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## (54) LIPID NANOPARTICLES FOR GENE EDITING SYSTEMS

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## (57) ABSTRACT

The present disclosure provides compositions and methods for treating and preventing localized nociception, inflammation, or morphological changes associated with joint disease or illness, back or spine conditions or disorders, and musculoskeletal diseases or dysfunction with an LNP-encapsulated CRISPR/Cas9 gene editing system.

Specification includes a Sequence Listing.

SEQ ID NO.	Chromosome	Protospacer (cRNA)	Cut site coordinate	Score
1	chr2	CCGAAGCCCCGGGUCAUCGGG	9555543	83
2	chr2	CUGACGCUGCAGUUUAAGG	9490227	82
3	chr2	UGGCCCGOAAGGGAUACACAG	9490422	82
4	chr2	UGGGGGCGAAUUAACAUAG	9535838	80
5	chr2	UCUCCUAUUCCUGACCAGCG	9555577	80
6	chr2	UUGGAGCUGCUGCGCGCAA	9490412	78
7	chr2	AGCGACUGCACGUUGAAAGGA	9505183	78
8	chr2	CAACAGCGACUGCACGUJUGA	9505187	78
9	chr2	AGUUUGUGGGAACUCGAGGG	9505265	78
10	chr2	AGCAGAAUUGUGCCCGAAUG	9510046	78
11	chr2	GUAGCGGGGCCGGAAACAUG	9555605	78
12	chr2	GCGAUUAUAGCUACUUGCAA	9502195	77
13	chr2	UAAAGUUUUGUGGGAACUGGA	9505268	77
14	chr2	GGACUUCUUCACUGGACACG	9536706	77
15	chr2	CCGGGACCUCUCCGGUAAGACC	9555537	77
16	chr2	AUGCAUCCCUUUCUGCGAGA	9497149	76
17	chr2	AAGUAGCAUUAUCGCCUCC	9502214	76
18	chr2	CCCUCCCUGGUCCUCAUUCG	9510047	76
19	chr2	UAGCUUAUUAUAGCCCAGUU	9517975	76
20	chr2	GGUCUUUACCGAGUCUCGG	9555517	76
21	chr2	CAGCACGAAAGGAACCCACGC	9555575	76
22	chr2	CACAGACUCACAUUAGGACG	9490334	75
23	chr2	GUGAUCGCCACUCACAGCUA	9510008	75
24	chr2	AACGUUCAGACUUCUAGUGC	9536808	75
25	chr2	CUGGGCCCGAAGGGAUACA	9490421	74
26	chr2	CGUCGAAAUUCUGAGCAGCA	9490499	74
27	chr2	CUGGGUCUUUACCGAGUCUC	9555514	74
28	chr2	CAGCGUGGUUCCUUUCUGUGC	9555562	74
29	chr2	GGUCCGGGCGCCAGCACGAA	9555564	74
30	chr2	CAACAUCGUUUGGGUCUGCC	9492953	73
31	chr2	GUCACAAAUCUACUGUAC	9494660	73
32	chr2	AAUGCAUCCCUUUCUGCGAG	9497150	73
33	chr2	GGGCACACUACUGCUAUUACC	9497247	73
34	chr2	AUAAGUULUGGGGAACUCG	9505269	73
35	chr2	GCGUUCUUGAAAACACUCU	9505308	73
36	chr2	GGAGCAGAACAGUAUCGGG	9510072	73
37	chr2	CCAAUUCAGAGUUGUAACC	9510118	73
38	chr2	CAUCUACGGAACACUUCAU	9523295	73
39	chr2	CAUCGUUCUACAGAUACAU	9526150	73
40	chr2	UCAUCGGCUUCUACAGAUACA	9526151	73
41	chr2	UUCUUACUGGACACGUGGU	9536702	73
42	chr2	GUCUUUACCGAGUCUGGU	9555518	73
43	chr2	CAA AUGUGAGAACAGAGUAC	9493806	72
44	chr2	UGCUGUUCCCUCUGCGAGAA	9497154	72
45	chr2	UGGGCUAGAACCCUAGAGUC	9535912	72
46	chr2	GCCCCCACCUCCGGAUAGACC	9555538	72
47	chr2	CGUGCUGGCGCCGGACCUUC	9555547	72
48	chr2	CGAAAGGAACCACCGCUGGU	9555580	72

SEQ ID NO:	Chromosome	Protospacer (cRNA)	Cut site coordinate	Score
1	chr2	CCGAAGCCGGGUCAUCCGG	9555543	83
2	chr2	CUGACGCUGCAGUUUAAGG	9490227	82
3	chr2	UGGCGCCGAAGGGAUACAG	9490422	82
4	chr2	UGGGGCCGAAUUAACAUAG	9535838	80
5	chr2	UCUCCUAUUCCUGACCAGCG	9555577	80
6	chr2	UUGGAGCUGCUGGCCGCAA	9490412	78
7	chr2	AGCGACUGCACGUUGAAGGA	9505183	78
8	chr2	CAACAGCGACUGCACGUUGA	9505187	78
9	chr2	AGUUUGUGGGAACUCGAGGG	9505265	78
10	chr2	AGCAGAAUGUGCCCCGAAUG	9510046	78
11	chr2	GUAGCGGGGCCGGAAACAUG	9555605	78
12	chr2	GCGAUUAAUUGCUACUUGCAA	9502195	77
13	chr2	UAAAGUUUUGUGGGAACUCGA	9505268	77
14	chr2	GGACUUUCUUCACUGGACACG	9536706	77
15	chr2	CCGCGACCUCCGGAUGACCC	9555537	77
16	chr2	AUGCAUCCCUUUCUGCGAGA	9497149	76
17	chr2	AAGUAGCAUAAAUCGCCUCC	9502214	76
18	chr2	CCCUCCCCUGGUCCUCAUUCG	9510047	76
19	chr2	UAGCUUAAAUAAGCCCAGUU	9517975	76
20	chr2	GGUCUUUACCGAGUCUCUGG	9555517	76
21	chr2	CAGCACGAAAGGAACCACGC	9555575	76
22	chr2	CACAGACUCACAUAAUGGACG	9490334	75
23	chr2	GUGAUCGCCACUCACAGCUA	9510008	75
24	chr2	AACGUUCAGUACUUGAUGUC	9536808	75
25	chr2	CUGGCGCCGAAGGGAUACACA	9490421	74
26	chr2	CGUCGAAAUGCUGAGCAGCA	9490499	74
27	chr2	CUGGGUCUUUACCGAGUCUC	9555514	74
28	chr2	CAGCGUGGUUCCUUUCGUGC	9555562	74
29	chr2	GGUCGCGGCCAGCACGAA	9555564	74
30	chr2	CAACAUCGUUGGGUCUGUCC	9492953	73
31	chr2	GUCACAAAUCCUACUGUAC	9494660	73
32	chr2	AAUGCAUCCUUUCUGCGAG	9497150	73
33	chr2	GGGCACUCACUGCUAUUACC	9497247	73
34	chr2	AUAAAGUUUGUGGGAACUCG	9505269	73
35	chr2	GCGUUCUUGAAAACACUCCU	9505308	73
36	chr2	GGAGCAGAACAUCAUCCGGA	9510072	73
37	chr2	CCAAUUCAGAGUUGUAACC	9510118	73
38	chr2	CAUCUAUCGGAACACUCAU	9523295	73
39	chr2	CAUCGUUCUACAGAUACAU	9526150	73
40	chr2	UCAUCGCUUCUACAGAUACA	9526151	73
41	chr2	UUCUUCACUGGACACGUGGU	9536702	73
42	chr2	GUCUUUACCGAGUCUGGU	9555518	73
43	chr2	CAAAUGUGAGAACGAGUAC	9493806	72
44	chr2	UGCUGUUCCCCUCUGCAGAA	9497154	72
45	chr2	UGGGCUAGAACCCUAGAGUC	9535912	72
46	chr2	GCCGCGACCUCCGGAUGACC	9555538	72
47	chr2	CGUGCUGGCGCCGCGACCUC	9555547	72
48	chr2	CGAAAGGAACCACGCUGGUC	9555580	72

FIG. 1

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
49	chr21	CUGCCGGUUGGCAUCGUACG	26840497	84
50	chr21	UACAUGUGCUGGCUGCAUCG	26840439	82
51	chr21	UGUGUCCAGUCACCGCUAUG	26842640	82
52	chr21	CGACGUCGGCGGCACGUGCG	26844362	82
53	chr21	GGAGACUGUUUCGAGAACGUG	26840005	81
54	chr21	UGGCCGUGUCGGACCGCACUC	26844807	81
55	chr21	AUGCAGCGAGCUGUGCCCAGA	26844937	81
56	chr21	GACUGUUCGAGAACGUGCGG	26840002	80
57	chr21	GCGCGUGGUCCCCGUGUCCCCG	26844747	80
58	chr21	GUACGCUGGCGUCUCACCAA	26839654	79
59	chr21	GUUCUCGAACAGUCUCCCCA	26840023	79
60	chr21	UCAAACCAAACACUUCCCCGU	26840361	79
61	chr21	GCACCUCGUACGAUGCCAAC	26840488	79
62	chr21	GCUUGUGGCAGACCAGUCGA	26842610	79
63	chr21	GGUUUCCACAUAGCGGUGAC	26842647	79
64	chr21	GCCGCUUCCGGAAACCGGACC	26844599	79
65	chr21	GCUUCACGCUCCGAGAACGUG	26844642	79
66	chr21	CGGCUUCACGCUCCGAGAACG	26844644	79
67	chr21	CUGGCCGUGUCGGACGCACU	26844808	79
68	chr21	CUUGCGCCUUCCGAACCCCU	26844933	79
69	chr21	GGUGAGACGCCAGCGUACUU	26839671	78
70	chr21	UGUUCGAGAACGUGCGGUGG	26839999	78
71	chr21	AACCAAACACUUCCCCGUGGG	26840358	78
72	chr21	GUGUACCGGCACCUCUGGUG	26840394	78
73	chr21	GGUGUACCGGCACCUCUGGU	26840395	78
74	chr21	GUCAUCCACGAUGAACAGAA	26842468	78
75	chr21	CCGGCCCCUUCUGUUCAUCG	26842475	78
76	chr21	UCCGCUUUCCCAGUCGGCCG	26844337	78
77	chr21	AAUCGGCAGGGCGACGUCGG	26844373	78
78	chr21	CGGAAUCGGCAGGGCGACGU	26844376	78
79	chr21	GGUGGCGAGGCGCUCGCUGG	26844462	78
80	chr21	GCGCCCGAGUGCGUCCGACA	26844816	78
81	chr21	AAUGCAGCGAGCUGUGCCCG	26844938	78
82	chr21	CCGCUGUUUCACUUUCGAUGU	26838232	77
83	chr21	GAGGAGUCCAGUACACGAUG	26839980	77
84	chr21	UGUAAACUGGCACUGGCCGU	26840485	77
85	chr21	CAUGUGGCAUGUUAAAACACG	26841159	77
86	chr21	UCGUAAUUCAGUUAGGCCUGG	26842505	77
87	chr21	GCUGGGGUGUUUGUACAAUC	26842542	77
88	chr21	GGGUCGUGGACGACGAGCCC	26844342	77
89	chr21	CCUGCCGAUUCGCCCGCAGG	26844398	77
90	chr21	AGCCUCUGCGAGGGCGUGCG	26844520	77
91	chr21	ACGUUCUGGAGCGUGAAGCC	26844658	77
92	chr21	ACUCGGCGCCCCUCCGAGG	26844791	77
93	chr21	GCAAGCUGGGCAGCGACAU	26844903	77
94	chr21	CCAACAUCGAAGUGAAACAG	26838220	76
95	chr21	GAAUGGAUUCCCAAGUACGC	26839668	76
96	chr21	AGGAGUCCAGUACACGAUGA	26839979	76

FIG. 2

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
97	chr21	GUAGACGUGCAGGAUCCGUG	26965830	85
98	chr21	GUUUCGCAGCUGGCGCGCGG	26965777	84
99	chr21	AGAAAAGGGCGCGUGUACG	26965847	82
100	chr21	CGGAGGCCACUGCUUCUAUCG	26965996	82
101	chr21	UCGUCUACGCGGGCGGCCGG	26966106	82
102	chr21	GUAAUUCCGUCUGCGUCCGG	26930013	81
103	chr21	GGACUGGUCCAAGAGCUGCG	26965683	81
104	chr21	CGAAACCCCCGCGGUCCACAC	26965748	81
105	chr21	CUGCACGUCUACACCGCGA	26965807	81
106	chr21	CCUGGAGCGAGAUGGUUCGG	26966072	81
107	chr21	GCCAUGAUUCAGGUGACCAGA	26930056	80
108	chr21	ACAACGGACGCUACUGCAC	26932915	80
109	chr21	UGGAUGUCUGUGCUCGCCUG	26934606	80
110	chr21	AUGGCGCGGUUGUAUGGCCG	26965540	80
111	chr21	UGGCUGACGCGUCCAUGGCG	26965554	80
112	chr21	AACCCCCGCGGUCCACACCGG	26965745	80
113	chr21	CUGGAGCGAGAUGGUUCGGU	26966071	80
114	chr21	AUCGACCAACUCUACUCCGG	26966146	80
115	chr21	CAUGGGCUACUCUGGCCACGA	26924369	79
116	chr21	CUAUAGCGGUUGGAGGCCACA	26924408	79
117	chr21	ACGUGGUGAGGAUUCUGAA	26924589	79
118	chr21	CAGUAAGGGUUACACUGACG	26924606	79
119	chr21	ACUUGGGAGCAGCGUACCAU	26932861	79
120	chr21	GCGGUGGAAGGGACGCCUUG	26934532	79
121	chr21	UGGUGUGCUGUGGUACGCCA	26934586	79
122	chr21	UGGCAUCGUAGGUCUGUCU	26934692	79
123	chr21	CCGGCCAUAACAACCGCGCCA	26965554	79
124	chr21	UCCGCUCUCUCGCCCGCUGG	26965651	79
125	chr21	CCGGCGCACAGCAACCCGAG	26965708	79
126	chr21	UGUACGGGGAUGGGGUCCGCA	26965833	79
127	chr21	AGUAUUUCCGUCUGCGUCCG	26930014	78
128	chr21	GUACUCCGUGUGUCCCGGCA	26934626	78
129	chr21	UCUGGUGGCUGACGCGUCCA	26965559	78
130	chr21	CCAGUGCUGCGCGUCCGCU	26965706	78
131	chr21	GUGCGCCGGAGCAUGCUCGU	26965734	78
132	chr21	AUGCUCGUGGGCCUCCGGUG	26965746	78
133	chr21	GAAGAAAAGGGCGCGUGUA	26965849	78
134	chr21	ACCCUAAAGCCACUGCUGCG	26965885	78
135	chr21	GGCUACCUCUGCUACGCGGG	26966113	78
136	chr21	GGUGGGCUACCUCUGCUACG	26966117	78
137	chr21	CUACUCCGGCGGCGGCAAGG	26966135	78
138	chr21	ACUAUAGCGGUUGGAGGCCAC	26924409	77
139	chr21	CUUGGGAGCAGCGUACCAUU	26932862	77
140	chr21	GGGAGCAGCGUACCAUUGGG	26932865	77
141	chr21	AACAAACGGACGCUACUGCAC	26932916	77
142	chr21	ACGUCAAGCCAUGGCAACUG	26933027	77
143	chr21	CUAUUCAGACGUCAAGCCA	26933036	77
144	chr21	AGUACUCAGGCCGAUGUC	26934656	77

FIG. 3

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
145	chr11	CGACGCGACAUCCAACCGAG	10305767	84
146	chr11	CGCUCGGUUGGAUGUCGCGU	10305780	82
147	chr11	UCACGUCAGCGAGCCGGUG	10306011	82
148	chr11	GCGCUUGACUCGGAUGCGGG	10306348	82
149	chr11	CUGCCGCUUCGGGACGUGCA	10306427	82
150	chr11	CUUCACGUACAGCGAGCCGG	10306013	80
151	chr11	GUAGCGCUUGACUCGGAUGC	10306351	80
152	chr11	GCACCGUGCACGUCCCCGAAG	10306418	80
153	chr11	CUAGGAAGGCAGCGAACCC	10305735	79
154	chr11	ACAAGGACAACGUUCGCC	10306492	79
155	chr11	UGGGGAGCACUUCCACUCGG	10306614	79
156	chr11	AGCGGUGUCAGCGCCUAGGA	10305749	78
157	chr11	AUUCGGCCCCAGGACAUAGAA	10306069	78
158	chr11	CCUGUCUUCGGGGCUUCGAG	10306082	78
159	chr11	GUGGGGAGCACUUCCACUCG	10306615	78
160	chr11	GAGCGAACCCAGGUACAUCA	10305725	77
161	chr11	UCCUAGGCGCUGACACCGCU	10305764	77
162	chr11	AGGCGCUGACACCGCUCGGU	10305768	77
163	chr11	CACCGGGCUCGCUGACGUGA	10306026	77
164	chr11	GGUAGCGCUUGACUCGGAUG	10306352	77
165	chr11	AGGGCGUAGUUACCUGUCUU	10306094	76
166	chr11	CGCCGGCGGCCGUAGCCCUG	10306515	76
167	chr11	UCCGUCGCCUGAUGUACCU	10305730	75
168	chr11	UUCACGUACAGCGAGCCGGU	10306012	75
169	chr11	GGCGUAGUUACCUGUCUUCG	10306092	75
170	chr11	CAUCCGGACUGCUGCGGGCG	10306325	75
171	chr11	UCUGGGGGUAGCGCUUGACU	10306358	75
172	chr11	ACCGAGCGGUGUCAGCGCCU	10305753	74
173	chr11	CCUGGGGCCGAAUAAGGGUC	10306047	74
174	chr11	CCAGACCCUUAAUUCGGCCCC	10306059	74
175	chr11	UAUUCGGCCCCAGGACAUAGA	10306068	74
176	chr11	GCGCCGGCGGCCGUAGCCCU	10306516	74
177	chr11	UUCUAAGCCACAAGCACACG	10306595	74
178	chr11	GGGAGCACUUCCACUCGGGG	10306612	74
179	chr11	ACCCAGGUACAUCAUCAGGGCGA	10305719	73
180	chr11	AUAAGUGGGCUCUGAGUCGU	10305971	73
181	chr11	CUGGGGCCGAAUAAGGGUCU	10306046	73
182	chr11	AUCCGGACUGCUGCGGGCGG	10306324	73
183	chr11	ACUCGGAUGCGGGCGGCAUC	10306341	73
184	chr11	AGCGGCAGCCAAGGUCCGG	10306400	73
185	chr11	GGCCUUCACGUACAGCGAGCC	10306016	72
186	chr11	UCAUGUCCUGGGGCCGAAUA	10306053	72
187	chr11	CACCAAGAGUCCGACCCGGGC	10306558	72
188	chr11	AGUGGGAGCACUUCCACUC	10306616	72
189	chr11	UUCCGUCGCCUGAUGUACC	10305729	71
190	chr11	CAGAGCCCACUUAAUUCACC	10305950	71
191	chr11	AAUAAGUGGGCUCUGAGUCG	10305970	71
192	chr11	CAUGUCCUGGGGCCGAAUA	10306052	71

**FIG. 4**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
193	chr1	GCUGUGCUGUGGUUCCGUGA	116392921	81
194	chr1	CAGGAACGGCCGCCCGUGGA	116395233	81
195	chr1	GCGGCCGUUCCUGAUGCCGU	116395255	81
196	chr1	GCCAAGGCCUGCGUAGUACA	116397909	81
197	chr1	UACCUAGGACUCGUUCUCCG	116393709	80
198	chr1	GCAUCAGGAACGGCCGCCCG	116395237	80
199	chr1	GUAGACAACAAUACCACGUG	116389447	79
200	chr1	UAUCAUCGUAGCCA AUGUGC	116389679	79
201	chr1	AUCGUAGCCA AUGUGC CGGA	116389683	79
202	chr1	GGUCAUCCCAGUCCACUCGG	116400948	79
203	chr1	UGACCGCUGGAUCAACGAUG	116400978	79
204	chr1	AUCAUCAGGC GACGCC CUGG	116403971	79
205	chr1	UCUUUUUAAGGCACCGCACG	116389446	78
206	chr1	AGCACUUCGACAUUUGCCCA	116395259	78
207	chr1	CGUUGGGACCAUCUCGCGCC	116387322	77
208	chr1	UUCUGAUUCACCAGUGAGCG	116388917	77
209	chr1	GUGGAUAACUCCUCGCUCAC	116388919	77
210	chr1	CAGCAAACCUUCCGGCACAU	116389678	77
211	chr1	CGCCCGUGGAGGGUCAUCA	116395223	77
212	chr1	UCAGGAACGGCCGCCGUGG	116395234	77
213	chr1	CAUGAUUGACCCUCCACGGG	116395236	77
214	chr1	CAAACAUUCCACUACCA CUG	116399040	77
215	chr1	GAAUGAGCGGCUGAUCAGCA	116399523	77
216	chr1	GCUGAGAUCCUGGCGCGAGA	116387326	76
217	chr1	GUGGUAUUGUUGUCUACACU	116389465	76
218	chr1	GACACUUACUGCCAAACGCA	116390238	76
219	chr1	UGUUGGGCUCCGAUGUGUU	116393554	76
220	chr1	CACCACGAUACUGACGAAGA	116401178	76
221	chr1	AGAGGAUCAACUUACUUGAG	116401653	76
222	chr1	AAUGCCCAUAAA UACCGCCA	116403973	76
223	chr1	CUCAUCAUCAGGC GACGCC	116403968	76
224	chr1	GACAAAGCGUCUGGUACCGUG	116388244	75
225	chr1	UAUCCACAUCAUCACGGGUG	116389574	75
226	chr1	UGAUGAAGGGCGCCCCAGAA	116393597	75
227	chr1	UCUGGGGUAGGGUGCUACG	116398628	75
228	chr1	UCACCUGUUGGGCCUCCGAG	116400948	75
229	chr1	ACGAUGUGGAAGACAGCUAC	116400992	75
230	chr1	CAGAGGAUCAACUUACUUGA	116401654	75
231	chr1	CUCCAAAUAGGCGUUCUGAA	116393678	74
232	chr1	AUCACUGCCGUGUACUACGC	116397904	74
233	chr1	CUCUACUCCAGUCACAAUUG	116398768	74
234	chr1	GAGUGGACUGGGAUAGACCGC	116400965	74
235	chr1	CCAGAGGAUCAACUUACUUG	116401655	74
236	chr1	GAAUGCCCAUAAA UACCGCC	116403974	74
237	chr1	CUCACCAUUUCGAAUCACAA	116388647	73
238	chr1	AGGCAAUGUUCCUCGUCUCC	116388978	73
239	chr1	CGUGGUAUUGUUGUCUACAC	116389464	73
240	chr1	CCUCAAGCCAGGUGUACUCA	116389625	73

FIG. 5

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
241	chr11	GAACUACCCAGUCGUACGUG	27657922	82
242	chr11	ACACGCUCAGCUCCCCUCGG	27658161	80
243	chr11	AACUACCCAGUCGUACGUGC	27657921	79
244	chr11	UAUUAGUGAGUGGGUAACGG	27658119	79
245	chr11	UCAUGCCAUACAGAACGCGUG	27674165	79
246	chr11	GGUAGACGCCAAAACAUGUG	27674187	79
247	chr11	AAGGGCCCGCACGUACGACU	27657928	78
248	chr11	ACGACUGGGUAGUUCGGCAC	27657942	78
249	chr11	CGACUGGGUAGUUCGGCACU	27657943	78
250	chr11	CUCUGACCCUGCCCCGCGAG	27658161	78
251	chr11	AGAGUGGCGCCGGACCCUCA	27658192	78
252	chr11	UCCCAUGGGUCCGCACACCU	27658458	78
253	chr11	UCUACCCACACGCUUUCUGUA	27674158	78
254	chr11	GUACGUGCGGGCCUUACCA	27657909	76
255	chr11	AGUGGACAUGUCGGCGGGGA	27658080	76
256	chr11	UUGACUACUGAGCAUCACCC	27658285	76
257	chr11	UGUGUACCUCCACGUUUCAG	27674103	76
258	chr11	CGAGACCAAGUGCAAUCCCA	27658002	75
259	chr11	ACUGCAGUGGACAUGUCGGG	27658085	75
260	chr11	ACUCUGACCCUGCCCGCGA	27658162	75
261	chr11	ACGCAGACUUGUACACGUCC	27658291	75
262	chr11	GGGACUCUGGGAGAGCGUGAA	27658427	75
263	chr11	ACCCAGGUGUGCGGACCCAU	27658447	75
264	chr11	UGUUAAAUCUCCACUGAAACG	27674106	75
265	chr11	CAUGCCAUCACAGAACGUGU	27674166	75
266	chr11	UAAGGGCCCGCACGUACGAC	27657927	74
267	chr11	GAAUUGGCUGGCGAUUCAUA	27657874	73
268	chr11	CUGCAGUGGACAUGUCGGGC	27658084	73
269	chr11	AUGGGUCCGCACACCUGGGU	27658462	73
270	chr11	GCACGUACGACUGGGUAGUU	27657936	72
271	chr11	GUCCCAUGGGUCCGCACACC	27658457	72
272	chr11	GGAUUAACCUUGUGUGCACUCA	27674256	72
273	chr11	UACCCAGGUGUGCGGGACCCA	27658448	71
274	chr11	GUAAACCAUGGGAUJUGCACU	27658009	70
275	chr11	GGACUCUGGGAGAGCGUGAAU	27658426	70
276	chr11	GAAGCAAACAUCGAGGGACA	27658481	70
277	chr11	ACACGUGAUAGAAGAGCUGU	27658356	69
278	chr11	CACGCUCUCCAGAGUCCAU	27658444	69
279	chr11	GCUCAGUAGUCAAGUGGCCUU	27658263	68
280	chr11	UGUGCGGACCCAUGGGACUC	27658440	68
281	chr11	CUUCAUUGGGCCGAACUUUC	27658338	66

**FIG. 6**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
282	chr11	UACCGCACCGCUUAGAUCUG	14968983	80
283	chr11	CCAACCCCAAUUGCAGUUUG	14968901	77
284	chr11	CAUGCUGGGCACAUACACGC	14968935	77
285	chr11	UUCUGGGGCAUGCACAAUAG	14968826	76
286	chr11	UUACCGCACCGCUUAGAUCU	14968982	76
287	chr11	AAUCUGAGUACUUGCAUGCU	14968949	75
288	chr11	AGCCCCAGAACUAAGCGGGUG	14968973	74
289	chr11	CGCUUCGUCCUCACUGAGCG	14970038	74
290	chr11	GCAUGCUAACAAUGAGGGCGA	14968833	73
291	chr11	AAGGGAUAAUGUCCAGCGACU	14968851	73
292	chr11	AUGGUCUCUCUCCAAGUCGC	14968852	73
293	chr11	GUUCCCCCAAACUGCAAUUG	14968893	73
294	chr11	UAAUCUGAGUACUUGCAUGC	14968950	73
295	chr11	UCGUCCUCACUGAGCGUGGC	14970042	73
296	chr11	GGACGAAGCGCGCCUCCUGC	14970013	71
297	chr11	CGUCCUCACUGAGCGUGGCC	14970043	71
298	chr11	AUUACCGCACCGCUUAGAUC	14968981	70
299	chr11	CUCCAACCCCAAUUGCAGUU	14968899	69
300	chr11	CGUUCCCCCAAACUGCAAUU	14968894	66
301	chr11	CCCCAAACUGCAAUUGGGGU	14968889	65

**FIG. 7**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
302	chr11	CGCGUCCUCUUUACUGAGUG	15075095	78
303	chr11	UAUCUUGGUCCUGUACCAGG	15074774	77
304	chr11	ACCUGAAUGGCGCCGCCUGG	15074789	77
305	chr11	UAUUGGCUGGACCUACCUGA	14978358	76
306	chr11	CCCGGCCACACUCAGUAAAG	15075102	76
307	chr11	CCUUUACAGAGAGGCGGCAU	15074718	74
308	chr11	CAGCCGAUGAGUCACACAGG	15077323	73
309	chr11	AAAGAGGCCUAACCUUCAGGU	14978359	72
310	chr11	CAGUAUCUUGGUCCUGUACC	15074771	72
311	chr11	AUCUUGGUCCUGUACCAGGC	15074775	72
312	chr11	UGCCAGCCGAUGAGUCACAC	15077326	70
313	chr11	AGGCUUGCUGAGCAGAUCAG	15077360	70
314	chr11	AUCCUUCCUUUACAGAGAGG	15074712	69
315	chr11	AGAGGGACGCGCGCCUCCUGC	15075120	69
316	chr11	GCUUUGGAACCCACAUUGGU	15077394	69
317	chr11	CAACUUCGUGCCCACCAAUG	15077396	69
318	chr11	GUCCCUGCGGCGCCUGCCAA	15077416	69

**FIG. 8**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
319	chr2	GUGGGUGUUAACAUUACACU	187380496	80
320	chr2	UGUACAGAUUGGAUUCCGCU	187352265	77
321	chr2	CCUACACACACUCAUUGUGG	187360654	76
322	chr2	AUCCGUGGUCCAUUUAUGGUC	187379006	76
323	chr2	GACAUACCUUCACUUUCUCG	187380477	76
324	chr2	CUGAUUCCAUGGCGACCUGA	187352174	75
325	chr2	GCAAUGGCAUGUAUACAAGC	187359253	75
326	chr2	CGUUUACUGCAACAGAACCU	187380769	75
327	chr2	CCUGAGGACUCAAUUCAGUU	187383258	75
328	chr2	UUAGGAGCCUAAGUUGCCAA	187363489	74
329	chr2	UACCUUGACCAUAAAUGGACA	187378996	74
330	chr2	CGUGGGUGUUAACAUUACAC	187380495	73
331	chr2	UGUGAGAGCUACUCUUUAUCU	187352223	71
332	chr2	GCACAUCCUUUAUGCACUCC	187352118	70
333	chr2	UGCUGGAACGAUGUUGCAGC	187380735	70
334	chr2	GCGUUUACUGCAACAGAACCC	187380770	70
335	chr2	CACUGCAGUGGCCAACAAACC	187363401	69
336	chr2	GGCUACUAAGGCCUGGUUGU	187363402	69
337	chr2	GCAUCACUGCUUAUCUCGCU	187378963	68
338	chr2	AGACAAUCCGUGUCCAUAUA	187379001	68
339	chr2	GCCAUACCUGGAAGUGCAUA	187352124	67
340	chr2	GGACAGUCAUGACUAUAACC	187346277	66

**FIG. 9**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
341	chr17	CAGCCACCUUCAUCCCCAA	34255407	74
342	chr17	GUUAUAACAGCAGGUGACUG	34256239	74
343	chr17	AGCGAGCCCJUGGGGAAUGA	34255401	73
344	chr17	UUGCUCUGGUCCAGGUGGUCCA	34256789	73
345	chr17	UAUGAGCAGCAGGCACAGAA	34255371	72
346	chr17	GGCGGCAGAGACUUUCAUGC	34255350	71
347	chr17	UACCUGGCUGAGCGAGGCCU	34255411	71
348	chr17	GCAGCCACCUUCAUCCCCA	34255406	71
349	chr17	AUAGGAAGAACUCAGUGCAG	34256281	71
350	chr17	GAGCAGCAGGCACAGAAGGG	34255368	70
351	chr17	CCUGGGCUGAGCGAGCCUUG	34255409	70
352	chr17	ACCUGGGCUGAGCGAGCCUU	34255410	70
353	chr17	AAGUUUAACAGCAGGUGAC	34256241	70
354	chr17	GAGCCUUGGGAAUGAAGG	34255398	69
355	chr17	AGUUUAACAGCAGGUGACU	34256240	69
356	chr17	GUGUUCAAGUCUUCGGAGUU	34256814	69
357	chr17	AUUGGUGAAGUUUAACAGC	34256248	68

**FIG. 10**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
358	chr17	AUUCUGUGGAAUCUGCCGGG	36089253	69
359	chr17	CCUGGAGCUGAGUGGCCUGAG	36088673	67
360	chr17	UCUGGACCCACUCCUCACUG	36088721	66
361	chr17	ACUGGCACUUACAUGACACC	36089188	65
362	chr17	ACUCACGUGAUGCAGAGAAC	36089997	65
363	chr17	GGUCUGUGCUGACCCCCAGUG	36088721	64
364	chr17	CCUAACCAAGCGAAGCCGGC	36088742	64
365	chr17	GACUGGCACUUACAUGACAC	36089187	64
366	chr17	GUGCUGACCCCAGUGAGGGAG	36088716	63
367	chr17	GCUUCCUAACCAAGCGAAGC	36088746	63
368	chr17	ACAGACCUGCCGGCUUCGCU	36088749	62
369	chr17	CCUGCCGGCUUCGCUUGGUU	36088754	62
370	chr17	AGAGAGCCAUGGUGCAGAGG	36090024	62
371	chr17	GGUGCAGAGGAGGACAGCAA	36090034	62
372	chr17	UCACCUGCUCAGAAUCAUGC	36090057	62
373	chr17	UAGUCAGCUAUGAAAUCUG	36089239	61
374	chr17	GAAAUUCUGUGGAAUCUGCC	36089250	61

**FIG. 11**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
375	chr17	CUGAGACUCACACGACUGCU	35878534	71
376	chr17	AGGUACCAUGAAGGUCUCCG	35880295	71
377	chr17	UAUGACUCCC GG CUGAACAA	35875570	69
378	chr17	GUAUGACUCCC GG CUGAACAA	35875571	69
379	chr17	GACUCACACGACUGCUGGGU	35878538	69
380	chr17	UACUCCUUGAUGUGGGCACCG	35878584	69
381	chr17	UAGCAAUGAGGAUGACAGCG	35880280	69
382	chr17	CUGGGUUGGCACACACUUGG	35872441	68
383	chr17	UCUUUGGCAUCCUUGACCUG	35875637	68
384	chr17	CUCUCCCACAGGUACCAUGA	35880304	68
385	chr17	UACCCCACCCGCUCCUUGAA	35872271	67
386	chr17	GGUAGAAUCUGGGCCUUCACU	35872270	67
387	chr17	UCUCUGGGUUGGCACACACU	35872438	67
388	chr17	AUACUCCUUGAUGUGGGCAC	35878583	67
389	chr17	GGUGUGGUGUCCGAGGAAUC	35878638	67
390	chr17	UCAAGACCAGGACUUACAUG	35880230	67
391	chr17	AGACCAGGACUUACAUGGGG	35880233	67

**FIG. 12**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
392	chr17	ACAUACAUUACAGCUUCCCCG	34271252	79
393	chr17	GUCCUGGACCCACUUUCUGUG	34271740	75
394	chr17	CCGGGGACAGUGGCUACUGG	34271235	74
395	chr17	CCUUGGACCCACUUUCUGUGUG	34271738	72
396	chr17	UCCUGGACCCACUUUCUGUGU	34271739	72
397	chr17	CUGGCUGAGCAAGCCCCUGG	34270349	71
398	chr17	UUACAGCUUCCCAGGGGACAG	34271245	71
399	chr17	ACAGAUCUCCUUGUCCAGUU	34271710	71
400	chr17	CACCAGUAGGCCACUGUCCCC	34271248	70
401	chr17	CAGCUGCUUUCAGCCCCCAG	34270348	69
402	chr17	GUCCAUUUGAGGGUGUGUCCU	34271103	69
403	chr17	AGUCCAUUGAGGGUGUGUCC	34271104	69
404	chr17	ACAAGAAGAAAAGUCCAUUG	34271115	69
405	chr17	GACCCAGGACACACCCUCAA	34271113	69
406	chr17	UUCCCCGGGGACAGUGGCUAC	34271238	69
407	chr17	CCACCAGUAGGCCACUGUCCC	34271247	69
408	chr17	CCAGGUGCUUCAUAAAGUCC	34271756	69

**FIG. 13**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
409	chr2	CUUCUGAUUCGCCGCAGAGG	227813970	79
410	chr2	GUGCUGCUACUCCACCUCUG	227813971	78
411	chr2	ACACGGCAGCUGGCCAAUGA	227815542	77
412	chr2	AUGUCAGUGUAACAUACCUG	227816384	77
413	chr2	UACCUUCUGAUUCGCCGCAG	227813973	76
414	chr2	CAAACUCUUGGUACAGCACA	227813915	72
415	chr2	GCGCAAUCCAAAACAGACU	227816353	72
416	chr2	AAUUUAUUGUGGGCUUCACA	227815525	71
417	chr2	CGCAAAUCCAAAACAGACUU	227816354	70
418	chr2	CUGUACCAAGAGUUUGCUCC	227813934	69
419	chr2	AGCAGCCAGGAGCAAACUCU	227813927	68
420	chr2	GCACUGACAUCAAAGCAGCC	227813940	68
421	chr2	CACCUCUGCGCGAACAGA	227813983	68
422	chr2	UGUGGGCUUCACACGGCAGC	227815532	68
423	chr2	AUUGAUGUCACAGCCUUCAU	227815543	68
424	chr2	AGCAACUUUGACUGCUGUCU	227815476	65
425	chr2	GUGAAGCCCACAAUAAAUUU	227815509	64

**FIG. 14**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
426	chr6	CGAACGUCCAUGCUGCACAG	131950425	84
427	chr6	AAGACUCGACUCACCGCGA	131950295	83
428	chr6	CUUGAACUCCACCGGCAGGG	131949413	81
429	chr6	ACCGUACCACCGAAGAUGCA	131950524	81
430	chr6	GACCCAUGCGACCCGCACAA	131950828	81
431	chr6	GAAGACUCGACUCACCGCG	131950294	80
432	chr6	GGUGGUACGGUGUACCGCAG	131950500	80
433	chr6	UCUGGAAGGACUCUCCGCGUG	131950498	80
434	chr6	CGGGUCGCAUGGGUCGCGCU	131950848	79
435	chr6	GUUCAAGUGCCCUGACGGCG	131949385	78
436	chr6	GGGGGUGCAGCAUCCGCCGU	131949446	78
437	chr6	CGAGCCCAGGACCAAACCG	131950315	78
438	chr6	CUGCAGUUUCUGGCCGACGGC	131950987	78
439	chr6	GC GGACGGGGCCC AUACUGG	131951160	78
440	chr6	UAUGAUUAGAGCCAACUGCC	131950106	77
441	chr6	UGCUCUCGCAGCUUACCGAC	131950154	77
442	chr6	UCCGGGACAGUUGUAAUGGC	131949338	76
443	chr6	GUUGUCAUUGGUAAACCGGG	131950044	76
444	chr6	GGCAGGCCAACACCGGUU	131950315	76
445	chr6	ACGAACGUCCAUGCUGCACA	131950424	76
446	chr6	CCCGCUGCAGUUCUGGCCGA	131950983	76
447	chr6	UCAGGGCACUUGAACUCCAC	131949405	75
448	chr6	UUCUGUGGAGUAGUACCGA	131949450	75
449	chr6	AGAGCCGCCUGUGCAUGGUC	131949987	75
450	chr6	GGCGUUGUCAUUGGUAAACCC	131950041	75
451	chr6	GGCCAAACGUGCUUCCAGU	131950151	75
452	chr6	AGCUUGACCCUCCUCGGAA	131950383	75
453	chr6	AGCACGAGGCUCACGCCGC	131950924	75
454	chr6	GACGGGGCCCAUACUGGCCG	131951163	75
455	chr6	AGCUCGGUAUGUCUUCAUGC	131949488	74
456	chr6	CGCAAGGCCUGACCAUGCAC	131949992	74
457	chr6	UACCGACUGGAAGACACGUU	131950141	74
458	chr6	GCUUGACCCUCCUCGGGAAG	131950384	74
459	chr6	GACGAACGUCCAUGCUGCAC	131950423	74
460	chr6	CCAGUGCACGUGCCUGGACG	131950450	74
461	chr6	CAAGUACCAGUGCACGCC	131950456	74
462	chr6	UCCUGCAUCUUCGGUGGU	131950513	74
463	chr6	UCCCCGGCCAACCGCAAGAU	131950786	74
464	chr6	CGGUGCACACGCCGAUCUUG	131950787	74
465	chr6	ACGCCGAUCUUGCGGUUGGC	131950795	74
466	chr6	AUUCUGUCACUUCGGCUCCC	131950802	74
467	chr6	ACCAUGACCGCCGCCAGUAU	131951158	74
468	chr6	AACCAUGACCGCCGCCAGUA	131951159	74
469	chr6	UGUCUCCGGGACAGUUGUAA	131949334	73
470	chr6	CCCAAGGACCAACCGUGGU	131950311	73
471	chr6	CAGCUUGACCCUCCUCGGGA	131950382	73
472	chr6	CGCAAGAUCGGCGUGUGCAC	131950774	73
473	chr6	CGACCCAUGCGACCCGCACA	131950829	73

FIG. 15

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
474	chr17	GGAGCGCCGGAUGUGCCGAC	40554804	80
475	chr17	ACGCAACUUUGAGCGCAACA	40555103	80
476	chr17	CAUCAGCAUUGACCACUACG	40555415	80
477	chr17	AGGCAGAAGAGUCGCCUAUG	40554744	79
478	chr17	CCGGCGCUCCUCCAUGAGUG	40554779	79
479	chr17	GCCGCGCUGUAGGCCAGAA	40555540	78
480	chr17	GAGGUACGGACGAUUAACAU	40555783	78
481	chr17	GCGCUCCUCCAUGAGUGUGG	40554776	77
482	chr17	UCCCUUCUGGGCCUACAGCG	40555529	77
483	chr17	GAGCAGGUAGGUACGGUCA	40555602	77
484	chr17	CCGUGACCUCAUUJUGACAC	40555808	77
485	chr17	GCGGAACUUGACGCCGAUGA	40554894	76
486	chr17	UAGGCCACGAAACAAAUGA	40555677	76
487	chr17	CAGGCAGAAGAGUCGCCUAU	40554743	75
488	chr17	GCCAGGACCACCCAUUGUA	40555042	75
489	chr17	CUACGACGUACCUACAGCC	40554941	74
490	chr17	GGUCUCGGCCUCCACACUCA	40554780	73
491	chr17	CCGAUGAAGGCGUACAAGAA	40554907	73
492	chr17	UGCACUCAAAGUUGCGUGCC	40555121	73
493	chr17	CUACAGCGGGCCAAGGUCCU	40555517	73
494	chr17	UGAUGGAGUACAUGAUAGGG	40555694	73
495	chr17	CCACACUCAUGGAGGAGCGC	40554791	72
496	chr17	CAUCAAGGUGAUCAUCGCGUG	40555079	72
497	chr17	UCGUGGGCCUACUGGGCAAU	40555653	72
498	chr17	ACGUGCGGAACUUUAAGCC	40555707	72
499	chr17	GUUGACGCAGCAGCGGACGC	40554930	71
500	chr17	CCAGGACUUGGCCCGCGCUGU	40555530	71
501	chr17	AAAGUUCCGCACGUCCUUCU	40555728	71
502	chr17	UGUUGAAGUUGGCCACCGUC	40555019	70
503	chr17	CCGCUACGUGGCCAUCGUCC	40555403	70
504	chr17	GAAACCAAUGAAAAGCGUGC	40558924	70
505	chr17	CAGGCUGUAGGUGACGUCGU	40554954	69
506	chr17	CCUACAGCGGGCCAAGUCC	40555518	69
507	chr17	GUACUCCAUCAUUUGUUUCG	40555670	69
508	chr17	CGAUGAAGGCGUACAAGAAA	40554908	68
509	chr17	UCUUCCAGCUGCCUACAAU	40555041	68
510	chr17	ACCAAUGAAAAGCGUGCGUG	40558921	68
511	chr17	ACCACCAAGCACGUUUUCAU	40558932	68
512	chr17	GUUGAAGUUGGCCACCGUCU	40555020	67
513	chr17	UCCUCUGGAGGUACUGUAC	40555292	67
514	chr17	UACUCCAUCAUUUGUUUCGU	40555669	67
515	chr17	CCUUUCUUGUACGCCUCAU	40554895	66
516	chr17	AUUUGUUUCGUGGGCCUACU	40555660	66
517	chr17	CACAGCGAUGAUCACCUUGA	40555092	65

FIG. 16

SEQ ID NO.	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
518	chr7	GAGUGAAGAGCGGCUCACGG	66152236	81
519	chr7	CCGGCGAGCACCUUGGCGCG	66114862	78
520	chr7	CCGCGCGCCAAGGUGUCGC	66114850	76
521	chr7	CAAGCUCCCCGGCAUGCAGCG	66114834	75
522	chr7	UCAGGGACUCUGGGUGCCUGCA	66134309	75
523	chr7	CCACAGCAGUCACAGGCCGG	66145472	75
524	chr7	GGGAUCGCCCGCGCUGCAUGC	66114829	74
525	chr7	GGUGGCCAGCUCCGCGCGCCA	66114860	74
526	chr7	GGCGAUCCCCGGCGAGCACCU	66114855	74
527	chr7	UGCGCUUUCUCAGUAACUACG	66127736	74
528	chr7	UGGCACCUUGGGCGCUGUUGG	66114888	72
529	chr7	UGGAUCUCCCACAGCAGUCAC	66145479	72
530	chr7	AGAGAGUGAAGAGCGGGCUA	66152233	72
531	chr7	CGCCGCCACCAACAGCGCCA	66114881	71
532	chr7	GGCGCGCGGAGCUGGCACCU	66114876	70
533	chr7	ACAUAUCAAAACACCAUGC	66134307	70
534	chr7	GCAGAAUGCUGGUGACGGUG	66152266	70

**FIG. 17**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
535	chr4	AGAGAGCAGCGCGGGCCAUG	73869472	76
536	chr4	ACCUGCUGCGCGCCGGCCAG	73869555	74
537	chr4	GAGCCACACCUAAGAAUGGG	73869922	74
538	chr4	CUAUGGGGAUGCAGGAUUG	73869941	74
539	chr4	GCACUGGCAGCGCAGUUCAG	73869686	73
540	chr4	GAGAGAGCAGCGCGGGCCAU	73869473	72
541	chr4	AGUGCCACUCGCAGGAGCCG	73869512	72
542	chr4	UUCUUCCCCUAGGAGCCGUCCG	73869675	72
543	chr4	ACUUUCGGUUUGGGCGCAGUG	73869772	72
544	chr4	CGGCCGGCCAGCGGCUACC	73869547	71
545	chr4	GAUGAUUUUCUUAACUAUGG	73869955	71
546	chr4	CUGGCCGGCGCGCAGCAGGU	73869570	70
547	chr4	CUUGGGGUGAAUUCCCUGCA	73869716	69
548	chr4	AGUGUGAACGUGAAGGUCCCC	73869760	69
549	chr4	GGGCGGCGGAGAGAGCAGCG	73869481	68
550	chr4	GGAGCAGCAGUGCCACUCGC	73869520	68
551	chr4	UGCUCCUGGUAGCCGCUGGC	73869555	68

**FIG. 18**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
552	chr4	UCUUAACCAUGGGCGAUGC	74098643	85
553	chr4	UCUCAACCCGCAUCGCCA	74098637	78
554	chr4	UUUCUUAACCAUGGGCGAUG	74098641	74
555	chr4	CAGCGCAGUUCAGUGGCCAG	74098912	74
556	chr4	UUCUUAACCAUGGGCGAUGC	74098642	73
557	chr4	AGAGCGUGGCGCGGGCCAUG	74099119	73
558	chr4	ACUUCGGUUUGGGCGCAGUG	74098819	72
559	chr4	GCACUGGCAGCGCAGUUCAG	74098905	72
560	chr4	UUCUUCCUAGGAGCGCCCC	74098916	72
561	chr4	CAAGAACAUCCAAAGUGUGA	74098844	70
562	chr4	CUUGAGGUGAAUUCUGCA	74098875	70
563	chr4	AGUGUGAAGGUGAAGUCCCC	74098831	69
564	chr4	ACCUGCUGCGCGCCGGCUGG	74099036	69
565	chr4	GGGACCCACCUGCUGCGCGC	74099029	68
566	chr4	GAGAGCGUGGCGCGGGCCAU	74099118	68
567	chr4	GGCGAGACUUACAUGACUU	74098803	67
568	chr4	CAGUGCUUGCAGACCCUGCA	74098876	67

**FIG. 19**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
569	chr4	UCUGAACCAUGGGGAUGCG	74038135	81
570	chr4	AGUGUGAAUGUAAGGUCCCC	74038322	78
571	chr4	CAGGGGGAGCACCAACUGAC	74037263	76
572	chr4	GAUGAUUUUCUGAACCAUGG	74038127	74
573	chr4	UUCUUCCCUAGGAGCGUCCG	74038407	74
574	chr4	ACUUUCGGUUUUGGGCGCAGUG	74038310	72
575	chr4	UCUCAACCCCGCAUCCCCCA	74038129	71
576	chr4	CGAUGAUUUUCUGAACCAUG	74038126	71
577	chr4	ACAUCCAAAGUGUGAAUGUA	74038330	71
578	chr4	GGGACCUUACAUUCACACUU	74038338	71
579	chr4	CAGUGCUGCGAGACACUGCA	74038367	71
580	chr4	GCAGCGCAGUUCAGUGACCA	74038402	71
581	chr4	AGAGCGUGGCGUGGGCCAUG	74038610	71
582	chr4	ACCUGCUGCGCGCCGGCUGG	74038527	69
583	chr4	UCGAUGAUUUUCUGAACCAU	74038125	68
584	chr4	GGGACCCACCUGCUGCGCGC	74038520	68
585	chr4	GAGAGCGUGGCGUGGGCCAU	74038609	68

**FIG. 20**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
586	chr4	UUUGGGAUGAACUCCUUGCG	73998282	79
587	chr4	ACAAGGAGCUCGAAGGACCG	73998544	79
588	chr4	AAUCUGCAAGUGUUCGCCAU	73998238	78
589	chr4	AGCGCGCACAAGGAGCUCGA	73998537	77
590	chr4	CAUAGGCCACAGUGCUCCA	73998221	76
591	chr4	CAGCCAGGGCCCAUCGCCAG	73998477	76
592	chr4	GUUJGUUUACAGACCACGCA	73998283	75
593	chr4	GCUCGAAGGACCGGGGACAC	73998551	75
594	chr4	AGGCCACAGUGCUCCAAGG	73998218	74
595	chr4	GCGCUCUCACCGCUGGCGAU	73998480	74
596	chr4	GCGCCAUGCGCUCUCACCGC	73998473	73
597	chr4	GCACAAGGAGCUCGAAGGAC	73998542	73
598	chr4	AGCUCGAAGGACCGGGGACA	73998550	73
599	chr4	AGCCUCCCUGAAGAACGGGA	73998078	71
600	chr4	CUUGGAGCACUGUGGGCCUA	73998234	71
601	chr4	CACAAGGAGCUCGAAGGACC	73998543	69
602	chr4	ACUUCCACCUUGGAGCACUG	73998226	68

**FIG. 21**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
603	chr4	UGGAGCACUGCGGGCCUGCG	73837066	79
604	chr4	AGGAGCUCUCACCGCUGGCG	73836854	77
605	chr4	GAGCUCUCACCGCUGGCGAG	73836852	76
606	chr4	CCGCCGGGGCCCCUCGCCAG	73836855	76
607	chr4	CGCAGGCCCGCAGUGCUCCA	73837081	76
608	chr4	ACACGGGCCGCGCGCUGGA	73836765	74
609	chr4	GCCCGAAGGACCCGGGACAC	73836781	74
610	chr4	AGCGCGCACAAGGAGCCGA	73836795	74
611	chr4	CUU GGAGCACUGCGGGCCUG	73837068	74
612	chr4	AGGCCCGCAGUGCUCCAAGG	73837084	74
613	chr4	ACAAGGAGCCCGAAGGACCC	73836788	73
614	chr4	GCAGUUUACCAAUCCUUUUG	73837036	73
615	chr4	AAACUGCAGGUGUUCCCCAC	73837064	73
616	chr4	GUGCCAGGAGCUCUCACCAC	73836859	72
617	chr4	CAAAACGAUUGGUAAACUGC	73837051	72
618	chr4	AGCCCGAAGGACCCGGGACA	73836782	71
619	chr4	CAGCCGCGCGGGCCCGUGUCC	73836782	70

**FIG. 22**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
620	chr4	AUUUCUGUGUUGGCGCAGUG	73741657	79
621	chr4	CUAAGUUCUUUAGCACUCCU	73741558	71
622	chr4	UGUAACAAUGACUUCCAAGC	73740669	70
623	chr4	CAACAGGUGCAGUUUUGCCA	73741553	70
624	chr4	CAAGAGAGCCACGGCCAGCU	73740671	69
625	chr4	GAACUGAGAGUGAUJUGAGAG	73741645	69
626	chr4	CAUGACUUCCAAGCUGGCCG	73740675	68
627	chr4	CAGAGCUGCAGAAAUCAGGA	73740698	68
628	chr4	UGAUGGAAGAGAGCUCUGUC	73741978	68
629	chr4	AGAGCUCUGUCUGGACCCC	73741987	68
630	chr4	AAAUUUGGGGUGGAAAGGUU	73741604	67
631	chr4	UCUGGACCCCAAGGAAAACU	73741996	67
632	chr4	UCUGCACCCAGUUUUCUUG	73741990	66
633	chr4	GUUCUUUGAUAAAUUUGGGG	73741614	65
634	chr4	GAAGGCUGCCAAGAGAGCCA	73740680	64
635	chr4	CACACAGAGCUGCAGAAAUC	73740702	64
636	chr4	AUUUCUGCAGCUCUGUGUGA	73740718	63

**FIG. 23**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
637	chr2	UUCCAGUCCAGUUUGCUAUG	218164649	76
638	chr2	ACUGACCCAGAAGCGUCACU	218164760	76
639	chr2	GCGCCGCAACAACAU CGGCC	218164361	75
640	chr2	GGGCCUUAAACAGUGUACGC	218164532	75
641	chr2	GCGAAA UUUUGGCCGAUGA	218164290	74
642	chr2	GUCUCAGUUUCUAGCAUACA	218165122	74
643	chr2	GACCUAUAGCAAACUGGAC	218164659	72
644	chr2	GAUGGUAGGCCUGGCGGAAA	218164691	72
645	chr2	AUCGGCCAAA UUUUCGCCA	218164273	71
646	chr2	UGUGAGCGCCGCAACAACAU	218164366	71
647	chr2	CGGAUCCUGCCUCACACCUU	218164588	71
648	chr2	CUUGACCAAGUGACGCCUUC	218164767	71
649	chr2	UGUCUCAGUUUCUAGCAUAC	218165121	71
650	chr2	AGCGCCGCAACAACAU CGGC	218164362	70
651	chr2	GGGCCCGGCCGAUGUUGUUG	218164370	70
652	chr2	ACUUGACCAAGUGACGCUUC	218164766	70
653	chr2	UCCAGUUUGCUAUGAGGUCC	218164643	69
654	chr2	CAACAACAU CGGCCGGGCC	218164355	68
655	chr2	CCAUGGAUUCUCAAGAUCC	218164256	65

**FIG. 24**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
656	chr2	GCUACUGGCCUGCAUCAGUG	218135223	78
657	chr2	UGGAUGAGUAGACGGGUCCUU	218135355	78
658	chr2	GAGCCCGGUGCGAUGUGAUUG	218135670	78
659	chr2	AAGGAUGCCCAGAAUCUCGG	218135699	77
660	chr2	UGUUAGCCCAGCCUGCUAUG	218135391	76
661	chr2	GAUGUGAUUGC GGCGCUCAC	218135660	76
662	chr2	GCGCCGCAAUCACAUCGACC	218135679	76
663	chr2	UAAGAUGACCAGCAUCACGA	218135006	75
664	chr2	ACUGACCCAGAACAGCGCUACU	218135280	75
665	chr2	CAGGCUCAGCAGGAAUACCA	218134973	74
666	chr2	AAGGAAGUCAACUUUCUAUAG	218135194	74
667	chr2	CAUGUAUAGCUAGAAUCUUG	218135781	74
668	chr2	UGUGGUCAUUAUCUAUGCCC	218134968	73
669	chr2	GUCCUUCGGAAAAGUAAGAC	218135341	73
670	chr2	GGGCCUUAAACAGCGUACGC	218135508	73
671	chr2	AUUGGCCAGAAGUUUCGCCA	218135767	73
672	chr2	UAUACAUGGCCUUGAUCAGCA	218135808	73

**FIG. 25**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
673	chr4	GAUGUGACGCCGCGGGCCGG	122826823	85
674	chr4	CGGACAGAAGAGCGGCCGAG	122826997	81
675	chr4	CGGCUGUACUGAAAAACGG	122827282	81
676	chr4	GCGGGUCGGCACUCACUGUG	122827351	81
677	chr4	CGGGUCGGCACUCACUGUGA	122827350	80
678	chr4	GAAAUCACCAAGUUGGUAG	122892299	80
679	chr4	AUGUGGCACUGAAACGAACU	122892316	80
680	chr4	CCGGGAUCCCGUUGCAACCG	122826858	79
681	chr4	UUCACGGAUGGGUGUCUCCG	122826919	79
682	chr4	AUUAGCGGACGCGGUGCCCG	122826853	78
683	chr4	GCGGCUGUACUGAAAAACG	122827281	78
684	chr4	AUGUAGAAGAUGUGACGCCG	122826815	77
685	chr4	GGGUGCCAGAUUAGCGGACG	122826844	77
686	chr4	CGGGAUCCCGUUGCAACCGC	122826857	77
687	chr4	CGCGGUUGCAACGGGAUCC	122826871	77
688	chr4	GUCUCCGCGGACGCCGCCUG	122826906	77
689	chr4	CCCCGUCAACUCGGCCGUCG	122827311	77
690	chr4	GACCCCGUCAACUCGGCCGU	122827313	77
691	chr4	CCCGGACGGCCGAGUUGACG	122827323	77
692	chr4	GCGGUGCCCGCGGUUGCAAC	122826863	76
693	chr4	AAGAGCGGCCGAGCGGCUCG	122827004	76
694	chr4	GCUGCCGCCAUCCUCGGGCA	122827211	76
695	chr4	UCCGCUAAUCUGGCACCCGC	122826827	75
696	chr4	CCGCGGUUGCAACGGGAUCC	122826870	75
697	chr4	GGGACCUGGGGUUCACGGAU	122826930	75
698	chr4	GACACCCAUCCGUGAACCCC	122826938	75
699	chr4	UUUUGCAGUACAGCCGCUUG	122827263	75
700	chr4	UUCCUGCGCAUCCACCCCGA	122827309	75
701	chr4	CGGCCGAGUUGACGGGGUCC	122827329	75
702	chr4	UAUGUGGCACUGAACGAAC	122892315	75
703	chr4	CCCGGCGGGUGCCAGAUUAG	122826838	74
704	chr4	CGCCGGCUCGCCGCGCACCA	122826969	74
705	chr4	GCCGGCUCGCCGCGCACAG	122826970	74
706	chr4	UCGGCCGCUUUUCUGUCCGC	122826981	74
707	chr4	CGUCAACUCGGCCGUCGGG	122827308	74
708	chr4	ACGGCCGAGUUGACGGGGUC	122827328	74
709	chr4	CGAACUGGGCAGUAUAACU	122892330	74
710	chr4	AUCUGGCACCCGCCGGGCG	122826820	73
711	chr4	ACCCCGUCAACUCGGCCGUC	122827312	73
712	chr4	UCUCCCGGACCCGUCAACU	122827320	73
713	chr4	CACCCCGACGGCCGAGUUGA	122827321	73
714	chr4	ACCCCGACGGCCGAGUUGAC	122827322	73
715	chr4	GAAGAUGUGACGCCGCCGCC	122826820	72
716	chr4	CCGCUAAUCUGGCACCCGCC	122826826	72
717	chr4	CGGGUGCCCGCGGUUGCAA	122826862	72
718	chr4	CUCUCCCCAGGCGGCGUCCG	122826914	72
719	chr4	CGGCCGAGCGGCCUCCGAGGC	122827009	72
720	chr4	GCCGCUUGGGGUCCUUGAAG	122827251	72

FIG. 26

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
721	chr8	UGUGGGUGAAUGUCCCCGUGCG	38414822	84
722	chr8	UGAGGAAUGAUCCCCAUUCGG	38424414	82
723	chr8	CCAGUGGCUAAAGCACACUCG	38424567	82
724	chr8	CAACCGCACCCGCAUCACAG	38429795	82
725	chr8	GCAUGGUUGACCGUUCUGGA	38421801	81
726	chr8	GUGAAGCUAACCGACUGCG	38423058	81
727	chr8	GAACUCCCACGUUGCUACCCA	38424637	81
728	chr8	AGGAAACCAUCCAUGGUGGA	38412452	80
729	chr8	GAGCUCUACGUGCUCCUCAG	38413728	80
730	chr8	GGAAGACCUGGACCGCAUCG	38413940	80
731	chr8	UUCCUCCAAGGAAACCGGUG	38412422	79
732	chr8	GGCCCAGCAGCCAUCGACCA	38412450	79
733	chr8	GCGCCUCUACUGCAUGGAUG	38412500	79
734	chr8	AUACUCCACGAUGACAUACA	38416052	79
735	chr8	ACCCAACCGUGUGACCAAAG	38417893	79
736	chr8	CGCAGGGGAGUAUCGUGCU	38421856	79
737	chr8	GAAGGGAGACCACUGUCUCGG	38424453	79
738	chr8	GUUGCCCGCCAACAAAACAG	38424648	79
739	chr8	ACCGGCCCAUCCUGCAAGCA	38424670	79
740	chr8	UUUUCAACCAGCGCAGUGUG	38427982	79
741	chr8	CAGCAGCCAUCGACCAUAGGA	38412446	78
742	chr8	AUCCAUGCAGUAGAGGCGCU	38412484	78
743	chr8	UGCCCAGCGCCUCUACUGCA	38412494	78
744	chr8	AUCGUGGAGUAUGCCUCAA	38416029	78
745	chr8	GAGCAUCCCUCUGCGCAGAC	38419543	78
746	chr8	AUACUCCCCUGCGUCCUCAA	38421878	78
747	chr8	CAACGUGGAGUUCAUGUGUA	38424615	78
748	chr8	GGUGUAGUUGCCUUGUCAG	38426197	78
749	chr8	AUUUUCAACCAGCGCAGUG	38427980	78
750	chr8	AAGCAUCUACCGAAAUCCC	38440354	78
751	chr8	UCCCCGACCUUGCCUGAACAA	38457360	78
752	chr8	CGGCCUAGCGGUGCGAGAGUG	38457395	78
753	chr8	GUUUCCUCCAAGGAAACCGG	38412424	77
754	chr8	AGCGCCUCUACUGCAUGGAU	38412499	77
755	chr8	CAUUUGCAGGUGGUUUUCG	38414281	77
756	chr8	UAGCAGACUUUGGCCUCGCA	38414823	77
757	chr8	UACUCCACGAUGACAUACAA	38416053	77
758	chr8	UCAGAGACCCUGCUAGCAU	38418290	77
759	chr8	GAUGACCGACCCCACCAUGC	38419649	77
760	chr8	AGUGUCUCACGCAUACGGUU	38428353	77
761	chr8	CAGCCACACUCUGCACCACGU	38457386	77
762	chr8	GAAGACGGAAUCCUCCCCUG	38413726	76
763	chr8	UCCUCGGGCUUACAGCUCGU	38414156	76
764	chr8	GAUCCGGUCAAAUAUAGCCU	38414596	76
765	chr8	AGGUCCCUUGUAUGUCAUCG	38416045	76
766	chr8	AAGUAUUAUUACCGUGUCCG	38418233	76
767	chr8	GUGGGACUCCCAUGCUAGCA	38418286	76
768	chr8	UUGGUUUGUUCCAGCAUUCG	38423172	76

FIG. 27

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
769	chr2	AUGGUGGUAGUAGCAACCAA	112781695	81
770	chr2	UAUGUAAUGCAGCAGCCGUG	112779528	80
771	chr2	GGUAGUAGCAACCAACGGGA	112781690	78
772	chr2	GGCCAUCGCCAAUGACUCAG	112781612	77
773	chr2	UGGUGGUAGUAGCAACCAAAC	112781694	77
774	chr2	CUUCUUCAGAACCUUCCGU	112781691	75
775	chr2	AGCUAUGGCCACUCCAUGA	112781791	75
776	chr2	AGCCGUGAGGUACUGAUCAU	112779541	74
777	chr2	GAGAUACCCAAAACCAUCAC	112775241	72
778	chr2	CUCGAAUUUAUCUUUGAUUG	112779564	72
779	chr2	GAUACUGGAAAACCAGGCGU	112775071	71
780	chr2	UUUGAUUGAGGGCGUCAUUC	112779576	71
781	chr2	GCUCUCCUUGAAGGUAGCU	112781730	71
782	chr2	UGAGACUCCAGACCUACGCC	112775071	70
783	chr2	GGGUGCUUAUAAGUCAUCAA	112778081	69
784	chr2	GCACCCAUGUCAAAUUCAC	112778110	69
785	chr2	UUCCUCUGAGUCAUUGGCGA	112781622	69
786	chr2	GGCUGCUGCAUUACAUAUC	112779510	65

**FIG. 28**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
787	chr2	CAUGGCCACAACAAACUGACG	112833479	80
788	chr2	GUGCAGUUCAGUGAUUCGUAC	112832771	79
789	chr2	CUUCGACACAUGGGAUACG	112832799	79
790	chr2	GAUCACUGAACUGCACGCUC	112832752	77
791	chr2	GGUGGUUCGGAGAUUCGUAGC	112833526	77
792	chr2	UCCGACCACCACUACAGCAA	112833501	75
793	chr2	CUCCGACCACCACUACAGCA	112833502	75
794	chr2	GAGGUGCUGAUGUACCAAGUU	112830466	74
795	chr2	CUUAUCAUCUUCAACACGC	112831327	73
796	chr2	AUCACUGAACUGCACGCUCC	112832751	72
797	chr2	AUCAUUUCACUGGCGAGCUC	112836212	72
798	chr2	AACCUAUCUUCUUCGACACA	112832809	71
799	chr2	UGGUGAAGUCAGUUUAUCC	112830398	70
800	chr2	AGAGGUGCUGAUGUACCAAGU	112830465	70
801	chr2	CAUCUUCAACACGCAGGAC	112831332	70
802	chr2	GCAGGCCGCGUCAGUUGUUG	112833472	70
803	chr2	AGUUUAUCCUGGCCGCCUU	112830408	69
804	chr2	UGAUGUACCAGUUGGGGAAC	112830473	68
805	chr2	UGAUAGCCCACUCUACAGC	112831299	65

**FIG. 29**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
806	chr2	UUCAGGACAUUACUAUUGCG	102164999	80
807	chr2	UGGGGCUAUCCGCUGGUACAG	102176596	80
808	chr2	GUGUAAAGUCCCCUGACCAG	102176593	79
809	chr2	UGGUCAGGGGACUUUACACA	102176609	79
810	chr2	AUGGGGCUAUCCGCUGGUCA	102176595	78
811	chr2	ACCACAUGCCAGUCCAGCGA	102176673	78
812	chr2	ACUUCUCCAUGCUACCCGAG	102176753	78
813	chr2	UGGAAGGGAAUGACUACGUUG	102175637	77
814	chr2	AGAAACAUGGGGCUAUCCGC	102176589	77
815	chr2	GAAGGUGACCGUCGCUGGAC	102176669	77
816	chr2	AAGCAGAACUACCCGUUGC	102165220	76
817	chr2	GCAAGCAAUAUCCUAUUACC	102166254	76
818	chr2	GAGCCCAGCUAAUGAGACAA	102168646	76
819	chr2	CUGGUCAAGGGACUUUACAC	102176608	76
820	chr2	GAGGCUUGUUCUGUAGAUAC	102164910	75
821	chr2	CAAGCAAUAUCCUAUUACCC	102166255	75
822	chr2	AUACAGUAUUAUAGCGUCAU	102175498	75
823	chr2	UAGAUGAAGGUGACCGUCGC	102176674	75
824	chr2	CAUGGGGCUAUCCGCUGGU	102176594	74
825	chr2	GAGAGGCUCACGUGGCCUCUC	102176751	74
826	chr2	GACAUUACUAUUGCGUGGUA	102165004	73
827	chr2	UAUGGAAGGGAAUGACUACGU	102175635	73
828	chr2	AUGGAAGGGAAUGACUACGUU	102175636	73
829	chr2	UAGUGGCUGGUGACAGUAAC	102176701	73
830	chr2	CUACCCGUUGCAGGAGACGG	102165229	72
831	chr2	AGUAAGCAAUGUCACUCAAC	102171840	71
832	chr2	AAUAGCUUCCCCUAGCACU	102171897	71
833	chr2	CACGUGCCUCUCGGGUAGCA	102176759	70
834	chr2	CCUGAAUCCUCCACCUUAGC	102164970	69
835	chr2	AAUUCUAUUACCCGGGUAAU	102166253	69
836	chr2	GGGUAGUUUCUGCUUAAAUA	102165200	68
837	chr2	AGUUGAGUGACAUUGCUCUAC	102171854	68
838	chr2	UAAAAGCUUCCCCUAGCAC	102171898	67
839	chr2	UUAAUGAAUUUAAUCGAUUC	102176552	67

**FIG. 30**

SEQ ID NO.	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
840	chr3	GUCUGGAUCACGUUCAAAGG	190656257	82
841	chr3	AUAGUCGGUUUGACACUGGA	190609151	81
842	chr3	ACUGGGGACUAGACACCAUG	190604157	80
843	chr3	CCAGAACGCGUGCGAUGACUG	190604141	79
844	chr3	CAUUCUCCAAAACUCCACGG	190645774	79
845	chr3	GACGUUGAUGUUGCCCCGAG	190648469	79
846	chr3	CGGUCUAGCAGUGAUGAGCA	190648669	79
847	chr3	CCUUACUAUAUGCGGUUCUCG	190604329	78
848	chr3	CUAGAAAAUAUGGCCUCUCG	190648468	78
849	chr3	AGUGAUAGUCGGUUUGACAC	190609155	77
850	chr3	UCUGAUGGAUUCUCGCAAUG	190627380	77
851	chr3	UUUGUUAUACUGACCCUCCG	190645772	77
852	chr3	AGCCCCAACUACGUGCUCCA	190648414	77
853	chr3	GCACAAUGUCCAAGCACCGA	190656350	77
854	chr3	AGUGGGCACUUGAUGCGAGC	190604192	76
855	chr3	AUCAGGCCUACCUUAACAUGC	190604406	76
856	chr3	GGAGCUACUCAUUCCCUGUA	190627344	76
857	chr3	UUGAUGUCACCAUUAAACGAA	190627445	76
858	chr3	UUGACCGAGACAGUCUGCCU	190645833	76
859	chr3	GGCACAAUGUCCAAGCACCG	190656349	76
860	chr3	CAGCUGGCCUUACUCUGAUC	190604268	75
861	chr3	UACUAAUGCGGUUCUCGGGG	190604326	75
862	chr3	AAACUAAAAUAGACCGUACA	190627345	75
863	chr3	GAUCUGGUAUUGGACUAGGC	190604284	74
864	chr3	ACCGACUAUCACUUGGUUA	190609177	74
865	chr3	AUUCUCCAAAACUCCACGGA	190645773	74
866	chr3	UUCCCUGGAGCACGUAGUUG	190648404	74
867	chr3	GCGGUCUAGCAGUGAUGAGC	190648668	74
868	chr3	CAUUGCACCAAGCUCAUUG	190656036	74
869	chr3	GUAUUGGACUAGGCAGGACC	190604290	73
870	chr3	UAGGCAGGACCGGGACCUUG	190604299	73
871	chr3	UGGAUUCUCGCAAUGAGGUU	190627385	73
872	chr3	UCAGUCUGGAUCACGUUCAA	190656254	73
873	chr3	GGUGCUUGGACAUUGUGCCU	190656333	73
874	chr3	GGAGGACUUCCCUCGGUGCU	190656347	73
875	chr3	CCCCGAGAACCGCAUUAGUA	190604341	72
876	chr3	UAUAGUUGCCAGUGUCAUUG	190604383	72
877	chr3	GACGUACGUUCAUCUCACC	190620413	72
878	chr3	GAUCCAGACUGAUGUCAGAC	190656231	72
879	chr3	GGGAGCAGGGGGUUCCAAUC	190656521	72
880	chr3	UCCAGAACGCCUGCGAUGACU	190604140	71
881	chr3	GCAACUAUACCUUGCAUGUUA	190604409	71
882	chr3	CCCAUGAUACCUUUCGUUAA	190627442	71
883	chr3	UUUGACCGAGACAGUCUGCC	190645832	71
884	chr3	AGUUGGGCUUAGAACAAACC	190648389	71
885	chr3	GGUUCCCUGGAGCACGUAGU	190648406	71
886	chr3	CGGUGCUCACGGGUCAUAAA	190648565	71
887	chr3	UCAUCACUGCUAGACCGCCU	190648651	71

**FIG. 31**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
888	chr5	ACUGCUGUGCAGUCGCACCC	132679802	82
889	chr5	UGGGCUUUGUAGGCGAGUCG	132677859	81
890	chr5	UGUGCCGGCAACUUUGUCCA	132674116	80
891	chr5	GGGACGGCUUCUUACCUUGG	132674504	80
892	chr5	GCUUUGUAGGCGAGUCGAGG	132677856	79
893	chr5	GGAAGCCAACCAGAGUACGU	132682520	79
894	chr5	AGUUGCCGGCAC AUGCUAGC	132674094	76
895	chr5	UGAUUAUCGCACUUGUGUCCG	132674121	76
896	chr5	GUUUUCCAACGUACUCUGGU	132682513	76
897	chr5	GAUGUCUGUUACGGUCAACU	132674474	75
898	chr5	UGAGAAGGACACUCGCUGCC	132679796	75
899	chr5	GUGUCCGUGGACAAGUUGC	132674108	74
900	chr5	GUAGGCGAGUCGAGGGUGGAA	132677851	74
901	chr5	UGUAGGCGAGUCGAGGUGGA	132677852	74
902	chr5	ACGGCUCGACAGGAACCUCU	132679871	74
903	chr5	CAAGUGCGAUUAUCACCUUAC	132674142	73
904	chr5	GAUCCUGAAACGGCUCGAC	132679861	71
905	chr5	CGACUGCACAGCAGUUCCAC	132679822	70
906	chr5	AGAAGUUUUCCAACGUACUC	132682517	70
907	chr5	AACGGCUCGACAGGAACCUC	132679870	68
908	chr5	CAACUCGGUGGCACAGAGUCU	132674459	67
909	chr5	GGUUCCUGUCGAGGCCGUUUC	132679853	66
910	chr5	AGCUGAUCCGAUUCUGAAA	132679852	66
911	chr5	UGUCGAGCCGUUUCAGGAAU	132679847	65

**FIG. 32**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
912	chr7	AUUCGUUCUGAAGAGGGUGAG	22727565	78
913	chr7	CAAAUUCGGUACAUCCUCGA	22727607	78
914	chr7	CAUUCCCUCAACUUGGUGUG	22729691	77
915	chr7	UCAGGGCUGAGAUGCCGUCG	22727609	76
916	chr7	CUUCCAAUCUGGAUUCUAUG	22728802	76
917	chr7	GGAGAAGGCAACUGGACCGA	22727452	75
918	chr7	CAUCUCAGCCCUGAGAAAGG	22727630	75
919	chr7	GUACCUCAUUGAAUCCAGAU	22728793	75
920	chr7	ACAUUUGCCGAAGAGGCCUC	22731554	75
921	chr7	ACAGUGUCCUGGACAACUCA	22729730	74
922	chr7	CACACCAAGUUGAGGGAAUG	22729676	73
923	chr7	UCUUCCCCCACACCAAGUUG	22729684	73
924	chr7	GUGUCCUCAUUCCCUCAACU	22729684	73
925	chr7	GACAGUGUCCUGGACAACUC	22729729	73
926	chr7	GGAAGGCAGCAGGCAACACC	22727486	72
927	chr7	CAUGUGUGAAAGCAGCAAAG	22728727	72
928	chr7	CUUCCCCCACACCAAGUUGA	22729683	72

**FIG. 33**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
929	chr1	CGTGACTCTGACCTGCCCGG	154429253	79
930	chr1	AGCUGCCCCAAAGAGUACGG	154465195	76
931	chr1	UCGAGGUAUUGUCAGACCCC	154465287	74
932	chr1	GCGUGACUCUGACCUGCCCG	154429252	72
933	chr1	UCGGUGCAGCUCCACGACUC	154429380	72
934	chr1	CUGCCAGUUAGCAGUCCCGG	154434602	72
935	chr1	GACACUACUGGCGACGCACA	154434638	72
936	chr1	UGUGCGUCGCCAGUAGUGUC	154434652	72
937	chr1	GUCAUCCACCGACGCCUGGAG	154436010	72
938	chr1	ACGACGCCUGGAGCGGCCUG	154436017	72
939	chr1	AAUGCAGAGGAGCGUUCCGA	154454551	72
940	chr1	AUCUGGUCGGUUGUGGGCUCG	154465304	71
941	chr1	CUGAUGUCAUAAGGGCUCCG	154465336	71
942	chr1	CACCCCAUCCUGACGACAA	154430579	70
943	chr1	UGCCAGUUAGCAGUCCCGGA	154434603	70
944	chr1	UGCAUCCGCCGUACUCUUUG	154465201	70
945	chr1	AGCUCAGAUUAUCGGGCUGAA	154435127	69
946	chr1	ACCUGUCCAAGGCGUGCCCA	154436096	69
947	chr1	UCAGGCCGCUCCAGGCGUCG	154436004	69
948	chr1	UGAUGUCAUAAGGGCUCCGU	154465335	69
949	chr1	AACUAUUCAUGCUACCGGGC	154429404	68
950	chr1	CACGUGCCUCAGGCCGCUCC	154436012	68
951	chr1	AGAGGAGCGUUCCGAAGGCC	154454546	68
952	chr1	GGAGCCGUGCCAGUAUUCCC	154434563	67
953	chr1	CUCCGGGACUGCUAACUGGC	154434590	67
954	chr1	UACGGUUUGAGCUCAGAUAU	154435118	67
955	chr1	AGCCAGCAGCGCGCAGCCGA	154405643	66
956	chr1	CAGCCUUUGUCGUAGGGAU	154430571	66
957	chr1	CUCCCUCCGGGACUGCUAAC	154434594	66
958	chr1	CGGAGGCCAUGGGCACGCCU	154436101	66
959	chr1	GUGGACACCUCGUUCUCAGC	154448145	66
960	chr1	CCUGCGCAGGGCAGCGCCUU	154405698	65
961	chr1	AUGUGCGUCGCCAGUAGUGU	154434651	65
962	chr1	CCGUGGCCAGAAACCCCCGC	154435043	65
963	chr1	CAUGCAUCCGCCGUACUCUU	154465199	65

FIG. 34

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
964	chr5	CCACAGACUGUACGGCAAGG	55941103	83
965	chr5	UAUUCUACCGUGGUACACAG	55941547	83
966	chr5	GAACUACUAGUCCUUCACUG	55941085	81
967	chr5	CACUGUCCAGUAUUCUACCG	55941557	81
968	chr5	UCUUAAAUGGUGCGAUGCA	55954828	81
969	chr5	UAAGCAAACAGGCACGACUA	55947566	79
970	chr5	GUGCAUCGCACCUUUUAAG	55954814	79
971	chr5	GGUGGAAUGGACUACUCCAA	55954923	79
972	chr5	AACUGUACAACUCGUGUGGA	55957213	78
973	chr5	GCGGCUACAUAGCCUCAGUGA	55941084	77
974	chr5	AGACUUGGACUGACGGAACU	55941527	77
975	chr5	GACUAGUGACACAUJUGUACA	55951541	77
976	chr5	AAGCACCCUGUAUCACAGAC	55954855	77
977	chr5	ACAGAACUGUACAACUCGUG	55957217	77
978	chr5	GUUUAGGAUUCGCUGUAUGA	55960470	76
979	chr5	CUUCCAAAGGACCUACUGUU	55952055	75
980	chr5	UGUGUACCACGGUAGAAUAC	55941563	74
981	chr5	GGACCAAAGAUGCCUCAACU	55963363	74
982	chr5	CAAUGUGUCACUAGUCAAAG	55951560	73
983	chr5	GCCAGGUUAUACCUCUUAAA	55954816	73
984	chr5	CCUGGUCCAUCAGCAUUAAC	55952305	72
985	chr5	ACUCGGAUCUUGAGAAGACU	55941512	71
986	chr5	UGUCCGAACAGUAGGUCCUU	55952064	71
987	chr5	UGGACUUACGAGCUUGUUUA	55952258	71
988	chr5	AUGGAGGCUGCGACUGAUGA	55941145	70
989	chr5	CACAUUGUACAAUGGUACGAA	55951532	69
990	chr5	CACCCAAAGCAUGUUAUCUU	55954954	66

**FIG. 35**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
991	chr1	UUGCCTUGGUCCUCUGACUG	206772395	75
992	chr1	AGGCGCAUGUGAACUCCCUG	206770951	74
993	chr1	CGGAGAUCUCGAAGCAUGUU	206772319	74
994	chr1	GAAGAUGUCAAACUCACUCA	206768698	73
995	chr1	GGCGCAUGUGAACUCCCUGG	206770950	73
996	chr1	AAGGCGCAUGUGAACUCCCCU	206770952	73
997	chr1	CAAGGCGCAUGUGAACUCCC	206770953	73
998	chr1	GCGCCGUAGGCCUCAGCCUGA	206770927	72
999	chr1	GGUCUUCAGGUUCUCAGCUCC	206770948	71
1000	chr1	GGGUCUUGGUUCUCAGCUUG	206770994	71
1001	chr1	GAUGAUCCAGUUUUACCUUG	206771013	71
1002	chr1	AAACUGGGAUCAUCUCAGACA	206771035	71
1003	chr1	GGAUCAUCUCAGACAAGGCU	206771040	71
1004	chr1	UCGUAUCUUCAUUGUCAUGU	206768659	70
1005	chr1	UGGCCUCACCCCAGUCAGG	206772398	70
1006	chr1	GAACCAAGACCCAGACAUCA	206770971	69
1007	chr1	UCACAUGCGCCUUGAUGUCU	206770974	69

**FIG. 36**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1008WO 2023/201276		GCGCCGCCAGCAGCACUACG 37/103	11798648PCT/US2023/065691	
1009	chr11	UGGCAUUCAGGGGUUACCUG	117999243	82
1010	chr11	UCGCUCUCCCGAAGUAACAAG	117994104	81
1011	chr11	GCGCCGCACAUACAGCUGGA	117995669	81
1012	chr11	UGGGUAGCUGAAUCUUCCCCG	117993295	80
1013	chr11	CUGGUUCCACUGUCCCCGUUG	117999441	79
1014	chr11	GCUGGUUCCACUGUCCCCGUU	117999442	78
1015	chr11	CGUCUUGGCUCAGACGCUA	117986530	77
1016	chr11	UGCAGGUAGGUCAUAGGACA	117989491	77
1017	chr11	GUUCCCCGGCACCUUGCGAA	117993387	77
1018	chr11	AGGCCUCCUCAUCAAGCGGG	117998788	77
1019	chr11	GCAAGCCUGCCUCAUCAACC	117999349	77
1020	chr11	CUUCAAACCAACAGACGGA	117988403	76
1021	chr11	CUGCGGUAGGUCAUAGGAC	117989492	76
1022	chr11	GU AUGAGAUUGCCA U UCGCA	117993388	76
1023	chr11	GAUUGCCA U UCGCAAGGUGC	117993394	76
1024	chr11	UUACUUUCGGGAAGCGACAGA	117994085	76
1025	chr11	CUCCUCAUCAAGCGGGUGGA	117998784	76
1026	chr11	UACGAGGCACGGCAGCAUCC	117986468	75
1027	chr11	GCCAGCAGCACUACGAGGC	117986479	75
1028	chr11	GUCGCUUCCCGAAGUAACAA	117994103	75
1029	chr11	GCUAUGAAGUGGCGCUCCUG	117988498	74
1030	chr11	GGUCACUGCGGUAGGUCAU	117989497	74
1031	chr11	GGUCCAAGGUCACUGCGUA	117989504	74
1032	chr11	UCGGGAAGAUUCAGCUACCC	117993309	74
1033	chr11	UGUCGCUUCCCGAAGUAACA	117994102	74
1034	chr11	ACACAGGGUGGCUCGGCCUU	117999167	74
1035	chr11	GAAGAAUCGCCUUGACAGA	117999314	74
1036	chr11	CCAGGCAUCUCCCGAAUUUG	117999331	74
1037	chr11	CGAGUCACUCACUUGACUGC	117999628	74
1038	chr11	CAGGAGCGCCACUUCAUAGC	117988483	73
1039	chr11	ACCUGGUAUCUCCUCAGGU	117989442	73
1040	chr11	AUUGCCA U UCGCAAGGUGC	117993395	73
1041	chr11	AAGCGACAGAUGGUUUCACC	117994075	73
1042	chr11	CGGAGGCCUUCGCCUACUGCC	117995658	73
1043	chr11	CACCAUCCACCCGCUUGAUG	117998794	73
1044	chr11	GGCUGGUUCCACUGUCGGU	117999443	73
1045	chr11	AGGAGUGACCAUUCCUCAGU	117999464	73
1046	chr11	AGCUCCAGUCAGAUAUUCCC	117999508	73
1047	chr11	UUGGACCUGUACCACAGCAA	117989532	72
1048	chr11	UCAUUUGCGGGGCCAUCUU	117993320	72
1049	chr11	CAAGGUGCCGGAAACUCA	117993406	72
1050	chr11	GGCGCCGCACAUACAGCUGG	117995670	72
1051	chr11	CAUCCACCCGCUUGAUGAGG	117998797	72
1052	chr11	UGGCCCGGUAGCCA U UCGUG	117989531	71
1053	chr11	UGUACCACAGCAAUGGCUAC	117989539	71
1054	chr11	CCCCCAGGCUGACAGAACGC	117998944	71
1055	chr11	ACCAGGCAUCUCCCGAAUUU	117999332	70

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1056	chr21	UCACAGCUCAGUACCUAAGG	33268516	78
1057	chr21	UGAGAAUCACAUUCCGUCA	33276636	78
1058	chr21	UGUGAGCAAACAACCCAUGA	33283237	78
1059	chr21	AUCUCAGGGCUUACCGUCAU	33283238	77
1060	chr21	CACAGCUCAGUACCUAAGGU	33268517	76
1061	chr21	CUUGUAAACGCACCACAGCA	33288190	76
1062	chr21	CCCAGCGUCCGUCCAUGGCG	33266469	75
1063	chr21	UUGAGAAUGAAUACGAAACU	33279844	75
1064	chr21	GAUGACGGCCACCAUCCAGG	33288118	75
1065	chr21	UCGGCUGCUUCGCCUUGCUG	33288190	75
1066	chr21	AAUAAGUUGGUCCAUGGCUCC	33283140	74
1067	chr21	CAUCUCAGGGCUUACCGUCA	33283239	74
1068	chr21	AUUCUCAUCCGACAAUGGAA	33296229	74
1069	chr21	CUUUCACAGCUCAGUACCUA	33268513	73
1070	chr21	CAAGGUGUGGUACCAUACU	33276666	72
1071	chr21	UUAGCCAUUAUUUGGACCCCC	33279765	72
1072	chr21	CUUAUUGUGUUCAAGUUCGA	33283169	72
1073	chr21	UCCCCAGCUUUGUUCCGAUC	33283186	72
1074	chr21	UUCCUGAUCGGAACAAAGCU	33283196	71
1075	chr21	CGUCCAUGGCGUGGAGCCUU	33266478	70
1076	chr21	GGACUUUAUAUGUGCAAUAC	33279892	70
1077	chr21	ACUUAUUGUGUUCAAGUUCG	33283168	70
1078	chr21	CAUUCAGGGGUCCAAUAA	33279757	69
1079	chr21	GUGACCACACCUUGAGAGUC	33276687	68
1080	chr21	CCGUCCAUGGCGUGGAGCCU	33266477	67
1081	chr21	CUUCCUGAUCGGAACAAAGC	33283195	67
1082	chr21	AGUACUUCUACUUGCAUUCC	33279771	66

**FIG. 38**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1083	chr5	UGUCCGAGACACCAAAAUCG	132660209	79
1084	chr5	GCAGCCUGACACGUUGAUCA	132659748	78
1085	chr5	GAGCGGAUUCUGCCCGACA	132659812	78
1086	chr5	GCAAGUGAGAGCAAUGACCG	132658250	77
1087	chr5	GAAUCCCUGAUCAACGUGUC	132659756	77
1088	chr5	GGUGUCUCGGACAUGCAAGC	132660187	76
1089	chr5	UGGUCAACAAAAGCGCCAUG	132658230	75
1090	chr5	CAUGGCGCUUUUGUUGACCA	132658245	75
1091	chr5	GCCCCGCACAAGGUCUCAGCU	132659823	75
1092	chr5	CCGAGACACCAAAAUCGAGG	132660212	75
1093	chr5	GAGCAGGUCCUUUACAAACU	132660223	74
1094	chr5	UGGAGCAUCAACCUGACAGC	132659462	73
1095	chr5	AAACUGGGCCACCUUCGAUUU	132660208	72
1096	chr5	CACGUUGAUCAGGGAUUCCA	132659739	70
1097	chr5	UGCCCGCACAAGGUCUCAGC	132659822	70
1098	chr5	UGCAGCCUGACACGUUGAUC	132659749	69
1099	chr5	UUAAAAGAAACUUUUUCGCGA	132660264	69
1100	chr5	GCUAGCCGACACUCACCUUC	132658359	67
1101	chr5	CGAGGUGGCCAGUUUGUAA	132660227	67
1102	chr5	UUUAAAAGAAACUUUUUCGCG	132660263	67
1103	chr5	GAAUCCGCUCAGCAUCCUCU	132659787	65
1104	chr5	CCACCUUCGAUUUUGGUGUCU	132660200	65

**FIG. 39**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1105	chrX	UGGACAUGGAAUCCACCCGA	118741093	81
1106	chrX	UAACAUGGUUAUGUAGAGUG	118773899	80
1107	chrX	AUGGACAUGGAAUCCACCCG	118741092	79
1108	chrX	AUAGUGCCUUUAACUUCGG	118758238	79
1109	chrX	CGAGGGUCGCACUCACCCGU	118727725	78
1110	chrX	UAGAAGUACCCCUGAAUGAG	118746999	78
1111	chrX	GUCGCACUCACCCGUAGGCG	118727720	76
1112	chrX	CGGGACUGGUAUUCUUCCA	118749736	76
1113	chrX	AGUAUAGUUAGUGUCGGGAC	118749750	76
1114	chrX	UUUGAGCUGGCUCCCCUCGGG	118741093	75
1115	chrX	CAAUUUGAGCUGGCUCCCUC	118741096	75
1116	chrX	CCAGCUAAAUGUAGUCUA	118741118	75
1117	chrX	UCGGGACUGGUAUUCUUC	118749737	75
1118	chrX	AUUGUUCCAGUCAUCGUCGC	118773937	74
1119	chrX	GGACUUCUAAAACUUACCAC	118758242	73
1120	chrX	GUGGAUUUGCGCUUCUUACC	118773880	73
1121	chrX	GAGACACUCAAAUUUGUCAC	118741033	72
1122	chrX	AUUGCACCUGCGACGAUGAC	118773931	71
1123	chrX	CACUCUCAUUGGUGCUACAC	118747030	70
1124	chrX	CCAUAGACUACAAUUGAGC	118741106	69
1125	chrX	UGAUGACCUUAUGUGCAAU	118761198	69
1126	chrX	CACAUAAUGUUUUUCUACGUA	118761290	69
1127	chrX	CUGACUGUGUUCAAAGUAUC	118766882	69
1128	chrX	GGCGGGGGCGCCGCCUAC	118727722	67
1129	chrX	AACUUACCACGGGAAGUUAA	118758232	67
1130	chrX	UCUAUUGAACGACGAGUUUC	118746968	66

**FIG. 40**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1131	chrX	UCUUCGAUAGGUUCUUCACCU	115005315	74
1132	chrX	GCUGAAUAGGAGCAUACCUU	115009591	74
1133	chrX	UCAAGAAGUACACCUAUGC	115013810	74
1134	chrX	GAACUAAAAUACCGAAACAU	115015691	73
1135	chrX	AUUGCUCAGAUGACGGAAUU	115007968	71
1136	chrX	CUUGCUGGAAUAGGUCCCAA	115009587	71
1137	chrX	CGAGUAAAAGUAAGAUAGAC	115009647	71
1138	chrX	CCUGGGCAGAACUACUUAU	115014439	71
1139	chrX	AUAGUGGAUCCCGGAUACUU	115015781	71
1140	chrX	AGGAUGCAGAUUUCCUAUU	115010748	70
1141	chrX	UAGAGAUAAACCUAAGUAUCC	115015784	70
1142	chrX	UCUUCAGACACCGAGAUAAA	115017180	70
1143	chrX	CUACGUUUUCUGGUACCAUU	115005272	69
1144	chrX	GUAGCAAAGUUUUCUUCGAU	115005303	69
1145	chrX	GUAAGAUAGACUGGCCGCAA	115009656	69
1146	chrX	GCUUUCGUUUGCUUGGCUAU	115017249	67
1147	chrX	AGAAAACUUUGCUACGUUUC	115005283	65

**FIG. 41**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1148	chr6	UGCAGGACUCACCACCAAUG	52186453	80
1149	chr6	GGUUGACCAUCACAGUCCGG	52187703	80
1150	chr6	AGGUUGACCAUCACAGUCCG	52187704	79
1151	chr6	GGAUCGGUUGUAGUAUCUG	52187768	79
1152	chr6	UCCCCUCUGCAGCCGCAAUG	52189060	79
1153	chr6	AGAGAUCCUGGUCCUGCGCA	52189192	79
1154	chr6	GGCCACACAUGGUGGACAAUCG	52189272	78
1155	chr6	GAACUUCCCCCGGACUGUGA	52187709	77
1156	chr6	GAGAAGAUACUGGUGGUCCGU	52189247	77
1157	chr6	ACAACCGAUCCACCUCACCU	52187792	76
1158	chr6	UGGGCUGCAUCAACGCUGAU	52189131	76
1159	chr6	CAUCAACGCUGAUGGGAACG	52189138	76
1160	chr6	CACCCCGAUUGUCCACCAUG	52189282	75
1161	chr6	UUGGGCUGCAUCAACGCUGA	52189130	74
1162	chr6	AAGAGAUCCUGGUCCUGCGC	52189191	74
1163	chr6	CAGUAUCUUCUCCAGGCCGGA	52189224	74
1164	chr6	GUGGACAAUCGGGGUGACAC	52189263	73
1165	chr6	GAGGCAAAGUGCCGCCACUU	52189112	72
1166	chr6	UAGGCCACAUGGUGGGACAAU	52189274	72
1167	chr6	CAGCGUUGAUGCAGCCCAAG	52189114	71
1168	chr6	AUCACAAUCCCACGAAUCC	52187665	69
1169	chr6	CUUCCGGCUGGAGAAGAUAC	52189237	69
1170	chr6	UUGGGCAUCCUGGAUUCGU	52187661	68
1171	chr6	GUAGUAAUCUGAGGACCUUU	52187759	67
1172	chr6	GGAGGCAAAGUGCCGCCACU	52189111	67
1173	chr6	UGGUAUUGGUAUUCCGGUUA	52187733	66

**FIG. 42**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1174	chr22	GUGUGACGUUGGAUCGCUGG	17103516	84
1175	chr22	UUCACGAUGCCGGUUCCCAG	17109058	84
1176	chr22	CAUUCCGUGAGUCCUCGG	17109635	82
1177	chr22	UCCAUUCGAUGUGAGGCCACG	17097911	81
1178	chr22	ACAAGGAGACCUGUUCCCCG	17097908	81
1179	chr22	CGUCCCCGACCUGUUUCGGCG	17108772	81
1180	chr22	AGACCCACACACGCCUACG	17109564	81
1181	chr22	ACUCGGGCUGGGACACGAUG	17109788	81
1182	chr22	UCGCAGGGAGGCCACCCG	17085176	80
1183	chr22	GCCAGCAUCCUGUACCUCGA	17098791	80
1184	chr22	GUGGGACCCAACAUCACCG	17102163	80
1185	chr22	GCAGAGUUACCGCAGGUUAUG	17102295	80
1186	chr22	CAGUUCCUGCUACCGCCUG	17108434	80
1187	chr22	CGUAGUCUGCUACUUCAGCG	17108733	80
1188	chr22	GGGCCGCAUGCACCACGUAG	17108868	80
1189	chr22	UGUCGGGGGACAACUACCUUG	17108894	80
1190	chr22	ACUGCGGCACCGUCAGCCAG	17109277	80
1191	chr22	GGUCAGGGUCAACCACAAAG	17100377	79
1192	chr22	AUGGGUAGAUUCGUUCCACA	17102210	79
1193	chr22	AAACAGUCGCGGAGUGUCUG	17104766	79
1194	chr22	CCCGCGGCACCGCGGCCAAG	17108579	79
1195	chr22	GCGACCACCGAAAGGCCGUG	17108642	79
1196	chr22	CCCCCGACAGCUCCCCUACG	17108868	79
1197	chr22	CGGGCCGCAUGCACCGCGUA	17108867	79
1198	chr22	UGGCUGACGGUGCCGCAGUC	17109293	79
1199	chr22	AGGAGGUACAGGAGACGCCAU	17109421	79
1200	chr22	GCGAGAGCAUCAAGCCUUCG	17109465	79
1201	chr22	CCAUUCCGUGAGUCCUCG	17109636	79
1202	chr22	GGGAGCAGACCAGCGCCGG	17085207	78
1203	chr22	CACGUCGAGUCUCACCGGCU	17085227	78
1204	chr22	ACCUGAGCUAUGC AUGGCG	17102028	78
1205	chr22	GCACAUUGCACCAUACCUG	17102298	78
1206	chr22	GCGUAGAGUGAGUGUGACGU	17103527	78
1207	chr22	CCCGCUCAUGGACAGGUUCG	17108805	78
1208	chr22	GCUCAUUUGACAGGUUCGAGG	17108808	78
1209	chr22	CCGGGCCGCAUGCACCGCGU	17108866	78
1210	chr22	AUUCGAACCAGUCGGGACAG	17108961	78
1211	chr22	GGGGCCCCGUUCACCGAUGC	17109068	78
1212	chr22	CGCCAGUGCCAGCCGGACUG	17109293	78
1213	chr22	GACGUGAGGGAGCACCUUGA	17109463	78
1214	chr22	GGGCUGCAGUAGACCCGCCA	17109534	78
1215	chr22	UCCUCCUCGUAGGGCGUGUG	17109556	78
1216	chr22	CCGGUGGUCCAGGAGUCGCA	17085191	77
1217	chr22	UCCCCGUGGCUCACAUUCGAA	17097922	77
1218	chr22	CAGAUAAUCUUGACCCUCG	17098793	77
1219	chr22	AGCCAGCAUCCUGUACCUUG	17098790	77
1220	chr22	GCUGUGUUCUUACCCGCCUG	17098883	77
1221	chr22	UGAUGAACCAAGUACACCCAC	17105868	77

**FIG. 43**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1222	chr11	CUUAUAAUAAAUAUGGUCCG	112148723	79
1223	chr11	UUAUUUCAGAUAAUGCACCC	112148728	75
1224	chr11	UUUGAAUCUUCAUCAUACGA	112143710	74
1225	chr11	AUAGUUACAGCCAUACCUCU	112148681	73
1226	chr11	AUGACUGAUUCUGACUGUAG	112150076	72
1227	chr11	GAAUCCUCCUGAUAAACAUCA	112143795	71
1228	chr11	GUAUCCUUGAUGUUUAUCAGG	112143803	71
1229	chr11	AGAUAGCCAGCCUAGAGGUA	112148679	71
1230	chr11	CAAGUUCUCUUCAUUGACCA	112150121	71
1231	chr11	GCAUCUUAAUUAUC AUGUCCU	112143747	70
1232	chr11	AAGGCUCAGUCUUACCUUAA	112148605	70
1233	chr11	UUACAGCCAUACCUUAGGGC	112148685	70
1234	chr11	UCAGUCAUAUUUCAAAUAG	112150103	70
1235	chr11	UUCUUUCAGAGAAGUGUCCC	112143752	69
1236	chr11	UAUAAAGAUAGCCAGCCUAG	112148684	69
1237	chr11	CAAAUAGAGGCCGAUUUCCU	112150116	69
1238	chr11	UGCAUCUUAAUUAUC AUGUCC	112143746	68

**FIG. 44**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1239	chr2	ACGCCGAGUUUGAAGAUCAG	102375976	80
1240	chr2	UUCACGUCCCCCACAUUACUG	102367852	75
1241	chr2	AUCAUGCAAAGCAAUUCUG	102367989	75
1242	chr2	AACGCCGAGUUUGAAGAUC	102375975	75
1243	chr2	UGUAAUUUAUUGGAUGUUCG	102384941	74
1244	chr2	GCAGACAAUGGCUGAUUAUCCC	102390072	74
1245	chr2	ACUUCCGGUUCGUCUCUACC	102396844	73
1246	chr2	CAAGCCAGGUAGAGACGAAC	102396852	73
1247	chr2	UAUAAGCACCCAAGGGUCA	102362685	72
1248	chr2	CCCCUUCAACCACAGUAAUG	102367849	72
1249	chr2	CCCUUCAACCACAGUAAUGU	102367848	71
1250	chr2	CCCCCACAUUACUGUGGUUGA	102367859	71
1251	chr2	GAGAAUUACCCUUGACCCUU	102362688	70
1252	chr2	UCGGCGUUCUUCUUUAUCGU	102375948	70
1253	chr2	AGAUCGCAGUAAUAGUUC	102381635	70
1254	chr2	GGGUAAUUCUCUACAAUUC	102362664	69
1255	chr2	GCAAUUCUCGAGGAACCUU	102367979	69
1256	chr2	CGGCGUUCUUCUUUAUCGUU	102375947	69
1257	chr2	ACUGCAACAUGGUUAAGCUU	102381657	69
1258	chr2	CUCUUGUGAAGACGUGGCCU	102390077	69
1259	chr2	CCUGUGUCAAUCAACUCAAC	102368030	68
1260	chr2	GAACGCCGAGUUUGAAGAUC	102375974	68
1261	chr2	CCUCUUGUGAAGACGUGGCC	102390078	67
1262	chr2	CAAUUCUCGAGGAACCUU	102367978	66

**FIG. 45**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1263	chr2	AGGUCGUUACUACCCAUAGAA	102423959	80
1264	chr2	AAGGAGCAACCGAAUCGUAG	102437252	80
1265	chr2	AGCAAGGAUCAGACGCUUGG	102447203	80
1266	chr2	UCUAAUGGAUCUCCAUUCGA	102424007	79
1267	chr2	UCCAGCGGUACCUGGUACA	102424410	79
1268	chr2	GAGCAAGGAUCAGACGCUUG	102447202	79
1269	chr2	UAGGUCGUUACUACCCAUAGA	102423960	78
1270	chr2	AUGUGAUGC GGAAUACUCAC	102424301	78
1271	chr2	GUACCGGACCUACCAGAGCA	102447187	78
1272	chr2	ACGACCUAUCUGAUGGUCAA	102423987	77
1273	chr2	CCCAGAAUUUAUUCACCCUG	102424089	77
1274	chr2	ACCUUGUACCAGGUUACCGC	102424399	77
1275	chr2	AGUCGGAUACUGUGAGUUCG	102437323	77
1276	chr2	UGGUACCAACAACCUUCGAA	102424007	75
1277	chr2	ACCCAGAAUUUAUUCACCCCU	102424090	75
1278	chr2	GUUGUUCAAGUGAGAACCAU	102437358	75
1279	chr2	AAAACUCUCAGGGAUUCACG	102452083	75
1280	chr2	GAUCUCCAUUCGAAGGUUGU	102424000	74
1281	chr2	UUCGAAUGGAGAACCAUUAJG	102424021	74
1282	chr2	AGAUAAUUCUGGAUCCUGUCG	102441351	73
1283	chr2	AAGCACGAUUUGGCUUUGAA	102443241	73
1284	chr2	GCUUGGCACCAUCGGGACCC	102447106	73
1285	chr2	GAGGGCACUCGCCGCCAGCA	102447120	73
1286	chr2	UUUCAAUCCAGUGCCUGUAG	102447142	73
1287	chr2	CGGGAGAACAAUGUCUGUU	102423285	72
1288	chr2	CGUUACUACCCAUGAAGGGC	102423955	72
1289	chr2	GACCCAGAAUUUAUUCACCCC	102424091	72
1290	chr2	GCUACAUCAUAGGGGUCCU	102424231	72
1291	chr2	AAACUUCAUCCACAUACGAUU	102437249	72
1292	chr2	UCACUCAGCGUGAUCUUCGC	102445262	72
1293	chr2	AGAGCUUUUUUCACGAGAUG	102451956	72
1294	chr2	ACCAGCUCAGAAUUACCUCU	102452107	72
1295	chr2	CCCCAGGGUGAAUAAUUCU	102424101	71
1296	chr2	GGAUCCUGUCGAGGACACAC	102441360	71
1297	chr2	UCUCAGUACCUGAGGCGAAA	102443319	71
1298	chr2	CGGCGAGUGCCCUCUCUAC	102447141	71
1299	chr2	UGAAAUAGUGCUGCUGUAC	102447171	71
1300	chr2	CAAGCGUCUGAUCCUUGCUC	102447187	71
1301	chr2	AGAGCAAGGAUCAGACGCUU	102447201	71
1302	chr2	AAGCCUCUCCAAGUAACUGU	102451989	71
1303	chr2	UUCUUUCAGUUGGACGGACU	102445313	70
1304	chr2	CUGGGGUCCAUUGACAUAGUU	102451833	70
1305	chr2	AGGCAUGUGGUAGCGCAUUU	102452044	70
1306	chr2	GUUGCAGGAGAGCGAAUUA	102423328	69
1307	chr2	CACAAAGUCCAGCGGUACC	102424403	69
1308	chr2	GCUUUGCAGCUAAUAGUUAA	102443210	69
1309	chr2	AUUAGCUGCAAAGCACGAUU	102443231	69
1310	chr2	ACUGGGGUCCAUUGACAUAGU	102451834	69

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1311	chr11	UGUGCACACGGAUACCCCA	102791447	82
1312	chr11	GAUGCUAUACUACGUUCG	102795203	82
1313	chr11	GCGCACAAUCCCUUCUACC	102792712	78
1314	chr11	AGAUGUGUUUGCUCCCAGCG	102797282	78
1315	chr11	CUCCCAUUCUACUGAUACG	102795538	76
1316	chr11	AACACAUCUGACCUACAGGU	102797256	76
1317	chr11	GAGCUAACUUCGGGUAGA	102792713	75
1318	chr11	UGAUGCUUAACUACGAUUC	102795204	75
1319	chr11	UCACUGAGGGAACCCUCGC	102797284	75
1320	chr11	GAUGGCAUCCAAGCCAUAUA	102795456	74
1321	chr11	UCGUUUUAUUAUCAUCAUACC	102790805	73
1322	chr11	GAUUGGCAGGGAAUAAGUAC	102791487	73
1323	chr11	AGCUCAACUUCGGGUAGAA	102792714	73
1324	chr11	UUGAUGCUUAACUACGAUU	102795205	73
1325	chr11	CUUACUGUCACACGCUUUUG	102795247	73
1326	chr11	UACCCUAGCUACACCUUCAG	102795507	73
1327	chr11	CACCACUGAAGGUGUAGCUA	102795517	73
1328	chr11	UAUAAAACGAUCUAUGGAUCC	102790779	72
1329	chr11	UGAUGAAUAAAACGAUCUA	102790786	72
1330	chr11	GCAAGAUGUGGACUAGUCC	102797992	72
1331	chr11	AAUCGUAGUUUAUAGCAUCAA	102795220	71
1332	chr11	CAUACUCACCAUUAUGGCU	102795460	71
1333	chr11	UCACCACUGAAGGUGUAGCU	102795516	71
1334	chr11	AAACCGGACUUCAUCUCUGU	102792629	70
1335	chr11	UGGUGAUGUUCAGCUAGCUC	102795487	70
1336	chr11	AGCCCCGAUAUCAGUAGAAU	102795548	70
1337	chr11	CUAAUAGCUGGUUCAACUGC	102790426	69
1338	chr11	AGCUUACUGUCACACGCUUU	102795245	68
1339	chr11	GCUUACUGUCACACGCUUUU	102795246	68
1340	chr11	UGAGCAUCCCCUCCAAUACC	102796719	67
1341	chr11	GCUAUCAUUUUGGGAUACC	102790773	66
1342	chr11	CACACGCUUUUUGGGUUUGU	102795255	66
1343	chr11	UGGCUUACCUUCUGAAUUGU	102796673	66

**FIG. 47**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1344	chr16	GUCGCGGUCGUAGUCCUCAG	55488671	84
1345	chr16	CCGCUGGUGCGGCACCACUG	55488669	83
1346	chr16	ACAUCUGGGUUGCCGCAGCG	55483058	82
1347	chr16	GAUUCGAGAAAACCGCAGUG	55484104	82
1348	chr16	GCCAGGGAGCGCUACGAUGG	55479481	81
1349	chr16	AUGGAGGGCGCUAAUGGCCCG	55479497	81
1350	chr16	GAUGAUGGGCGACGGCGCGG	55479567	81
1351	chr16	UACGAUGAUGACCGCAAGUG	55489802	81
1352	chr16	ACCAUGCGGAAGCCACGCUG	55483058	80
1353	chr16	GUCCGUCCUUACCGUCAAAG	55485320	80
1354	chr16	UCCUUCAGCGUUGCCGCCA	55488557	80
1355	chr16	CUUGAUGAUGGGCGACGGCG	55479570	79
1356	chr16	ACCAGCGGUAGCCAUCCGUG	55488642	79
1357	chr16	CUUGGGGUACCCUCGCUCCA	55498324	79
1358	chr16	GCCGCCUGCAGGUCCACGA	55502870	79
1359	chr16	GAUGCCGUCGUGGACCUGCA	55502878	79
1360	chr16	UUGUCCCACUUGGGCUUGCG	55483100	78
1361	chr16	GAGUCCGUCCUUACCGUCAA	55485322	78
1362	chr16	GACGAUGAGCUAUGGACCUU	55485411	78
1363	chr16	ACCAUGGGCGGCAACGCUGA	55488568	78
1364	chr16	ACGGAUGGCUACCGCUGGUG	55488658	78
1365	chr16	ACACACAUCUUUCCGUCACUG	55489750	78
1366	chr16	CUACGAUGAUGACCGCAAGU	55489801	78
1367	chr16	GAUGGCAUCGCUCAGAUCCG	55493264	78
1368	chr16	AAAGAUUGAUGCGGUUAACG	55497024	78
1369	chr16	CGACGGCAUCCAGGUUAUCG	55502853	78
1370	chr16	GCCGCAGCGUGGCUUCCGCA	55483047	77
1371	chr16	UGACGAUGAGCUAUGGACCU	55485410	77
1372	chr16	UUAGUGGUCCGUGUGAAGUA	55485617	77
1373	chr16	AGUGGCACCCACCGGUCUCA	55488711	77
1374	chr16	GCCGCAGUGACGGAAAGAUG	55489761	77
1375	chr16	AGGGCCUCGUACCUUGGUCA	55489819	77
1376	chr16	CAAACUCGUGGGCUGGCCACG	55491819	77
1377	chr16	CAUGGCGUGGCCAACUCGU	55491830	77
1378	chr16	UCCACUCGCUUGGACAUCAGG	55498368	77
1379	chr16	UAAAGGCGGCAUCCACUCGC	55498379	77
1380	chr16	AUGGCCGGGGCGCGCUCAC	55479509	76
1381	chr16	GCGCGCUCACGGGUCCCCUG	55479519	76
1382	chr16	UCGCCGGGAACUUGAUGAU	55479581	76
1383	chr16	UGGCUUCCGCAUGGUCUCGA	55483038	76
1384	chr16	UGGAUUCGAGAAAACCGCAG	55484106	76
1385	chr16	CGGUUUUCUCGAAUCCAUGA	55484124	76
1386	chr16	UGAAGUAUGGGAACGCCGAU	55485630	76
1387	chr16	GAAGUAUGGGAACGCCGAUG	55485631	76
1388	chr16	CCUCAGUGGUGGCCGACCAAG	55488657	76
1389	chr16	UACGACCGCGACAAGAAGUA	55488694	76
1390	chr16	GCUCCCGGAAAGAUUGAUG	55497015	76
1391	chr16	GCUUGGGGUACCCUCGCUCC	55498325	76

FIG. 48

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1392	chr11	GUACGAGCUGGAUACCCAAG	102838661	82
1393	chr11	AAUCCUACUGUUGCUGUGCG	102843515	81
1394	chr11	CACGGUUGGAGGGAAACCUA	102838642	80
1395	chr11	CUUGCAGAUACUUCAU AUG	102839149	80
1396	chr11	CAGGUUCCGUGGGUACCAGG	102840222	80
1397	chr11	UAUGCGGCAUCCACGCCUGA	102839165	79
1398	chr11	UCGAUUUUCUCACGGUUGG	102838631	77
1399	chr11	UGCUCAGCCUAUCCAUUGGA	102843487	77
1400	chr11	CGAUUUUUCUCACGGUUGGA	102838632	76
1401	chr11	AAUCAAUUCUGGGCUAUCAG	102838691	76
1402	chr11	GCGCAUCACCUCCAGAGUGU	102842775	76
1403	chr11	UCACGGUUGGAGGGAAACCU	102838641	75
1404	chr11	ACUUUUGGCCAAAUCCCUC	102839224	75
1405	chr11	GAGACAGGCAGAACCGAGUC	102840479	75
1406	chr11	UACCUAGAACAGGUUCAUGC	102843456	75
1407	chr11	GCAUCGAUUUUCUCACGGU	102838628	74
1408	chr11	CGCAUAUGAAGGUUACUAGCA	102839136	74
1409	chr11	UCUGGAGGGACAGGUUCCGU	102840212	74
1410	chr11	UGGGCAUCUCCAUAAAUCCC	102842209	73
1411	chr11	GAAGCUGGACUCCGACACUC	102842774	73
1412	chr11	UGCAGCUCCAUCCAAUGGAU	102843492	73
1413	chr11	UCACAGUUGGCUGGCGUCCC	102840188	72
1414	chr11	UUCUGGAGGGACAGGUUCCG	102840211	70
1415	chr11	GCAUGGGCCAAACAUUUC	102842245	69
1416	chr11	GUUCUGAAGUGACCAACAU	102842734	68
1417	chr11	CACCAGCAUGAACCUUGUUC	102843446	66

**FIG. 49**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1418	chr11	CGAUGAGGAUGAACGCUGGA	102524956	82
1419	chr11	AUGAUUGGCCUUUGCGCGAGG	102527528	79
1420	chr11	AUCAUGAUUGGCCUUUGCGCG	102527531	79
1421	chr11	AAUCGAUCCACUGUAAUAUG	102527642	79
1422	chr11	AGACUUACCGCAUAAUACAG	102527637	78
1423	chr11	GUUCAUGAGUUGCAGCAUAC	102523386	77
1424	chr11	AGUGGAUCGAUUAGUGUCAA	102527619	77
1425	chr11	GCAGUGAUGUAUCCAACCUA	102523310	76
1426	chr11	UGAUGGGCCAGGAAACACGC	102525025	76
1427	chr11	CAUCACUGCAUUAGGAUCAG	102523336	75
1428	chr11	UGGACCGGAUGGUAGCAGUCU	102524940	75
1429	chr11	AGGAGAUGCUCACUUCUGAUG	102524971	75
1430	chr11	CUGCAUUAUUUCUAUGACGC	102527860	72
1431	chr11	GGGACUCCUACCCAUUUGAU	102525041	71
1432	chr11	CUUCCAAAGUGGUACCCUAC	102527761	71
1433	chr11	GGGGACUCCUACCCAUUUGA	102525042	70
1434	chr11	UCUGCAUUAUUUCUAUGACG	102527859	69
1435	chr11	UCCUACCCAUUUGAUGGGCC	102525036	68
1436	chr11	AGUGAGUAUUCUGCAACAU	102527819	66

**FIG. 50**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1437	chr11	GAUGCUAUCACCACACUCCG	102716331	80
1438	chr11	GCAACCAGUAUCAGUCUACA	102722625	78
1439	chr11	UCAAUUGCUGGACGCCUGCU	102714630	77
1440	chr11	AGUUAGUGCGUUCCCACUUG	102722456	77
1441	chr11	AUGAUGAAAAAGCCUCGCUG	102722506	77
1442	chr11	UGUGACGGGAAGCCAAGU	102722540	77
1443	chr11	CAGCCUGUAUACCAGUUGGA	102715363	76
1444	chr11	UUGGUCCAGUAGGUUGGAUA	102716401	76
1445	chr11	CUCCCUAAGAUGACAUCGA	102718436	76
1446	chr11	GUAGACUGAUACUGGUUGCU	102722641	76
1447	chr11	AAGUAUUUCUCCACGGAGUG	102716333	75
1448	chr11	AUGCCAUCGAUGUCAUCUUG	102718445	75
1449	chr11	CUUGAUUAACUUACUUGCGG	102721404	75
1450	chr11	ACCCUAGAGAUACACCUGU	102722430	75
1451	chr11	GGUGGUUUUAUGUUAACCCC	102722470	75
1452	chr11	UAAGCAGCCUGUAUACCAGU	102715359	74
1453	chr11	AGCAUAGUUGGGAUACAUC	102718495	74
1454	chr11	CCCAAGGAUUAUCAAACUA	102714645	73
1455	chr11	AACGCACUAACUUGACCUAC	102722433	73
1456	chr11	GGUUAUCCCCAAAAGCAUAC	102713809	72
1457	chr11	GAU AUGCUUUUGGGAUACC	102713824	72
1458	chr11	AUCCCUUCCAACUGGUUAAC	102715354	72
1459	chr11	AAGUUAGUGCGUUCCCACUU	102722455	72
1460	chr11	UUUUGGGUUGAAUGUGACG	102722553	72
1461	chr11	UGCCAUCGAUGUCAUCUUGA	102718446	71
1462	chr11	CAAGUUAGUGCGUUCCCACU	102722454	71
1463	chr11	AAACAGCUGCGUCAAUUGC	102714619	70
1464	chr11	AUUGACAGGCAACCAAUACU	102714700	70
1465	chr11	CCGUUAGGUACUUCUGGAGA	102715427	70
1466	chr11	AAAUGUCAAACUGGGGUCAC	102716366	70
1467	chr11	CUUGGUCCAGUAGGUUGGAU	102716400	70
1468	chr11	GUAUCCCCAACUAUGCUUUCA	102718476	70
1469	chr11	GGCAUGAGCAAGGAAUCCAU	102721491	70
1470	chr11	GUUUACGUUACCUCCAGAAU	102714562	68
1471	chr11	GUCAAUUGCUGGACGCUGC	102714629	68
1472	chr11	UAACUUACUJUGCGGAGGUGU	102721410	68
1473	chr11	AGAUCAAAUGCGUAAUACU	102713429	67
1474	chr11	UUUCCUACUUACGUUCUUGC	102713759	66

FIG. 51

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1475	chr11	CAAGUUCAUGAGCAGCAACG	102776758	83
1476	chr11	GUACAAGCAGGUUAUCCAAG	102772957	80
1477	chr11	UCACAUCAUCUUGCGAAAGG	102776650	80
1478	chr11	UUGCAGCGGACAAUACUGG	102772851	78
1479	chr11	AUCUUGCGAAAGGCCGAACU	102776657	77
1480	chr11	AGUCAUCAGCUAUUAGCUA	102772074	76
1481	chr11	GAGCUGAAGUGACCAACGUC	102779566	74
1482	chr11	UCCAAGAGGCAUCCAUACCC	102772943	73
1483	chr11	AUUUUUGGCGAAGAUCCCAC	102775302	73
1484	chr11	GCUGGGCAUCUCAGAUCCCGA	102776369	73
1485	chr11	AAGUCAUCAGCUAUUAGUCU	102772073	72
1486	chr11	CACCAGGGGUUCCUCAGUAG	102776407	72
1487	chr11	CAUUCACAUCAUCUUGCGAA	102776647	72
1488	chr11	CCUUUCCUGGCAUGCCGAAG	102779532	71
1489	chr11	AAAGUACUACAACCUCGAAA	102779717	71
1490	chr11	CUCCAACAAGGAUCUUGCCC	102780491	71
1491	chr11	CUGCAUAUGAAGGUUACAGC	102775215	70
1492	chr11	UGCCUCUACUGAGGAACCCC	102776397	70
1493	chr11	UACCUUGGGCAAGAUCCUUGU	102780501	70
1494	chr11	UCAACCUUAGGCUCAACUCC	102772049	69
1495	chr11	UCUUUGCAGCGGACAAUAC	102772854	68
1496	chr11	UGGGUUUUUCCUCCAACCAUA	102772923	67
1497	chr11	CAUCUUGCGAAAGGCCGAAC	102776656	67

**FIG. 52**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1498	chr11	CACACCUGACAUGAACCGUG	102871923	81
1499	chr11	CUCUCUGCUGAUGACAUACG	102867930	80
1500	chr11	AAGUAGGUCCUAUAAAAACG	102865817	79
1501	chr11	AUGAUGCACGCACCUCGAUG	102872942	79
1502	chr11	AGUAGGUCCUAUAAAAACGU	102865818	78
1503	chr11	UCAGUGUUUUGGUGAUACGU	102863143	77
1504	chr11	UGUCAGGAUUUGGCAAGCGU	102867372	77
1505	chr11	CGUGGGCAUUCAGUCCCCUGUA	102867912	77
1506	chr11	UGGCCAUUCUAGUGAUCCAA	102868000	77
1507	chr11	AGGAUGUUGACUACGCAAUC	102871903	77
1508	chr11	ACAUCGGGGACUCCACAUCG	102872942	77
1509	chr11	UAAUCAGUUUGGGAUACCA	102864220	76
1510	chr11	GUGGGGAACAUUACGGCCUU	102867996	76
1511	chr11	CAUACACCAACGUUACCUGU	102872866	76
1512	chr11	UUUGAUGCUGUCACUACCGU	102867302	75
1513	chr11	UUGGUGGUUUUUGCCCGUGG	102871808	75
1514	chr11	UGCCAGGGGGGCCCGUAUGG	102872890	75
1515	chr11	CCCUGAAAUGAUGGACAUCG	102872928	75
1516	chr11	GCAACUGGACACAUCAUACCC	102872967	74
1517	chr11	GAGGUGCGUGCAUCAUCUCC	102872961	74
1518	chr11	GUAAUCAGUUUGGGAUACCA	102864219	73
1519	chr11	UUUUGAUGCUGUCACUACCG	102867303	73
1520	chr11	CGGCCUUUGGAUCACUAGAA	102868009	73
1521	chr11	ACUAGAAUGGCCAAGACCBA	102868022	73
1522	chr11	AUUUCGAUGAGGACGGAAUUC	102871615	73
1523	chr11	GGGUACACAUUACUCCAUACU	102871884	73
1524	chr11	AGGGGAUGCACUUUCGAUG	102871626	72
1525	chr11	UUGCGUAGUCAACAUCCUA	102871920	72
1526	chr11	ACAUCCUCACGGUUCAUUGUC	102871931	72
1527	chr11	AAAUGCCAGGGGGCCCGUA	102872893	72
1528	chr11	CCCGAUUCCUUGGAAGUUCU	102864197	71
1529	chr11	UUAUGGCCAACCUUGCCAUC	102866373	71
1530	chr11	CUCACUGCUGUUCACGAGAU	102868038	71
1531	chr11	CCCCGAUGGUCCAUCAUUCA	102872916	71
1532	chr11	AAGAAUUCACUCACCAUAC	102867910	70
1533	chr11	CCUGAACAGCUCUACAAGCC	102874867	70
1534	chr11	UUUUGGUGAUACGUUGGAGU	102863149	68
1535	chr11	CAAUUUUAGGCCGAUUCCU	102864187	68
1536	chr11	CACGAGAUUGGCCAUUCUU	102868026	68
1537	chr11	UCCCCGAUGGUCCAUCAUUUC	102872917	68
1538	chr11	GGCAAGCGUUGGUUCUCUUU	102867383	66
1539	chr11	GCAAGCGUUGGUUCUCUUUU	102867384	65
1540	chr11	CUAGCCCCAUGGUUUUUGGACC	102871661	65
1541	chr11	UGGUUUUUGCCCGUGGAGGU	102871804	65

FIG. 53

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1542	chr11	AAUUUUACCAGACUUCACGA	102954498	74
1543	chr11	GAUGCCAUUACCAGUCUCCG	102950139	73
1544	chr11	CUUAGGUCUUGACCACUCCA	102952113	72
1545	chr11	CUACCAUCCUACAAAUCUCG	102955459	71
1546	chr11	UUCUUCGGCUUAGAGGUGAC	102955377	70
1547	chr11	CUGGCAUGACGCGAACAAUA	102944305	70
1548	chr11	UGGACCAUUUAAGAGUUCGA	102955287	70
1549	chr11	UAGGAUGGUAGUAUGAUCUC	102955482	70
1550	chr11	UUCCCGCGAGAUUUGUAGGA	102955467	69
1551	chr11	UAUGCAGCAUCAAUACGGUU	102949079	69
1552	chr11	UACCAUCCUACAAAUCUCGC	102955458	69
1553	chr11	CCGAGUUACCAUAGAGAGAC	102952019	69
1554	chr11	GGCCAGAGGGCCCACUAAA	102954259	68
1555	chr11	UUCUCGGAGGCCUCUCAGUCA	102955420	68
1556	chr11	CUUGUUGCUGCGCAUGAGUU	102952142	68
1557	chr11	ACCUGGACAAGUAGUUCCAA	102954160	68
1558	chr11	AAAGUGGCCUUUUGCCGGUGU	102952071	68
1559	chr11	GCGACUUCUACCCAUUUGAU	102954258	67
1560	chr11	CCUCCUGGGCCAAAUUAUGG	102954208	67
1561	chr11	AGCAAUGCCAUCGUGAAGUC	102954502	67
1562	chr11	AUAUGCAGCAUCAAUACGGU	102949078	67
1563	chr11	ACAUCAGGAACCCCCGCAUCU	102955328	67
1564	chr11	AGGAUUCCCGCGAGAUUUGU	102955463	66
1565	chr11	UUUCCUAUCUACACCUACAC	102952073	66
1566	chr11	GAUACUCUACCUUCUGCAAAC	102955592	65
1567	chr11	CUUGGAGUGGUCAAGACCWA	102952125	65
1568	chr11	AAAUCUCGCGGGAAUCCUGA	102955447	65

**FIG. 54**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1569	chr11	ACUGCUUCUCGACAUCUCCG	19055436	80
1570	chr11	GUUCCUACCAGCCGACCA	19056276	79
1571	chr11	AGUGAAGGAUGCUUCCGUCA	19055449	78
1572	chr11	CGACCGCUGACAGGUGUCUG	19055975	76
1573	chr11	UUGAUUUCAUCACUGCAGCG	19055838	75
1574	chr11	GUAGGCACAGGUCAUCACAG	19056081	75
1575	chr11	CAGUGAAGGAUGCUUCCGUC	19055450	74
1576	chr11	GCUGAGGUGGAUCACAGUGA	19055464	74
1577	chr11	CUACUGCUGAGCAUCUUGGA	19055914	74
1578	chr11	CCGCCCCAGACACCUGUCAG	19055966	74
1579	chr11	AGACUGCUUCUCGACAUCUC	19055434	73
1580	chr11	UCAGCAUGCUCAGGCCUGCA	19056059	73
1581	chr11	GAGCAUGGAUCCAACCACCC	19056389	73
1582	chr11	GACUGCUUCUCGACAUCUCC	19055435	72
1583	chr11	ACAUCUCCGGGUGGCCUGA	19055447	72
1584	chr11	CCUAAUAUUAUGGAUCUGGA	19055654	72
1585	chr11	CGGCGGCAGCGAUACCAGAU	19055998	72

**FIG. 55**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1586	chr1	GACCCAAUCCGUUGACAG	115286238	80
1587	chr1	GAUCUGGACUUUCGAGGCUCGG	115286475	80
1588	chr1	UGGGUGCUAACAGCACACG	115286538	80
1589	chr1	CGAUAGCUGCACGCGUGGCG	115286606	79
1590	chr1	GCGAUAGCUGCACGCGUGGC	115286607	78
1591	chr1	AGCGGCGAUAGCUGCACGCG	115286611	78
1592	chr1	GCGGUUUUAUCCGGAUAGAUA	115286119	77
1593	chr1	CUUGACAAAGGUGUGAGUCG	115286186	77
1594	chr1	GCCCCGGCACCCGCUGUCAA	115286240	77
1595	chr1	ACACCGAGAAUUCGCCCGUG	115286407	77
1596	chr1	GGCGAUAGCUGCACGCGUGG	115286608	77
1597	chr1	UUUUCUGAUCGGCAUACAGG	115286746	77
1598	chr1	AUCUAUCCGGAUAAACCGCC	115286135	76
1599	chr1	CUUUGUCAAGGCGCUGACCA	115286161	76
1600	chr1	GUGUGACAGUGUCAGCGUGU	115286374	76
1601	chr1	UAAACAGCACACGGGGUGAA	115286545	76
1602	chr1	GCAGACCCGCAACAUUACUG	115286584	76
1603	chr1	ACACACAGGCCGUACUAUC	115286122	75
1604	chr1	CAUGGUACAGCGCCUUGACAA	115286174	75
1605	chr1	CCCGUUGACAGCGGGUGCCG	115286229	75
1606	chr1	GCAACAUUACUGUGGGACCCC	115286576	74
1607	chr1	AUGCUGAAGUUUAGUCCAGU	115286687	74
1608	chr1	UGUCAACGGGAUUUGGGUCC	115286254	73
1609	chr1	GCCGCUUUUUAAAACAGCCUG	115286572	73
1610	chr1	AGCUUUUCUGAUCGGCAUAC	115286749	73
1611	chr1	ACCCAAAUCCCGUUGACAGC	115286237	72
1612	chr1	GGGAUUUGGGUCCCGGCACU	115286261	72
1613	chr1	CAGGAUCUGGACUUUCGAGGU	115286478	72
1614	chr1	GUGCAGCUAUCGCCGUGCC	115286629	72
1615	chr1	GGGGCAUUGACUCAAAGCAC	115286210	71
1616	chr1	ACCCGCUGUCAACGGGAUUU	115286248	71
1617	chr1	AAUUCGCCCUUGUGGAAGAU	115286415	71
1618	chr1	UCGCCGCUUUUUAAAACAGCC	115286570	71
1619	chr1	UCCCGUUGACAGCGGGUGCC	115286230	70
1620	chr1	CCCCGGCACCCGCUGUCAAC	115286241	70
1621	chr1	GUGGUGCUGCCCCCUUCAAC	115286456	70
1622	chr1	AUCCCGUUGACAGCGGGUGC	115286231	69
1623	chr1	CACCCGCUGUCAACGGGAUU	115286247	69
1624	chr1	CUUCCACAGGGCGAAUUCU	115286398	69
1625	chr1	AGGCUGCCUGGCGGUUAUC	115286129	68
1626	chr1	UGCAGCUAUCGCCGUGCCG	115286630	68
1627	chr1	CGUGCAGCUAUCGCCGUGC	115286628	67
1628	chr1	CGCCGCUUUUUAAAAGCCU	115286571	65

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1629	chr17	UGAGACGACUGGGCGCUGCG	49506455	85
1630	chr17	GAGCAUGUCGGCGCCGUGCG	49506386	84
1631	chr17	UUGCAAGCAGGGCGCGAACG	49512873	83
1632	chr17	GUGUCCUCGCACACGGUGCA	49506591	82
1633	chr17	CAAGCAGAACACCGUGUGCG	49506527	81
1634	chr17	AGACCUAUAGCCAGCACGG	49510523	81
1635	chr17	UCGCUGGAUGCGGCGCAGGG	49512938	81
1636	chr17	AGCUUCUCAACGGCUCUGCG	49512788	80
1637	chr17	ACGGUCUGGUUGGUUCCACA	49502167	79
1638	chr17	AGGCGUGACGUUCUCCGACG	49506314	79
1639	chr17	CAAGCCGUGCACCGAGUGCG	49506356	79
1640	chr17	GCGUCGUCGGCCUCCACGCA	49506387	79
1641	chr17	CAUGUCGGCGCCGUGCGUGG	49506389	79
1642	chr17	CGAGGCGUGCCGCGUGUGCG	49506473	79
1643	chr17	CGACGGCACGUUUCCGACG	49506557	79
1644	chr17	UCCCCAGCCCGUGGUGACCCG	49510575	79
1645	chr17	GACAGGGAUGAGGUUGUGCG	49510588	79
1646	chr17	GCUUCUCAACGGCUCUGCGG	49512789	79
1647	chr17	CCCCGGUUGGGCUCACACCG	49513004	79
1648	chr17	GGCGCGGCCGUCCAUGGCG	49495443	78
1649	chr17	CAGCAGGCGCGGCCGUCCA	49495448	78
1650	chr17	GCCACCGGCCGCCAUGGA	49495447	78
1651	chr17	CUCGGUCGCGCUCACCACGU	49506316	78
1652	chr17	AGCCCCACGCACUCGGUGCA	49506348	78
1653	chr17	AGCCGUGCACCGAGUGCGUG	49506358	78
1654	chr17	GCCGUAGGCGCAGCGGCACA	49506409	78
1655	chr17	CUGCGCCUACGGCUACUACC	49506431	78
1656	chr17	CGUCGGAAUACGUGCCGUCG	49506542	78
1657	chr17	CUCGUCGGAAUACGUGCCGU	49506544	78
1658	chr17	GCGCUCGGUGUCCUCGCACA	49506598	78
1659	chr17	CGAGGUCGGCUCGCUGGAUG	49512948	78
1660	chr17	CAUCCAGCGAGCCGACCUCG	49512963	78
1661	chr17	GGUGCACGGCUUGCACGGCU	49506334	77
1662	chr17	AAGCCGUGCACCGAGUGCGU	49506357	77
1663	chr17	GUCGGGGCACUCCUCGCACA	49506526	77
1664	chr17	GUGUGGACCGUGUAUCCAA	49510427	77
1665	chr17	GCUUGCAAGCAGGGCGCGAA	49512875	77
1666	chr17	GGGUGGCCAGCUUGCAAGC	49512885	77
1667	chr17	GGGCUCACACACGGUCUGGU	49502177	76
1668	chr17	GGUAGUAGCCGUAGGCGCAG	49506416	76
1669	chr17	GUGUGCGAGGAGUGCCCCGA	49506540	76
1670	chr17	ACCGUGUAUCCAACGGCCA	49510421	76
1671	chr17	GCAGCCCUCAACGGUGACGG	49512721	76
1672	chr17	GGUGGCCAGCUUGCAAGCA	49512884	76
1673	chr17	GGCUCGCUGGAUGCGGCGCA	49512941	76
1674	chr17	UCUCCACGAGGUGCGGCUCGC	49512954	76
1675	chr17	CCGAGCCGCCUCGCACACG	49506470	75
1676	chr17	GGCGUGCCGCGUGUGCGAGG	49506476	75

FIG. 57

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1677	chr12	AUCACGGCGGAAACGGUACG	5494630	86
1678	chr12	UGGCGAACAGAACAUACCGG	5494617	85
1679	chr12	UCAUCGGCCAUCGACAUUCG	5494718	83
1680	chr12	AGAGACGCUACAACUCACCG	5494518	82
1681	chr12	GCGGAGCAUAAGAGUCACCG	5494649	82
1682	chr12	GAGACGCUACAACUCACCGC	5494519	81
1683	chr12	CACAUACCGAGUACUCCCCU	5494654	81
1684	chr12	GGAGCAUAAGAGUCACCGAG	5494651	81
1685	chr12	CGGAGCAUAAGAGUCACCGA	5494650	80
1686	chr12	UGAAGUCAGUGCUCGGACGU	5494884	80
1687	chr12	UCGGUGUCCAUUGCAUACAC	5494469	79
1688	chr12	CCCGCCCUUUGUAUCUCAUGG	5494573	79
1689	chr12	AUGUUCUGUUCGCCACACG	5494597	79
1690	chr12	GGAUUACGUGGGCAGCCCCG	5494594	79
1691	chr12	GAAGCCAGGCCGGUAAAAAA	5494820	78
1692	chr12	GCCGGUCAAAAACGGUUGCA	5494828	78
1693	chr12	ACAAUAAACUCGUGGGCUGG	5494923	78
1694	chr12	GUAUCUCAUGGAGGAUACG	5494582	77
1695	chr12	AUAUUUCUCGCUUAUCUCG	5494253	76
1696	chr12	GUAAUCCUCCAUGAGAUACA	5494566	76
1697	chr12	UUACGUGGGCAGCCCCGUGG	5494597	76
1698	chr12	GUGGGUGACCGACAAGUCAU	5494702	76
1699	chr12	CUGGUGUCCCCGAAUGUCGA	5494713	76
1700	chr12	CAUCGACAUUCGGGGACACC	5494726	76
1701	chr12	UUUUUAUGAAACGCGAUGUA	5494798	76
1702	chr12	GCAACCGUUUUUGACCGGCC	5494812	76
1703	chr12	GGGGUAUUGAUGAUAAACAC	5494848	76
1704	chr12	CAGUGCUCGGACGUAGGUUU	5494878	76
1705	chr12	UCAGUGCUCCGACGUAGGUU	5494879	76
1706	chr12	UCUCGACAAGGCACACACAC	5494953	76
1707	chr12	AUUCCAGCCGGUGAUUGCAA	5494474	75
1708	chr12	GCCCCCGCCCUUUGUAUCUCA	5494570	75
1709	chr12	CUCCAUGAGAUACAAGGGCG	5494560	74
1710	chr12	GAGUCACCGAGGGGAGUACU	5494660	74
1711	chr12	GUCGAUGGCCGAUGACUUGU	5494698	74
1712	chr12	CCGGUCAAAAACGGUUGCAG	5494829	74
1713	chr12	UCAGAGAACAAUAAACUCGU	5494916	74
1714	chr12	UGAUGUUCUGUUCGCCACCA	5494599	73
1715	chr12	CCCCUGCAACCGUUUUUGAC	5494817	73
1716	chr12	UCCAUGAGAUACAAGGGCGG	5494559	72
1717	chr12	AGUCAUCGGCCAUCGACAUU	5494716	72
1718	chr12	GGCCGGUCAAAAACGGUUGC	5494827	72
1719	chr12	GCUUAUCUCCGUGGCCAUCCA	5494262	71
1720	chr12	UGUCCAUUGCAAUCACCGGC	5494465	71
1721	chr12	GGACACCAGGUACACGGUGCU	5494739	71
1722	chr12	UCUUCCGAUUUUUCUCGACA	5494965	71
1723	chr12	UACCGUGGCAAAAGUAACCA	5432328	70
1724	chr12	ACAGAACAUACACGGCGGAAA	5494623	70

FIG. 58

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1725	chr19	GCGCCCACGCAAGUCCACAG	49061667	84
1726	chr19	CGGACCGCUGUGGACUUGCG	49061659	82
1727	chr19	CUUCUUUGAAACCCGUGCA	49061576	79
1728	chr19	CCUAGACAGGACUACUCGGG	49061865	79
1729	chr19	UGUGGACUUGCGUGGGCGCG	49061651	78
1730	chr19	GGACCGCUGUGGACUUGCGU	49061658	78
1731	chr19	AAACUGCACCGAGCGAGUCGU	49061730	78
1732	chr19	CAGUUUCGCUCACCCCCACGC	49061759	78
1733	chr19	CUGCAAGGCUGUAACGCUG	49061561	76
1734	chr19	CACGCCAAGGUCCACAGCGGUC	49061672	76
1735	chr19	UGGGACUCAAUUGGCACACU	49061941	76
1736	chr19	ACUGCACCAGCGAGUCGUCG	49061728	75
1737	chr19	UUGGGACUCAAUUGGCACAC	49061940	75
1738	chr19	AGCUCACCCCGACGACUCGC	49061734	74
1739	chr19	UUUCAAAGAAGUACUGGCAGG	49061600	73
1740	chr19	UCAAAGAAGUACUGGCAGGAG	49061602	73
1741	chr19	CGGCCAACCGCAGCCGGCGU	49061760	72
1742	chr19	GUGCCCCGGCCAACCGCAGC	49061766	72
1743	chr19	AGCGUUUAUCAGCCUUGCGAGC	49061577	71
1744	chr19	AGCGGGUUUCAAAGAAGUAC	49061594	71
1745	chr19	AGCGAGUCGUCGGGUGAGC	49061720	70
1746	chr19	AACUGCACCGAGCGAGUCGUC	49061729	70

**FIG. 59**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1747	chr1	UGGUAGCCAGGUCGCUGCG	156879222	85
1748	chr1	CCGUCCGCCUCGCAGCAUCG	156860935	84
1749	chr1	CUUUGAGUAUAUGCAGCACG	156876548	82
1750	chr1	GCAAUCGACUGCAUCACGCA	156881474	82
1751	chr1	GGCGCCGCCGCGAUGCUGCG	156860940	81
1752	chr1	ACGGAGACCACUCUUACGA	156864735	81
1753	chr1	CGGCAGCCAUGAGACCGUG	156873803	81
1754	chr1	GCCCACUAGACAGUUGCGUG	156879276	81
1755	chr1	GUACCAGCUCUCCAACACGG	156880153	81
1756	chr1	CAGGAAGAACGUGACGUGCU	156871699	80
1757	chr1	UGAUGGCGUAGACCUCUGGU	156881512	80
1758	chr1	GAGAGACUCCAGAGCGUUGA	156866923	79
1759	chr1	CCCUUUCGAGUUCAACCCCG	156873942	79
1760	chr1	CUUCUUCGGCGUCUGCACCG	156876506	79
1761	chr1	UCACCACCGAGAGCGACGUG	156880080	79
1762	chr1	GGCAAUCGACUGCAUCACGC	156881473	79
1763	chr1	UCUAGCAGCCGCAACUGCA	156842147	78
1764	chr1	GCCGCCGCGAUGCUGCGAGG	156860943	78
1765	chr1	GAAGAGUGGUCUCCGUUCG	156864754	78
1766	chr1	CCAGGUGCCAAUGCCUCGG	156868541	78
1767	chr1	GCGCAGACACCCGUGCCGCA	156873806	78
1768	chr1	GUUGCCGUUGUUGACGUGGG	156873839	78
1769	chr1	CCGGGACAUCGUGCUCAAGU	156876116	78
1770	chr1	UCGGCAGGACUUCCAGCGUG	156876443	78
1771	chr1	CGGUACAGGAUGCUCUCGGG	156880040	78
1772	chr1	AUGAUGGCGUAGACCUCUGG	156881513	78
1773	chr1	AGGUUUUCUACCAUCACCG	156868642	77
1774	chr1	UUACGGUACAGGAUGCUCUC	156880043	77
1775	chr1	GCUCCACACGUCGCUCUCGG	156880071	77
1776	chr1	GUACCAGGGCUGCUUGCCGU	156880122	77
1777	chr1	CAGAGGUCUACGCCAUCAUG	156881529	77
1778	chr1	CGCAGUCCCCGAGGAGCCGUG	156861063	76
1779	chr1	UCGCAGUCCCGAGGAGCCGU	156861064	76
1780	chr1	CCUCGGGACUGCGAUGCACC	156861085	76
1781	chr1	CCCAGCACCAUCGUGAAAGAG	156864740	76
1782	chr1	ACUCACAGGCGACUGAGCCG	156864788	76
1783	chr1	GGGCACCUGGACCUCAGCG	156868516	76
1784	chr1	GUCGUCCCCCACAUCCACCG	156868543	76
1785	chr1	GAAGCUGGUCUCAUUGAGCA	156873746	76
1786	chr1	CAACGGCAACUACACCGCUGC	156873864	76
1787	chr1	GCAGUCUCCUUCUCGCCGGU	156874396	76
1788	chr1	CGGUGGUGAACUUACGGUAC	156880054	76
1789	chr1	AGUUGCGGGCUGCUAGAUCU	156842165	75
1790	chr1	GACUGCGAUGCACCCGGGAU	156861091	75
1791	chr1	CGGACACUCACAGCUCAGUC	156861140	75
1792	chr1	AUCUGGAGCUCCGUGAUCUG	156864400	75
1793	chr1	AGGCAUCUGGCGCCACGAAA	156864754	75
1794	chr1	GGCACCUUGGACCUUCAGCGU	156868515	75

FIG. 60

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1795	chr9	GUCGCUGCACCAGAUCCGAG	84670872	83
1796	chr9	AUUACGUACAAUGCACAGUG	84724348	83
1797	chr9	GCUUCAGUGGUUCUUAUACG	84727759	81
1798	chr9	AUCAUCAUUGCUGUAACGG	84867254	81
1799	chr9	ACGCAAGGACUUCCACCGUG	84948505	81
1800	chr9	ACCAAUACACAGGAGUACCA	84727826	80
1801	chr9	GCCCCAGAGCCGCGCCAUGG	84670782	79
1802	chr9	ACCACUGAAGCGCUGGUUUG	84727735	79
1803	chr9	GGUUUUAUACAGUGACGUCUG	84745038	79
1804	chr9	CAAGUUCUAUGGCGUCUGCG	84948565	79
1805	chr9	AUCGCGGUGGCACGAAGUGCU	84955404	79
1806	chr9	GUCAGUGCUGUACACGUCCC	84955491	79
1807	chr9	CAUUGUGUGGCCACCGACCU	85020206	79
1808	chr9	ACCCCAAACCAGCGCUUCAG	84727746	78
1809	chr9	GCCGUGGUACUCCGUGUGAU	84727815	78
1810	chr9	AACUGCAGCGAAUGACAUCG	84744997	78
1811	chr9	CACCGAGAGAUGUUCCGAC	84745059	78
1812	chr9	ACGUCACUGAUAAAACCGGU	84745056	78
1813	chr9	UGUGGUGGUGAUUGCUGUCUG	84752011	78
1814	chr9	GUUCUAUGGCGUCUGCGUGG	84948568	78
1815	chr9	GACUGCGUCAGUUCCGUGGG	84955328	78
1816	chr9	CUGCGACUGCGUCAGUUCCG	84955332	78
1817	chr9	GGCUGAGGGCAACCCGCCA	84955327	78
1818	chr9	UAUAGCCCAGCAGAUCGCCG	84955375	78
1819	chr9	CAUGUACAGGAAAUUCACGA	85020273	78
1820	chr9	GUACCAAGCUGUCAAACAAUG	85020360	78
1821	chr9	GGUGAUAGAGUGUAUCACUC	85021268	78
1822	chr9	GCACUGCAUUJUGCAGGACGU	84670850	77
1823	chr9	CGACCCUUUCUCCUGGCAUCG	84670903	77
1824	chr9	CGUCACUGAUAAAACCGGUC	84745057	77
1825	chr9	CGCCAGGUAGACCAUGCCCG	84955380	77
1826	chr9	GUACAGCACUGACUACUACA	84955513	77
1827	chr9	GCAUUGUGUGGCCACCGACC	85020207	77
1828	chr9	UCACGACGGAAAGCGACGUC	85020287	77
1829	chr9	AAGGUGGCAUGGACCCGCCA	84670780	76
1830	chr9	CAUGCCCGCGCGAUCUGCU	84955368	76
1831	chr9	AGUCAGUGCUGUACACGUCC	84955492	76
1832	chr9	ACCUCCUGGGGGCACGUGCG	85021296	76
1833	chr9	GCGACCCCGCACGUGCCCCC	85021304	76
1834	chr9	GGCAUGGACCCGCCAUGGCG	84670785	75
1835	chr9	CAUCGUGGCAUUUCCGAGAU	84670918	75
1836	chr9	GUAUUGGGAUGUUGGUAAACC	84723690	75
1837	chr9	GCGCUUCAGUGGUUCUAUAA	84727757	75
1838	chr9	AACCGGUCGGGAACAUUCUCU	84745069	75
1839	chr9	ACUGCGUCAGUUCCGUGGGC	84955327	75
1840	chr9	UGCGACUGCGUCAGUUCCGU	84955331	75
1841	chr9	CCAGCAGAUCGCCGCCGGCA	84955381	75
1842	chr9	AAUGCAGUGGCCUCUCGGAU	84670875	74

FIG. 61

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1843	chr2	GCCUGGCCAGGAGGCUAACUA	237877264	79
1844	chr2	UUCCAUCCGCAGGAGCUACA	237911533	78
1845	chr2	AGUAGCGGCCAUGCACUGCC	237911620	78
1846	chr2	CUGGCAGAGCAAGCGCACUG	237911767	78
1847	chr2	GCGCACUGAGGGCAUUGUGU	237911779	78
1848	chr2	ACCGUAGUUAGCCUCCUGGC	237877253	77
1849	chr2	UGGCAGAGCAAGCGCACUGA	237911768	77
1850	chr2	GGGCACCGUAGUUAGCCUCC	237877257	75
1851	chr2	ACACCACAGCGUCUCCCCGA	237877325	75
1852	chr2	AGGCCGUCGGGAGACGCUG	237877334	74
1853	chr2	GAGACGCUGUGGUGUGACUG	237877345	74
1854	chr2	CGACUGCACCUUCCACAUUGG	237911563	74
1855	chr2	GUGAUGGGGACCACGAUGAA	237911702	74
1856	chr2	GCGCCGCGGGAGGCAGCACA	237859691	73
1857	chr2	AGUGGUCAUGAAGAGGGUGAU	237877229	73
1858	chr2	AGGUUCUUCCUGGCAGUGCA	237911624	73
1859	chr2	GGGUAGAGGAUGCUGCCGGG	237911681	73

**FIG. 62**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1860	chr2	GUCACCCGGUCAUAGGAAGG	165991323	80
1861	chr2	AAAGAGCUGCAUGCCGACCA	166038003	80
1862	chr2	CGUCAUUGGAUAGUGCUCCA	166041284	80
1863	chr2	GUGUCGUCCCCGGCACAAACA	166043887	80
1864	chr2	CUGAGUCAUUAGUCGAAACA	166047671	80
1865	chr2	CCGGGUGACAAGGCCAUUG	165991297	79
1866	chr2	CAGCUUUAGAGGGCGAGCAA	166043970	79
1867	chr2	UUUGUCACCCGGUCAUAGGA	165991320	78
1868	chr2	GCGUGGGAGUUGACAAUCAC	166037946	78
1869	chr2	UGGCGAGUCCAAGUUCUACC	166039478	78
1870	chr2	AGAACUUGGACUCGCCAUG	166039460	77
1871	chr2	UGGCUUUGUCACCCGGUCAU	165991316	76
1872	chr2	UUAACACCACAACUGGUGAC	166002609	76
1873	chr2	AAAGACGAUUAAGACGAUGU	166012236	76
1874	chr2	UUGUGUUCCGCGUGCUGUGU	166037877	76
1875	chr2	AACUGAAGGUCCACCAACCA	166043752	76
1876	chr2	UCCCAGUGCAACUCAGUCA	165991810	75
1877	chr2	GGGUGGCUUACUGUUGAGAA	165992075	75
1878	chr2	GGUACAGUCACAGUAAGACU	166036147	75
1879	chr2	CAU GGAGCACUAUCCAAUGA	166041271	75
1880	chr2	GACCCUAAUAAGUUAACCC	165992034	74
1881	chr2	GUCAAACCUGUCACCAGUUG	166002615	74
1882	chr2	UGUAGAAGAACAGCCGUAG	166015654	74
1883	chr2	UUAAGUGGGUACAUACCACU	166037777	74
1884	chr2	ACGCUUAGCCUGGUAGAACU	166039474	74
1885	chr2	UCAGUUCCUACAUCCGCCUGU	166043723	74
1886	chr2	CAGUAGGUCAUCCCGGAUGC	166043820	74
1887	chr2	ACGGCUCUCGUUAUCCUCAA	166043917	74
1888	chr2	CUUGUUUCGACUAUAGACUC	166047658	74
1889	chr2	CUGAAAUCGUCUUCAAUGCU	166052878	74
1890	chr2	CAAUGAGUAACCCUCCUGAU	166056430	74
1891	chr2	UGGUAGUCACAUUUUCACUC	165994301	73
1892	chr2	AGCUGUCCAACAGGGCGAUGU	166043729	73
1893	chr2	UGGCAGUGUUUCCAGCGAAU	166043800	73
1894	chr2	AUGACCUACUGGUCUGACUC	166043844	73
1895	chr2	UGCGUCUCUCUCCGUGUGUCGU	166043874	73
1896	chr2	CAUUUUGUCACGCAUCAAUC	165994268	72
1897	chr2	CAGAUUGAUGCGUGACAAAA	165994281	72
1898	chr2	CUUUAGUAGUGAAUCGGAU	166036067	72
1899	chr2	CAGCAUCCGGGAUGACCUAC	166043833	72
1900	chr2	UUGUUUGUGCCCCGACGACA	166043873	72
1901	chr2	CGUCUCUCUCCGUGUGUCG	166043876	72
1902	chr2	UAGUCUUUGUUGAGCAUCCG	166044035	72
1903	chr2	UGUCAAUCGGUUCCCCUCAA	166045090	72
1904	chr2	ACUCUUGGAACUACUUAG	166045243	72
1905	chr2	GGAUGCUCUACGAAUACAGA	165991612	71
1906	chr2	UCCGAAUAGUUGAACAUAC	166013797	71
1907	chr2	UGGUACAGUCACAGUAAGAC	166036146	71

FIG. 63

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1908	chr2	UUUGGUACACUAUCAUACG	165389729	83
1909	chr2	UUGGUACACUAUCAUACGA	165389728	80
1910	chr2	CAGGUUCAGAGGUCGAGCAA	165323235	79
1911	chr2	AUCCAUCCAUCCCUUAAACG	165377603	79
1912	chr2	CUCUUGUUUGCGUUUCAACG	165389504	79
1913	chr2	AGAGAUGUUUGAUGUAAGCG	165374845	78
1914	chr2	GUCACACUAUCAUACGGAGGG	165389725	78
1915	chr2	UUGAGUCAUGAGACGAAAUA	165313717	77
1916	chr2	GGAGCCACGGAUGCUUCAGUA	165323162	77
1917	chr2	GGUAGAAGGGCCCCGACCA	165323453	77
1918	chr2	UAUGGAGCACUAUCCCAUGA	165331528	77
1919	chr2	AUGAUUGUGAACUCCCACGC	165344756	77
1920	chr2	GAUAUUGGAGCUCCCGCCGA	165365199	77
1921	chr2	GGUAGCGUCUGUAAGCCCUC	165389547	77
1922	chr2	CCAGUCUGGAGGGUUACUCA	165307906	76
1923	chr2	CUCUCCAAGACGCAACAGUA	165323202	76
1924	chr2	ACUGCUGAACUGCUCCGUCA	165331530	76
1925	chr2	AGUUCUCCGAUCAUUCCGGC	165342465	76
1926	chr2	AUCAUACGAGGGUGGGAGACG	165389717	76
1927	chr2	CAGUCAGUGCUGGUACCGCC	165295847	75
1928	chr2	UGUAGGCCUGAAGACCAUUG	165310335	75
1929	chr2	CUGCUGAACUGCUCCGUCAU	165331529	75
1930	chr2	CCAUGAGUAACCCUCCAGAC	165307918	74
1931	chr2	CUAAGCGUGUUUGCGCUAAU	165310417	74
1932	chr2	UAGUCCUUACUGAGCAUCCG	165323170	74
1933	chr2	GCACGAACAGAGAGUCUU	165323307	74
1934	chr2	UGGUGUGGUUCUCCUGGUCG	165323454	74
1935	chr2	UUGAGAGCAAUCAAACUGCC	165374898	74
1936	chr2	CGUUUCAACGUGGUUCGUAAU	165389494	74
1937	chr2	GCGUUGUCAAUAGCAGCAA	165295890	73
1938	chr2	GAGACCCAAACAGGAACGCA	165295945	73
1939	chr2	ACGGGAUCCAUGGAAUUGGU	165308767	73
1940	chr2	AAAUGACAGAGGUCCGAAAUA	165315661	73
1941	chr2	CUCGCCUACUGUUGCGUCU	165323194	73
1942	chr2	UCUCUCCAAGACGCAACAGU	165323201	73
1943	chr2	CAUAGCGCUGUGGACUGCAA	165323434	73
1944	chr2	CUGCUAAAGUGGGUUGCAUA	165370239	73
1945	chr2	GCUCUUCCAUCUGUAAUCGA	165389433	73
1946	chr2	AAUUUACCAGCCGGAAUGAU	165342459	72
1947	chr2	GCGUGGGAGUUCACAAUCAU	165344741	72
1948	chr2	GCAUACUCACAUACAACAAUC	165370292	72
1949	chr2	AACUUUGUAACGUAGGACU	165374939	72
1950	chr2	CACGUUGAACGCAAACAAG	165389517	72
1951	chr2	GUAGGCCUGAAGACCAUUGU	165310336	71
1952	chr2	GGGUAGAAGGGCCCCCGACC	165323454	71
1953	chr2	AUUCAGCGAGCAGUCAGAUA	165354652	71
1954	chr2	UGUCUUCAGAUUCCGAUUUU	165315661	70
1955	chr2	GCAUGGUGGGCAUUUCUGUC	165331347	70

FIG. 64

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
1956	chr2	CCGUUGUCAUGUUACACAG	165097400	81
1957	chr2	AUGGUGGAAACGGGAUGACCA	165092414	80
1958	chr2	CCUAGACAUUACGUGCUGCU	165154646	80
1959	chr2	CUCUUGGCAAGCAAGCUCGG	165097317	79
1960	chr2	UCAAAAGCAGAAUCGCUUGG	165162677	79
1961	chr2	GACCAAGCUCCAUAACUG	165131305	78
1962	chr2	GUUGAUCCGGGACAAAACUA	165092392	77
1963	chr2	AGUCAUGGGGUAGUGCUCCA	165137932	77
1964	chr2	AACCGGGAUAGGCUCUUAG	165100332	76
1965	chr2	CUAGGGUCAUGUAUUUGCCC	165092409	75
1966	chr2	GUGCUCAACAAUACUGUAGC	165113887	75
1967	chr2	GAUGUUGUUCUACCCCGAGA	165115519	75
1968	chr2	CAGUUUCAGAGGUAGGGCAA	165140919	75
1969	chr2	CAGUCUCUCUUGAGUAUCCG	165140984	75
1970	chr2	UCAUGAGUCGAAAUAGAGAC	165155811	75
1971	chr2	UCAAGACUCUGAAAGUUUCGA	165163849	75
1972	chr2	GACCCUGACACAAUUCACCC	165090957	74
1973	chr2	AAUGAACACUAGGUUGAUCC	165092380	74
1974	chr2	CUAUCUUGCACUGCUUCAAG	165097256	74
1975	chr2	CCUUCUCAGUAGUGGUGCAU	165113820	74
1976	chr2	CAAUUGGCACUGUGACGGUG	165127725	74
1977	chr2	UCCAGUACCUACACCACUGG	165127810	74
1978	chr2	UGUACUGCGAUCAUUCAGAC	165131248	74
1979	chr2	UUACGUUUUCUUCGUGAUCCA	165164429	74
1980	chr2	GAUGGUGGAAACGGGAUGACC	165092415	73
1981	chr2	AGAGCCUUUAUCCGGUUUGA	165100314	73
1982	chr2	CCAAUGCACCACUACUGAGA	165113832	73
1983	chr2	UGUUUUGCUAUUGCGUCUUG	165140962	73
1984	chr2	UUCAAAAGCAGAAUCGCUUG	165162676	73
1985	chr2	UAACUCCACUAACCCUGUU	165170464	73
1986	chr2	CUGCCAGGGUGAAUUGUGUC	165090966	72
1987	chr2	UCUUACUGCUGGGCGAGGUA	165094385	72
1988	chr2	ACCCGAAGAAGACCUUAAAC	165115478	72
1989	chr2	ACGCAACAGUAACGUUAGUC	165140793	72
1990	chr2	GACCUCUAAGAGCCUUAUCC	165100322	71
1991	chr2	UCGGUGCCAUCAAUCAUUA	165100361	71
1992	chr2	AGCCUCAGUUUAUGGAGCU	165131295	71
1993	chr2	GACAGAAAUGUCCGCCAUGC	165138085	71
1994	chr2	UCAGCUCUAACGUCACCUAC	165140672	71
1995	chr2	CUCAUUUGGACCUUACCAAC	165155763	71
1996	chr2	ACCCAAAACACGCUUUGUAA	165090584	70
1997	chr2	ACUACUACUUCACUUAUAGGC	165092299	70
1998	chr2	GGUUGAUCCGGGACAAAACU	165092391	70
1999	chr2	CAGAAAGCUUCUUUACCGAC	165162795	70
2000	chr2	CUUACUGCUGGGCGAGGUAU	165094386	69
2001	chr2	CCGGAAGCUUGUUUACUGA	165115459	69
2002	chr2	ACCACCAGUGGUGUAGGUAC	165127799	69
2003	chr2	CAUUUGAAGACAGCGAAAGC	165140854	69

**FIG. 65**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2004	chr17	GCGGAUCACACGGAACAGCG	63941946	85
2005	chr17	UAUGGAACUGCUGCGCCCCG	63961351	85
2006	chr17	GUGUUGGUGUCGUUGAACGG	63968201	85
2007	chr17	GGGACGCCGGACCCACUAUG	63940869	84
2008	chr17	AGGGCACUGUCCGAUUCGA	63947057	82
2009	chr17	CGUCUUCCGCAUCCUGUGCG	63957262	82
2010	chr17	CAUGGU AUGCCAACGACACG	63968092	82
2011	chr17	GUUGGUGUCGUUGAACGGCG	63968203	82
2012	chr17	UGUCCUGCGGCUGAUCCCG	63941894	81
2013	chr17	GAUGAACAUUGCUGAACAGCG	63972224	81
2014	chr17	CGUGUCCUGCGGCUGAUCCG	63941896	80
2015	chr17	GAGGGCACUGUCCGAUUCG	63947058	80
2016	chr17	GACGAGUUCCCCAUCAUAGAG	63949511	80
2017	chr17	GCCCAACUUGAUGCGCCC	63951798	80
2018	chr17	CAGCCUGGACACAUCGCAAG	63963737	80
2019	chr17	UCAGGGCUGAAGACGAUCGU	63968342	80
2020	chr17	CUUCAGCGUAGUCAGGCGCG	63972379	80
2021	chr17	CCCGGCCUGACUACGCUGAA	63972393	80
2022	chr17	CUACGGAGACCCCCCGCCGG	63972624	80
2023	chr17	GAUCUACGGAGACCCCCCGC	63972627	80
2024	chr17	CCUGUUCGAGAUCACCACGU	63941681	79
2025	chr17	CACGCCCAAUCCGCGCCAGG	63941927	79
2026	chr17	GAAGUGGUGGACUCUGCGCA	63948693	79
2027	chr17	AGCUGACUACAAGCCCCCG	63949464	79
2028	chr17	CGACCUGGAGAUGCCCCACCG	63951467	79
2029	chr17	UCGGACUCCUCGGAGGCGAU	63951499	79
2030	chr17	GCGCGGCAGGUUGCAGUCCA	63957332	79
2031	chr17	GCUUUUGUGUACCAGACGGA	63959270	79
2032	chr17	CUAGGCCUGGCCAACGUACA	63959298	79
2033	chr17	CCCUACGAGUAUUUCCAGCA	63959367	79
2034	chr17	GAUCGUCAUGGACCCGUUCG	63961302	79
2035	chr17	AUGAGUGCAUCAAGACCGGG	63966209	79
2036	chr17	CCAAUCAAAGGUAUCGUUGG	63968062	79
2037	chr17	CGUGUCAUUGCUGUACCACG	63968179	79
2038	chr17	GUCAGGGCUGAAGACGAUCG	63968343	79
2039	chr17	CUCAUCAAGAUACUGGCCCG	63971799	79
2040	chr17	GGGAUGCCGAUGACCUCCGG	63972623	79
2041	chr17	GAUGCCGAUGACCUCCGCG	63972625	79
2042	chr17	AUGUACGCCACAGCCACGA	63941008	78
2043	chr17	AGAUCCAGAGGGCCUACCGC	63941067	78
2044	chr17	UUGGGCUUGGCAAUCCUCAG	63941329	78
2045	chr17	CGGAUCACACGGAACAGCGU	63941947	78
2046	chr17	CGAAGGCCUGCUUCGUCACG	63943065	78
2047	chr17	CCUAUCCCACUAUACUUAGG	63943848	78
2048	chr17	UGAGAGGUUCGACAUUCUCG	63945478	78
2049	chr17	GAUGUCGAACCUCUCAGAGG	63945497	78
2050	chr17	GUCCCGAUUCGAGGGCAUGA	63947049	78
2051	chr17	CAUCUCGCCAUCCUCAUCCG	63951837	78

FIG. 66

SEQ ID NO.	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2052	chr3	GUGCAUCUCAGGCCGACUGG	38579381	83
2053	chr3	AGUUCGCACCUGGAUCACUG	38609742	82
2054	chr3	AGUCGGCGAGAACUUCACUG	38550379	81
2055	chr3	GAUGAGGCCUCUCGCUAG	38550545	81
2056	chr3	UCGACAUACUUGGUCCAGGG	38622417	81
2057	chr3	AGCGAUAAACCUCCAGGUGCG	38550402	80
2058	chr3	CCGCACCUGGAGGUUAUCGC	38550416	80
2059	chr3	ACUCCGGCGCAAGCACGAAG	38550670	80
2060	chr3	UUCGUGCUUGCGCCGGAGUG	38550686	80
2061	chr3	ACUUGUAUACCCACCCACGA	38554282	80
2062	chr3	GUGGUGUCCACCGCACAGCA	38575429	80
2063	chr3	UGGGCGAGACGGUUCCAGCA	38597933	80
2064	chr3	AUCUACACACGGAGGCCUGGG	38599005	80
2065	chr3	CUUCCACCCCACUCCGGAGAG	38630338	80
2066	chr3	CCAACAGCAAUUCCACUCCG	38551157	79
2067	chr3	CUGUGGAUGGUCCAUCAAGCA	38597870	79
2068	chr3	AUGCUCCCGCGGCUGGAACG	38604033	79
2069	chr3	CGUGGUGCGCAAUUCCGCUAUG	38606042	79
2070	chr3	UGGCUGCAGGACAUGUCCGG	38606802	79
2071	chr3	CGGCACCAACGGCUCCGUGG	38609787	79
2072	chr3	CCAGGCCGAGGCUCACACGA	38550326	78
2073	chr3	CACCUUCCGAGGUUAUCGCUUG	38550419	78
2074	chr3	UGGGGACCGCAUCCAUUGCA	38550823	78
2075	chr3	CGCAUCCUCAGACUGAUCCG	38551461	78
2076	chr3	GCGAGACGGUUCCAGCAUUG	38597936	78
2077	chr3	ACUCUUCCGCCUGAUGACGC	38606700	78
2078	chr3	UUUAGGCACCGGUAGCCCUC	38606798	78
2079	chr3	CAACGGCACCAACGGCUCCG	38609790	78
2080	chr3	AUGGCGGUGAAGGUUACCU	38620971	78
2081	chr3	CACUCGACAUACUUGGUCCA	38622414	78
2082	chr3	AGGCUCACACGAUGGACUCA	38550334	77
2083	chr3	GAUGACUCCGAAAGAGCGUCG	38551514	77
2084	chr3	CGGAGAAUUGUACUGGACCA	38560205	77
2085	chr3	UCCCAAACUUCCCCGCAAAG	38560325	77
2086	chr3	UGGGCCCCAUCAAGUCACUG	38562474	77
2087	chr3	CUUCGAGGACAUCUACCUAG	38566563	77
2088	chr3	CAAGACCUGCUACCACACG	38575361	77
2089	chr3	GUGCUCCACGAUGGUUAGC	38575368	77
2090	chr3	CAUUACCUACCGCACCCUG	38579345	77
2091	chr3	CCCUACUACUACUCCAAAC	38587502	77
2092	chr3	UCCGUGUGUAGAUGGUUCG	38598983	77
2093	chr3	UCCUCGAAGCCCAUCUACACA	38598994	77
2094	chr3	CACUCAGACCACGCCAUCGG	38599040	77
2095	chr3	GCUAGAGCACCCGCCAGACA	38603716	77
2096	chr3	GGAGGGCUACCGGUGGCCUA	38606784	77
2097	chr3	CGCAACUUCACAGCGCUCAA	38609807	77
2098	chr3	CUCUGCCAUGCGCUUCUCGA	38633235	77
2099	chr3	UGUAGUCAGACCCCCCGCACC	38550403	76

FIG. 67

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2100	chr12	UGUCUUUCGAGUGUUGUGCG	51765927	84
2101	chr12	CGUGAAGCGAACACGCACGG	51721835	83
2102	chr12	CUCCUGCUUGCGACGCAGUG	51807154	83
2103	chr12	UCUGUCCUUCCGCCCCGUAGG	51745975	82
2104	chr12	GCAGAUGGAAGAGCGGUUCG	51807104	82
2105	chr12	CUGAGUCUAGCAGACGUGGA	51762643	81
2106	chr12	GUACCCGUACAGUCAAGUUG	51769235	81
2107	chr12	GUCCUGCAGCGUGCCUACCG	51807204	81
2108	chr12	GAACAGCUGCAGUCCGAUCA	51699665	80
2109	chr12	CGCGGAUGACGAGCACAGCA	51721676	80
2110	chr12	CAGCUACCGAUGACAGUGUA	51721942	80
2111	chr12	AUUCCGGUGGAUGUCUGCAC	51769114	80
2112	chr12	GGGUACCCGUACAGUCAAGU	51769237	80
2113	chr12	ACCUAGUUCCGAGUAGGCCA	51780682	80
2114	chr12	GAGUUUGCCUUCAUCUACG	51663023	79
2115	chr12	GAUUCUGUCCUUCCGCCCGU	51745978	79
2116	chr12	GGUACCCGUACAGUCAAGUU	51769236	79
2117	chr12	GUGCUGCCAGGUACAACAU	51770565	79
2118	chr12	UGCAGCUGUAGAUUCCCGGA	51788744	79
2119	chr12	ACCGGGGACAUUUGGCAAGG	51807220	79
2120	chr12	GGGAUGUCCCCGUAGAUGAA	51663018	78
2121	chr12	UCCUGACUGGUCGAAGAAUG	51686451	78
2122	chr12	CAGGAGACGCCGCCGAUGU	51721885	78
2123	chr12	ACCAAUUAGCCUCCUACGGG	51745978	78
2124	chr12	AUJAGCCCUACGGGCGGA	51745982	78
2125	chr12	GGCCCACUUUAAGCAGCGUG	51769040	78
2126	chr12	GCCUGGGCUACUCGGAACU	51780693	78
2127	chr12	GCCUAGAUAAAGGAACACCCA	51806668	78
2128	chr12	CAAUGAACUGGGUGGCAUCG	51806879	78
2129	chr12	AACCACACUGCGUCGCAAGC	51807164	78
2130	chr12	GUGCAAAGGAAAGACGUAC	51706557	77
2131	chr12	GUAGCUGCUCCGGCGCUCGC	51721735	77
2132	chr12	UGUAGCUGCUCCGGCGCUCG	51721736	77
2133	chr12	GGCGUGGUGUCCCUCaucgg	51721866	77
2134	chr12	GUAGGAGGCUAUUUGGUCCA	51745960	77
2135	chr12	ACUGAGUCUAGCAGACGUGG	51762642	77
2136	chr12	GUGUGUCUGCAAGAUCAACC	51765852	77
2137	chr12	AGUCAUGCAUAUGCCAGCGA	51765873	77
2138	chr12	UCAGUGAUCCGUAUCAAGAA	51768978	77
2139	chr12	AACUGUAUCGCCAAUCACAC	51769107	77
2140	chr12	GGUGCAGACAUCCACCGGAA	51769128	77
2141	chr12	AGCAGCGAGUCGGAUCCUGA	51769320	77
2142	chr12	GAGCAGGGUACGAAUCCUU	51806392	77
2143	chr12	AGCCUAGAUAAAGAACACCC	51806667	77
2144	chr12	UGACUGCCAUCCGGCUUUGG	51662931	76
2145	chr12	GUCCUCCCGAUGACUGCCAU	51662941	76
2146	chr12	ACAUUCUUCGACCAGUCAGG	51686437	76
2147	chr12	UCUGUUGUCUGGCGAGCCGAA	51706668	76

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2148	chr2	AGGAAACCGUGACCCAUUCUG	166228834	80
2149	chr2	GGCUGAGCGUCCAUCAACCA	166284536	80
2150	chr2	AUAGGCAGCACAUAGAAAAG	166286359	80
2151	chr2	UAUGCCCUUCGACACCAAGG	166286391	80
2152	chr2	CGUGUGUAGUCAGUGUCCAG	166293364	80
2153	chr2	CGAAUCCUACGUCAAGUCAA	166199767	79
2154	chr2	AAAGCCAAUUCUCGACCAG	166204364	79
2155	chr2	GUGUCCGAAGGGAUUUUAUG	166233407	79
2156	chr2	GUCGUUGUGAAUGCACUCAU	166228955	78
2157	chr2	UUGGGUCAUUAGCCUAAACA	166293275	78
2158	chr2	CCACCAAUGCUGCCGGUGAA	166284585	77
2159	chr2	CCGUUCACCGGGCAGCAUUGG	166284597	77
2160	chr2	GUUCACCGGCAGCAUUGGUG	166284599	77
2161	chr2	CUUAUAGACGUUACCGCUUA	166198926	76
2162	chr2	GCCAAGUUAACAUAGAGUCA	166242615	76
2163	chr2	UGACAGUGCCAAUUGCACCU	166272473	76
2164	chr2	GGGCUGAGCGUCCAUCAACCC	166284535	76
2165	chr2	GAGGUUGUCUACCCCCAAUC	166286340	76
2166	chr2	UCACUUUUUCUUCGUGACCCG	166305829	76
2167	chr2	CCAUGAAUAAACCCACCGGAC	166306529	76
2168	chr2	CCAGUCCGGUGGGUUUAUC	166306541	76
2169	chr2	CAGCAAUGCGUUGUUCAUAG	166311689	76
2170	chr2	AUGUUAGUCAAAAUUGUGCGA	166228756	75
2171	chr2	AAAAGCUCUCAGUGUCCGAA	166233396	75
2172	chr2	CUGUGCAUUUUUCCGUUCAC	166284585	75
2173	chr2	CGUUACCCGGCAGCAUUGGU	166284598	75
2174	chr2	UGUUACUGCUGCGUCGCUCC	166284640	75
2175	chr2	GUUACUGCUGCGUCGCUCCU	166284641	75
2176	chr2	UUACUGCUGCGUCGCUCCUG	166284642	75
2177	chr2	ACCUUGGUGUCGAAGGGCAU	166286377	75
2178	chr2	AGUGUAUUAAACACCACAGAU	166228834	74
2179	chr2	UUAAAGCUCUCAGUGUCCGA	166233395	74
2180	chr2	AGUUCUGCGAUCAUUCAGAC	166278144	74
2181	chr2	GUUUCCACCUUGGUGUCGAA	166286383	74
2182	chr2	UCGACAUUUUUGGUCCAGUC	166306527	74
2183	chr2	UUGCUCCUUUGACUAGACGU	166199774	73
2184	chr2	AAAAGCCAAUUCUCGACCA	166204365	73
2185	chr2	GCAAAACAAUUCGGAACGAUU	166228799	73
2186	chr2	UAUGAAUUCGAUGAGCCAG	166251785	73
2187	chr2	AGUUUCCACCUUGGUGUCGAA	166286384	73
2188	chr2	GCCUAUGCCCUUCGACACCA	166286388	73
2189	chr2	AGCUCGUGUAGCCAUAUCA	166293325	73
2190	chr2	UCGUGACCCGUGGAACUGGC	166305819	73
2191	chr2	ACAUUUUUGGUCCAGUCCGG	166306530	73
2192	chr2	ACUACUACUUCACUGUAGGA	166204000	72
2193	chr2	CUAGGUCAAAUAUACAUCCU	166204212	72
2194	chr2	UCUGGCUCAUCCGAAUUCAU	166251799	72
2195	chr2	UUUUCCAAAUCGGAUUCCCC	166272468	72

FIG. 69

SEQ ID NO	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2196	chr3	GUGACACUCUCAUAGGACGG	38697463	83
2197	chr3	UAGUUUCGAGGGAUCCAAUG	38793997	83
2198	chr3	UUCGAACCCUUCGCGCUCUG	38713999	82
2199	chr3	UCAUCGCUAUUCGACUGUG	38726716	82
2200	chr3	UGUAGUCACCAUGGCGUAUG	38756765	82
2201	chr3	GUGACUCCGGAGUAAAGCGA	38793967	82
2202	chr3	CAAGACUUUGCUACCGUAUCG	38722322	81
2203	chr3	GCACUUACCCUCAAGGACGG	38750082	81
2204	chr3	CUAUGUGCUGCACCGCUCCA	38697611	80
2205	chr3	GAAUGUCCUAGGAGAACCG	38697776	80
2206	chr3	GCACAGCAAUAGAACUCCGU	38763571	80
2207	chr3	GGCACAGCAAUAGAACUCCG	38763572	80
2208	chr3	GGGGCAAGCUCACUAGUGGG	38752255	79
2209	chr3	ACUAGCCCAGCGUUUUCCAG	38752491	79
2210	chr3	AGGUAAAAGGUGAUCCAUUG	38755912	79
2211	chr3	GCUCCCAGCAGAACUGAUCG	38793787	79
2212	chr3	AGGCUGGAGUGUUCUCACUA	38697349	78
2213	chr3	UGACACUCUCAUAGGACGGU	38697464	78
2214	chr3	GUGCAGCACAUAGCUCCGAU	38697633	78
2215	chr3	UUCUUGCUUCCAUUCGGAGAG	38697684	78
2216	chr3	AGACUGAUCCGAGCGGCCAA	38698450	78
2217	chr3	UGCAGCUGUUGAUUCCCCGGG	38710848	78
2218	chr3	GGAUACCACCAAGAGUCCAU	38722364	78
2219	chr3	CAUCGCUAUCCGACUGUGU	38726715	78
2220	chr3	CACCAUCACCUUJUGUGCAUCG	38742373	78
2221	chr3	UCUCAACAGGCAUUCGAUGC	38750176	78
2222	chr3	GGAGACCACGAAAGCCAUCG	38752375	78
2223	chr3	UGGUUGUAAGGAUCAGAGCG	38755815	78
2224	chr3	GGCCCGAGUGGCACUAAACC	38792129	78
2225	chr3	UCCGUUUCUACAGCACACACC	38793745	78
2226	chr3	GUAUGGAGCUGAUUGCCCCU	38697360	77
2227	chr3	GGCAAUCAGCUCCAUCUGG	38697378	77
2228	chr3	AGGUGCAUCAACUUAUACCGA	38712308	77
2229	chr3	CAGUGGCCGCAGAGCGCGAA	38714005	77
2230	chr3	CAGAGCGCGAAGGGUUCGAA	38714014	77
2231	chr3	CAGCGGGGCCAGUCUUCAUG	38728710	77
2232	chr3	UGUUACGGUAGUUUUCCCU	38728753	77
2233	chr3	GGAGCAUGGCUUCGAAAGGUA	38742322	77
2234	chr3	AUCAUAACCUCCGUCCUUGA	38750077	77
2235	chr3	GCAAGCUCACUAGUGGGCGG	38752258	77
2236	chr3	GAGUGAGUAUCUCGGCCAG	38752443	77
2237	chr3	GCUCCCCGAUCAGUUUCUGCU	38793796	77
2238	chr3	GGGAGCUCACCAUAGAACUU	38793816	77
2239	chr3	CAACUCCGUCGCUUUACUC	38793961	77
2240	chr3	GGUCAACAUUGCAACCCAAAGU	38709599	76
2241	chr3	UCACAAUCGACAAAGGUACAA	38712288	76
2242	chr3	CGAGAUGCUGCUUAAGUGGG	38718720	76
2243	chr3	GGUUGGCAAGCAGCUCCUAG	38728756	76

FIG. 70

SEQ ID NO.	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2244	chr3	UCCGGACUCUACGAGCACUG	38872234	81
2245	chr3	GGUAUUCCGCAUCCUCUGCG	38896951	81
2246	chr3	UUCACCGCUAGGGUACUCGG	38846988	80
2247	chr3	CCAAUCAUGCUGACGCCA	38894646	80
2248	chr3	UUUACGGCACUGGCACAUGG	38896996	80
2249	chr3	AGAGCCACCGAGUACCCUAG	38846996	79
2250	chr3	UUCGGCCAUCCGAGCCAAG	38847670	79
2251	chr3	ACAAUUCUGAAGAGCGUCGG	38847693	79
2252	chr3	CAAUUCUGAAGAGCGUCGGA	38847694	79
2253	chr3	UUACCACUUGCAGCAGAGCG	38871458	79
2254	chr3	CAGUGCUCGUAGAGUCCGGA	38872249	79
2255	chr3	AGUAGAGCCCAGUAGUACGC	38909187	79
2256	chr3	GUAGAGCCCAGUAGUACGCA	38909188	79
2257	chr3	GAAGAGCUGCUGACCUACCA	38921160	79
2258	chr3	AACACACGGAAGGUACGCAG	38925464	79
2259	chr3	UGAAGAUCGCCUCCACUGCA	38847025	78
2260	chr3	GCGACAUCAUCAGAGCAAAG	38847607	78
2261	chr3	GAUGCGGAAUACCACUAGGA	38896973	78
2262	chr3	ACUGCGUGUUCAUGGCUACA	38945457	78
2263	chr3	AUUGUCCGCUUGGCUCGGAU	38847663	77
2264	chr3	UGUUUACGGCACUGGCACAU	38896998	77
2265	chr3	UJGGGUCAUCAGCCCGAACAA	38910110	77
2266	chr3	CUUCUUCACAGAGCGUAGCA	38921229	77
2267	chr3	GAACACACGGAAGGUACGC	38925463	77
2268	chr3	GGUUCAUACAACUUUCUUGAG	38846919	76
2269	chr3	CGGACAAUUCUGAAGAGCGU	38847690	76
2270	chr3	GUGGUGGUCAAUGCUCUCAU	38871691	76
2271	chr3	AACUGGGACAGCGCACGAAG	38872223	76
2272	chr3	GCCUCAGUGCUCGUAGAGUC	38872245	76
2273	chr3	AGGUUCCACCAAAUGACCCA	38883335	76
2274	chr3	CUGCUUUCCAUGCUGUAGCG	38883360	76
2275	chr3	GACUUGGUUGGCACCCACUUG	38894612	76
2276	chr3	UGGCAUCCAAGUACCUUCGUG	38904041	76
2277	chr3	GGGGGCAACAGUUCCACACG	38904040	76
2278	chr3	GGAAUGUCGCCAUAGAGCUU	38950171	76
2279	chr3	GAAGCGGAUUGCCAUCAAA	38950262	76
2280	chr3	UUUGGUUUUUGCGACACGCAA	38847099	75
2281	chr3	UUUGGUAGCAGGUUUUCCGC	38883315	75
2282	chr3	CUCUUGUCCACGCUACAGCA	38883365	75
2283	chr3	GACCAUACAGGAUCCCCGAA	38886129	75
2284	chr3	CCUAGGAAAAUUACCUUUCG	38886128	75
2285	chr3	AGCACUGGAUCGGAUUCCGCC	38894849	75
2286	chr3	GUUUACGGCACUGGCACAUG	38896997	75
2287	chr3	GCCGUAAACAUGAGACUGUC	38897023	75
2288	chr3	GCCUCGGCGAAAGUAGUGGU	38900013	75
2289	chr3	UCUUAACGCACAGCCACUGG	38904022	75
2290	chr3	UGAACCUUGAAAUGCAUCUCG	38921115	75
2291	chr3	UCAGGUCUGAAGGUCAUCGU	38921242	75

FIG. 71

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2292	chr7	GUACGACACCGACCAGAUCA	97732731	76
2293	chr7	UUCUUCUGCAAACAGCUGAG	97732658	73
2294	chr7	AUCUUCUGCAGAGAAUCGCC	97733765	73
2295	chr7	UUAAUGGGCAAACGGGAUGC	97733815	73
2296	chr7	ACCAUAAGAGCCUUUAACA	97734277	73
2297	chr7	CUUUGGAUUAUGGGCAAAC	97733808	72
2298	chr7	GCCCUGUUAAAGGCUCUUUA	97734288	71
2299	chr7	AAAAGACUGCCAAGGCCACG	97732626	70
2300	chr7	UCUCUGCAGAAGAUGCUAA	97733744	70
2301	chr7	AUCUGAAUUACUGGUCCGAC	97732709	69
2302	chr7	UGUUUUUCAGUGGCUUAUGAA	97739882	69
2303	chr7	GGCACGAGGAUUUCAUGU	97732613	68
2304	chr7	CUGGUCGCUGUCGUACCAGU	97732712	68
2305	chr7	GAAGGGGCCUCACCUUGAUC	97732731	68
2306	chr7	GGGCUCCGGCAGUUCCUCCU	97733723	68
2307	chr7	CCAAUGAUGAUCUGAAUUC	97732700	67
2308	chr7	GGCCACUUGUUUUCAUUG	97734256	66

**FIG. 72**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2309	chr12	UUACCCUAUUCUGCUCUCGG	57012391	83
2310	chr12	CUGCUCUCGGGGAUACUUG	57012401	79
2311	chr12	UAUCCCCCGAGAGCAGAAUA	57012382	76
2312	chr12	GCAAUCCCUCCCAGAGAUGAG	57013622	75
2313	chr12	GUAGAGAUCUGGAUCCCUCU	57013671	75
2314	chr12	UCACAUCCGUAGGAGAGUCU	57012452	74
2315	chr12	CAGCCUAGCUCAGAGCUUUG	57015742	74
2316	chr12	GUAUCCCCGAGAGCAGAAU	57012383	73
2317	chr12	CUUUACCCUAGACUCUCCUA	57012446	73
2318	chr12	UUCUCUUGAUUCACAUCCGU	57012442	73
2319	chr12	AGGGGAGACUUACCUUGCUG	57015688	73
2320	chr12	CUUACCCUAUUCUGCUCUCG	57012390	72
2321	chr12	UGAGCCAGGCUAGCACAGGU	57013578	72
2322	chr12	AAAAGCCACUCAUCUCUGGA	57013615	72
2323	chr12	GAGCUUUGGGGCUGUCUGUA	57015730	72
2324	chr12	UUGCUCUCCCAGGCACCAUG	57015797	72
2325	chr12	AGAGGAGCGUCCAGGCCAGGU	57012822	71

**FIG. 73**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2326	chr2	UCAGCGCCGGCGACUAUGAG	75049662	83
2327	chr2	CUGGGUCUGGAGAUACCGGG	75049638	82
2328	chr2	AAUGGAUACGUCCUCCGG	75198918	82
2329	chr2	UUGCUCGUGGUAGCGGUCAG	75053640	81
2330	chr2	GUGGUUGAGUAGUAGCCCUG	75120663	81
2331	chr2	CAUCAGCGCCGGCGACUAUG	75049664	80
2332	chr2	UAGUCGCCGGCGCUGAUGAA	75049681	79
2333	chr2	UGGCUAUGCAUACACCGUAG	75053687	79
2334	chr2	CACGAACUGAUUGGGUUCGG	75198874	79
2335	chr2	AUCAGCGCCGGCGACUAUGA	75049663	78
2336	chr2	UCGCUCUCCAGGUUCCGUCU	75049717	78
2337	chr2	CCGUAGUGGGAAUCACACUA	75053673	78
2338	chr2	CCAUGAGUGAUUCCCCACUA	75053685	78
2339	chr2	GAGUCGUGUGCAUGAUCGAA	75120611	78
2340	chr2	UGGUUGAGUAGUAGCCCUGG	75120664	78
2341	chr2	GGCCGUAGUACCAUUCGUUG	75198648	78
2342	chr2	GGUGAACCUUGGCCUUCGCGG	75198705	78
2343	chr2	CCGUGUAGGCAGCUGCCCAA	75198831	78
2344	chr2	AGAGGUCUGAGUCCACCGGG	75198918	78
2345	chr2	CUUGAAGCCCAGACGGAACCC	75049722	77
2346	chr2	GCUUGGCAGAGACUUCGUUG	75053627	77
2347	chr2	AUCAUGCACACGACUCUGCU	75120630	77
2348	chr2	GGUCACCACAAUGACCGUGU	75198817	77
2349	chr2	CGUAGUGGGAAUCACACUAU	75053672	76
2350	chr2	CUGGCGAAGACAGCGGCGAU	75198596	76
2351	chr2	GGAGUCCCCGGGAUCUCAC	75053661	75
2352	chr2	UAUCUACUCCAUGACGGCUG	75198561	75
2353	chr2	CCUAUGCUGUCCACAACGAA	75198646	75
2354	chr2	AGCCAUGGAGGCCUCCGCGA	75198706	75
2355	chr2	UUCCAGCCCCCUAUAGUCGC	75049668	74
2356	chr2	UCUACUGCUGCCUCAAUGAC	75051255	74
2357	chr2	GGCUAUGCAUACACCGUAGU	75053686	74
2358	chr2	GGCUGGUUGCACGAACUGAU	75198865	74
2359	chr2	GCUGGUUGCACGAACUGAUU	75198866	74
2360	chr2	AGUCGCCGGCGCUGAUGAAG	75049682	73
2361	chr2	UGUGUACAAAGUCAGCCGCC	75049598	72
2362	chr2	CGCCGGCGACUAUGAGGGGC	75049658	72
2363	chr2	UGCUGCCCCUUCAUCAGCGC	75049675	72
2364	chr2	CUCGCUCUCCAGGUUCCGUC	75049718	72
2365	chr2	GGGAUCCUCACCUUGUCAUUG	75051257	72
2366	chr2	GGUAAAUGAUGAUUGUCG	75051428	72
2367	chr2	AUGUAUGAUGGCCAUGUACC	75120767	72
2368	chr2	AUCAAAGGCCACAGCCGUCA	75198565	72
2369	chr2	AGCUGCCUACACGGUCAUUG	75198810	72
2370	chr2	UGGGCUUCAAGCAUGCCUUC	75049698	71
2371	chr2	CUGUGGUUGAGUAGUAGGCC	75120661	71
2372	chr2	AGCCGUCAUGGAGUAGAUAC	75198577	71
2373	chr2	GUCCACAACGAAUGGUACUA	75198638	71

FIG. 74

SEQ ID NO.	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2374	chr10	CAUCUACUUCCUGCCGCU CG	69409034	80
2375	chr10	GCUACAAACAUCACC CGCGAG	69409033	80
2376	chr10	AGAUGAGGGCGAUCACCACG	69409064	80
2377	chr10	GUGACACCUGUUGACUCUG	69404984	79
2378	chr10	GUGCCC GGACAUCAGGC GCA	69408961	79
2379	chr10	GGGUGCCACCAAGUGCGUGG	69414987	79
2380	chr10	CCAGGCCACCACG CACU UGG	69414994	79
2381	chr10	GGAUCAUCCUGGCCAUCGG	69416140	79
2382	chr10	GACGAUGGCCAUUACCCGUCA	69416179	79
2383	chr10	GGCGUCCCCAGGAUGGAUCA	69404885	78
2384	chr10	CCCCUCCGAGGCUACCAGUG	69404914	78
2385	chr10	UGAGGGCGAUCACCACGAGG	69409067	78
2386	chr10	GCCUGGGCCCAGAACAGCGG	69414965	77
2387	chr10	GACUGUGCGCAUCCUCCGAU	69416140	77
2388	chr10	AGUCAACAGGUGUCACACUA	69404965	76
2389	chr10	GCAAUAACCGCCUUGGUGCU	69415091	76
2390	chr10	ACCAUAGCCCUGAUCCAUC	69404890	75
2391	chr10	CCGGAAUCCAGAGCGAAACC	69405083	75
2392	chr10	CGGAAUCCAGAGCGAAACCU	69405084	75
2393	chr10	GCACCGUGCGCCUGAUGUCC	69408970	75
2394	chr10	UCAUCGGCCUCACG CUCUGG	69408990	75
2395	chr10	CGUGAGGCCGAUGACGCUGU	69409011	75
2396	chr10	UCCACCGUCACCAUGGACCA	69415007	75
2397	chr10	UGACCGCCAUUGCUGCCGAC	69415936	75
2398	chr10	AUGCCCGUGGUGUUGCUCUC	69416273	75
2399	chr10	UCGAGCUUAUCUUCUUGGU	69405030	74
2400	chr10	GGCACCGUGCGCCUGAUGUC	69408969	74
2401	chr10	CUGCGCGCCUCCAGAGCGUG	69408995	74
2402	chr10	UUGCCUGUCGGCAGCAA UGG	69415945	74
2403	chr10	UGACUGUGCGCAUCCUCCGA	69416139	74
2404	chr10	GCCCCCUCCGAGGCUACCAG	69404916	73
2405	chr10	CGCUCCCCCAGGUUU CGCUC	69405078	73
2406	chr10	GAUGCCAGCAAUAACCGCCU	69415084	73
2407	chr10	GGGCGUCCCCAGGAUGGAUC	69404886	72
2408	chr10	CCCCACUGGUAGCCUCGGAG	69404926	72
2409	chr10	ACCUGUUGACUCUCGUGGAG	69404989	72
2410	chr10	CCACGCACUUGGUGGCACCC	69415002	72
2411	chr10	ACCCUGGUCCAUGGUGACGG	69415018	72
2412	chr10	CAACUACUUCAUCGUCAUC	69416103	72
2413	chr10	UGCGCAUCCUCCGAUGGGCC	69416145	72
2414	chr10	CCCCCUCCGAGGCUACCAGU	69404915	71
2415	chr10	CAGCUCGAGCUUAUCUUCU	69405026	71
2416	chr10	GCGCGCAGUGCCCGGACAUC	69408968	71
2417	chr10	UUUGUAGCCUACAGCGUCAU	69409006	71
2418	chr10	UCCCCCAGAGGCCUUACAGG	69414945	71
2419	chr10	GCCCCCGCUGUCUUUCGGGCC	69414976	71
2420	chr10	UCCCCACUGGUAGCCUCGG	69404925	70
2421	chr10	CUCCACCGUCACCAUGGACC	69415008	70

FIG. 75

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2422	chr4	UCUACGCGCUUCAUAGCGAG	103719228	82
2423	chr4	CAGAGCGGAUGCGCCAGGA	103719430	82
2424	chr4	GGGGUCAAACACGACUGUCA	103589852	80
2425	chr4	GCAGAAACCUGGAUAGACGG	103719640	80
2426	chr4	UAUGUACACCGUGACCAGAA	103589866	79
2427	chr4	UUGCCGGUUUGGAUGAAACC	103589906	79
2428	chr4	CUACUCCAUGACGGCCAUG	103719140	79
2429	chr4	CCGGUUUGGAUGAAACCUGG	103589909	78
2430	chr4	GGACGGCUGCACGAACUGGU	103719447	78
2431	chr4	CGCCCCGGCAGCUAGCGAGG	103719597	78
2432	chr4	CUGCUAUGAUACUUGUCAC	103656229	77
2433	chr4	AUUGACCAACGUGUUGAAGG	103719267	77
2434	chr4	GCGCUCUGGUCCCUGGCGUA	103719403	77
2435	chr4	ACAGGCAGUCCCAGCGCGGA	103719508	77
2436	chr4	CAGGCAGUCCCAGCGCGGAA	103719509	77
2437	chr4	ACAAAGCAGAGAGUACGGCC	103658265	76
2438	chr4	AGUACGGCCUGGCAUGACUU	103658276	76
2439	chr4	CUGGCGAACACAGCUGUGAU	103719178	76
2440	chr4	UUCCCGCGCUGGGACUGCCUG	103719494	76
2441	chr4	UGGUGGGUGUCUGCAUCGUUG	103589833	75
2442	chr4	GAGCUCUAGCUCAUCAUAGC	103589936	75
2443	chr4	UGACGGCCAUGCGGGUGGAC	103719132	75
2444	chr4	CUCCAUGACGGCCAUGCGG	103719137	75
2445	chr4	UCCUCACCUGUCCACCGCAA	103719138	75
2446	chr4	GCGCAUCGCGCUCUGGUCCC	103719410	75
2447	chr4	CACCACCAACCAUACGCCA	103719405	75
2448	chr4	ACCAGUUCGUGCAGCCGUCC	103719432	75
2449	chr4	GGACCAGAGCGCGAUGC GCC	103719426	75
2450	chr4	GCCAGGACGGCUGCACGAAC	103719443	75
2451	chr4	CUGAACUGCUGGACCAAGC	103719535	75
2452	chr4	CCACGGGGGCAGUUGAGACU	103719561	75
2453	chr4	GAAACCUGGAUAGACGGGGG	103719637	75
2454	chr4	CAAGUAUCAUGAGCAGCUAA	103656210	74
2455	chr4	GACAGUCCUCAUGCGCUUGU	103719336	74
2456	chr4	AGGCAGUCCCAGCGCGGAAG	103719510	74
2457	chr4	GGCCGCCCGGCAGCUAGCG	103719594	74
2458	chr4	CCAACGAUGCAGACACCACC	103589819	73
2459	chr4	CUGGUGGUGUCUGCAUCGUU	103589832	73
2460	chr4	UACAUACUGCUUUGCCGGUU	103589895	73
2461	chr4	GUCCACCGCAAUGGCCGUCA	103719147	73
2462	chr4	UCCUGGCCACAAGCGCAUG	103719330	73
2463	chr4	UGACAGUCCUCAUGCGCUUG	103719335	73
2464	chr4	CGGUGUACAUACUGCUUUGC	103589890	72
2465	chr4	UACUUGUCACAGGUACUCC	103656239	72
2466	chr4	CAUGGCCGCCUCAACACGU	103719260	72
2467	chr4	GCUGCACGACUGGUUGGUG	103719452	72
2468	chr4	GACCGCCUUCGCUAGCUGCC	103719587	72
2469	chr4	UGACCGCCUUCGCUAGCUGCC	103719588	72

FIG. 76

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2470	chrX	GUAGACGAACCGGAUGUCAG	47585241	84
2471	chrX	GAAUUGCAGAAGGCCGUCUG	47583511	83
2472	chrX	GCGCGGCCUAGCAACCACGA	47585396	82
2473	chrX	CAGGUCCCACAACCGCAGCG	47585311	81
2474	chrX	GGCGCGGCCUAGCAACCACG	47585395	81
2475	chrX	UCAUCAGGGCCAAGUUUCGUG	47584953	80
2476	chrX	GGUUGCAGGCCGCGCCCCA	47585377	80
2477	chrX	CUCAUAACGCUGGUUAAGG	47584983	79
2478	chrX	CAAUGAGAAACUCCUCGCGUG	47585311	79
2479	chrX	AAUUGCAGAAGGCCGUCUGU	47583510	78
2480	chrX	UCCGCAGACACUCUCCAUGG	47585268	78
2481	chrX	AUCAGAGCCUCGCCCGUCG	47585398	78
2482	chrX	GAUCUCAUAACGCUGGUUA	47584986	77
2483	chrX	CGCGGCCUAGCAACCACGAG	47585397	77
2484	chrX	CGGGCCUAGCAACCACGAGG	47585398	77
2485	chrX	ACCGAGGUCCGGAAUJUGCAGA	47583521	76
2486	chrX	GUAUCCGCAGACACUCUCCA	47585271	76
2487	chrX	AGUCAUCAGGGCCAAGUUUCG	47584951	75
2488	chrX	UCAUCUUGAUUCUCAUAACGC	47584993	75
2489	chrX	UGGCGGGGGUGUAGACGAAC	47585251	75
2490	chrX	GGGUGAGGACUCACCGAGGU	47583533	74
2491	chrX	UAGGGGAUGCCGCUGACAUC	47585244	74
2492	chrX	CGCAGACACUCUCCAUGGCG	47585266	74
2493	chrX	UUCACCAAGACCUACACUGU	47585646	74
2494	chrX	UCAGGCCUAUCUGGGACCGCA	47586674	74
2495	chrX	UUCUGGUGUCCCCACGAACU	47584950	73
2496	chrX	GUCAUCAGGGCCAAGUUUCGU	47584952	73
2497	chrX	UGGUUGCUAGGCCGCGCCCC	47585378	73
2498	chrX	GUUCGUCUACACCCCGCCA	47585266	72
2499	chrX	GUGUCUGCGGAUACUUCCAC	47585292	72
2500	chrX	CUCGCCCUUCGUGGUUGCU	47585390	72
2501	chrX	UAAAGGGUUCCAAGCCUUAG	47585227	71
2502	chrX	GCAGACACUCUCCAUGGCGG	47585265	71
2503	chrX	GAAACUCCUCGCUGCGGUUG	47585305	71
2504	chrX	CUAGCAACCACGAGGGGGCG	47585403	71
2505	chrX	CUGAGCUUAGCUCAGCGCCG	47585622	71
2506	chrX	AACGCUGGUUAAGGUGGUUC	47584978	70
2507	chrX	AUAAAGGGUUCCAAGCCUUAA	47585226	68
2508	chrX	AAACUCCUCGCUGCGGUUGU	47585304	68
2509	chrX	CGGGCAGGAUUCAGGCUAUC	47586684	66

**FIG. 77**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2510	chr22	AUGAUGGCAAGAUGUACACG	32858043	82
2511	chr22	CACGAUGAGCCCGAGCCAAG	32802011	81
2512	chr22	GUGUAGACCAGCGUGCCGAA	32849501	80
2513	chr22	GGUGUAGACCAGCGUGCCGA	32849502	79
2514	chr22	GACUCUCGGAAGCUUCGUA	32857295	79
2515	chr22	UUGAUGAUGCUUUUAUCCGG	32859344	79
2516	chr22	GCACGAUGAGCCCGAGCCAA	32802012	78
2517	chr22	CGUAUGGAUGUACUGCACAU	32857279	78
2518	chr22	UACGCCUGCAUCCGGCAGAA	32859302	78
2519	chr22	CAGCAGCGGCAAUGACCCCU	32802008	77
2520	chr22	CCUGUAGGUUCGCGUCUAUGA	32858027	77
2521	chr22	CCGGAUGCAGGCGUAGUGUU	32859282	77
2522	chr22	GAUGAUGCUUUUAUCCGGGG	32859342	77
2523	chr22	UGAUGAUGCUUUUAUCCGGGG	32859343	77
2524	chr22	AUUGAUGAUGCUUUUAUCCG	32859345	77
2525	chr22	CCAGGAGCGCUUACCGAUGU	32802119	76
2526	chr22	UUUUGCCCACAGAUGUACCG	32857254	76
2527	chr22	CACGGGGCUGUGCAACUUCG	32858059	76
2528	chr22	UGCAGGCGUAGUGUUUGGAC	32859277	76
2529	chr22	CUACGCCUGCAUCCGGCAGA	32859301	76
2530	chr22	CGGCAAUGACCCCUUGGCUC	32802014	75
2531	chr22	GGGCGAGCAUGUGCACGCCU	32802068	75
2532	chr22	CCGUAUGGAUGUACUGCACA	32857280	75
2533	chr22	AUAGUUCAGCCCCUUGCGCU	32858094	75
2534	chr22	UUACCUUGCAGUUACAACCC	32858125	75
2535	chr22	GUACCGAGGAUGGGCCCCCCC	32859340	75
2536	chr22	AGCACGAUGAGCCCGAGCCA	32802013	74
2537	chr22	GCGGCAAUGACCCCUUGGCU	32802013	74
2538	chr22	GCCUUCUGCAACUCCGACAU	32802118	74
2539	chr22	GUAUGGAUGUACUGCACAUG	32857278	74
2540	chr22	GGUCCAGAGACACUCGUUCU	32859222	74
2541	chr22	GCCGCCUUUCUGCCGGAUGC	32859294	74
2542	chr22	GGAGGGGCCUUUCGGCACGC	32849506	73
2543	chr22	GGCUGUGCAACUUCGUGGAG	32858064	73
2544	chr22	UGUGCAACUUCGUGGAGAGG	32858067	73
2545	chr22	GCUGAACUAUCGGUAUCACC	32858119	73
2546	chr22	AGUGUUUGGACUGGUAGCCA	32859268	73
2547	chr22	ACUGCAGCUGGUACCGAGGA	32859330	73
2548	chr22	CUCGGGCUCAUUGUGCUCCU	32802031	72
2549	chr22	GUUGGCCAUUACCUUGUCAGC	32857355	72
2550	chr22	UCACCCUCUCCCAGCGCAAG	32858097	72
2551	chr22	CCAAACACUACGCCUGCAUC	32859294	72
2552	chr22	ACCGAUGUGCGGAGUUGCAGA	32802107	71
2553	chr22	GAUAGUUCAGCCCCUUGCGC	32858095	70
2554	chr22	GUAGCCAGGGUAACCGAAAU	32859255	70
2555	chr22	UGCAGUAGCCGCCUUUCUGC	32859301	70
2556	chr22	UAUCCGGGGGGCCAUCCU	32859331	70
2557	chr22	CUGCAGCUGGUACCGAGGAU	32859331	70

FIG. 78

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2558	chr6	UUUGGGAUCAUUGCCCUGUG	31577534	68
2559	chr6	CAGCAAACCUCAAGCUGAG	31577130	67
2560	chr6	GGUGUGGGUGAGGAGCACAU	31577283	67
2561	chr6	GCGUACAGGCUUUGUCACUCG	31576787	66
2562	chr6	UCUGGUAGGAGACGGCGAUG	31577311	66
2563	chr6	GGCGCUCCCCAAGAACAGACAG	31575803	65
2564	chr6	GCGCUCCCCAAGAACAGACAGG	31575804	65
2565	chr6	AAGCACCGCCUGGAGGCCUG	31575816	65
2566	chr6	UUGGAGUGAUCGGCCCCAG	31575916	65
2567	chr6	CCAGGAGGGCAUUGGCCGG	31577158	65
2568	chr6	GUGCAGCAGGCAGAACAGCG	31575880	64
2569	chr6	CCAGAGGGCUGAUUAGAGAG	31576550	64
2570	chr6	GUGGGUGAGGAGCACAUAGGG	31577280	64
2571	chr6	GCUGAGGAACAAGCACCGCC	31575826	63
2572	chr6	UGGAGUGAUCGGCCCCAGA	31575917	63
2573	chr6	UGAUUAGAGAGAGGUCCUG	31576541	63
2574	chr6	GCUGAGAGAUACCAGCUGG	31577206	63

**FIG. 79**

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2575	chr12	GGUGGGAAUAUACCCUCAG	6334210	77
2576	chr12	GUGGACGGGACACCGUGUG	6333467	77
2577	chr12	CCACGGCGUACAGCGUCGCG	6329609	77
2578	chr12	UAAUGUAUCGUACCAACGG	6330625	75
2579	chr12	AGAGGUGCACGGGUCCAUUG	6333386	74
2580	chr12	AAAGUUGGGACAGUCACCAGG	6329930	74
2581	chr12	AGGUGGCACCACCCUAUCAG	6329880	74
2582	chr12	GGUGGCACCACCCUAUCAGG	6329879	74
2583	chr12	UGCGCUGGAAGGAAUUCGUG	6329557	74
2584	chr12	GAAUCCUUCAGCGCAACG	6329574	73
2585	chr12	ACAGCAUGCUGGCGACCUGG	6329461	73
2586	chr12	UUGGUGGGAAUAUACCCUC	6334212	72
2587	chr12	GACCAGUCCAAUAACCCUG	6334207	72
2588	chr12	AGAGCUUGGACUUCCACCGU	6330623	72
2589	chr12	CUUUGCGGCUCCCCGAGAG	6329900	72
2590	chr12	AAUCCUUCAGCGCAACGG	6329575	72
2591	chr12	UGGACUGGUCCCUCACCUAG	6334183	71
2592	chr12	CAGCUGCUCCAAAUGCCGAA	6333742	71
2593	chr12	GUUUAAUGUAUCGUACCAA	6330628	71
2594	chr12	GGGUCAGCCCCUGAUAGGG	6329883	71
2595	chr12	CGCGACGCUGUACGCCGUGG	6329595	71
2596	chr12	UGAGCGACCACGAGAUCGAU	6329524	71
2597	chr12	UCCAGCGUGGCCUCGCGCCG	6329440	71
2598	chr12	GACAUCGAGGGAGGCGCUUUG	6329357	71
2599	chr12	UGGUGGGAAUAUACCCCUA	6334211	70
2600	chr12	UGGACAGUCAUUGUACAAGU	6333856	70
2601	chr12	AGGAACUACUACUAAGCCCC	6330038	70
2602	chr12	CUGCGGGGAGCCGCAAAGUU	6329916	70
2603	chr12	CGGUGACUGUCCAACUUUG	6329915	70
2604	chr12	CGGGGGCACGUUCUCCACCA	6329592	70
2605	chr12	ACGAAUUCUUCAGCGCAA	6329572	70
2606	chr12	CGUGCCCCCGUUGCGCUGGA	6329568	70
2607	chr12	AAUACAGCAUGCUGGCGACC	6329464	70
2608	chr12	CGAGGGCUGUCGCAAGGAUG	6329863	69
2609	chr12	GAGGUGGCACCACCCUAUCA	6329881	69
2610	chr12	AAGUCCAAGCUCUACUCCAU	6330602	69
2611	chr12	UGCAGCCACACACGGUGUCC	6333473	69
2612	chr12	ACCAGUCCAUAACCCCUGA	6334208	69
2613	chr12	CGAGGGCAGAACAGCAUGC	6329472	69
2614	chr12	ACCACGGCGUACAGCGUCGC	6329608	69
2615	chr12	UACUUGUACAAUGACUGUCC	6333843	69
2616	chr12	CACCACGGCGUACAGCGUCG	6329607	69
2617	chr12	GAACGGGCGCUGCCUGCGCG	6329490	69
2618	chr12	GCGAGGGCUGUCGCAAGGAU	6329862	69
2619	chr12	AGCUGCUCCAAAUGCCGAAA	6333741	68
2620	chr12	CUCACCAAUGGAGUAGAGCU	6330609	68
2621	chr12	ACAGUCACGGGGGUUAUAGG	6329939	68
2622	chr12	UGCAGUCCGUAUCCUGCCCC	6333831	68

FIG. 80

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2623	chr1	CGUGUUGGAGAACGUCCCCG	12192472	82
2624	chr1	UUGCCUGCCGAUAAGGCCCG	12201990	82
2625	chr1	GUGGGGCCUGCAAAUACCG	12192502	80
2626	chr1	GUGCUGCAGCAAUGCUCGC	12188890	79
2627	chr1	CCCGGGCGGCACUUGCAGCAG	12191886	79
2628	chr1	GGGGACCUGCUCGUCCUUCG	12206883	79
2629	chr1	CUCCAGUCCGACGGCCAGCG	12167119	78
2630	chr1	GUACUGCGCGCUGAGCAAGC	12191855	78
2631	chr1	AGGAACUGAAACAUCAGACG	12192446	78
2632	chr1	GUAACGUGGUGGCCAUCCCU	12192882	78
2633	chr1	GAUAAGGCCGGGGUACACA	12201999	78
2634	chr1	GCCUGUGGCUGGUUCCGAGU	12202098	78
2635	chr1	UGCCUGUGGCUGGUUCCGAG	12202099	78
2636	chr1	GGCAUUUACACCCUACGCC	12188815	77
2637	chr1	UCUCGCUGCUCUAGACCAGG	12191777	77
2638	chr1	GCGGUUCUGUUCCCCGAGUGC	12191797	77
2639	chr1	GUCGUGUUGGAGAACGUCCC	12192474	77
2640	chr1	CCACCCGGAGUAUGGCCCA	12192942	77
2641	chr1	CACCCGGAGUAUGGCCCA	12192943	77
2642	chr1	CGUCCUUCGGGGACUCCGAG	12206872	77
2643	chr1	GGAGUCACACCGUGUCCG	12190992	76
2644	chr1	GAGAACGUCCCCGGGGCACA	12192465	76
2645	chr1	UCGUGUUGGAGAACGUCCC	12192473	76
2646	chr1	ACUUGCCUGCCGAUAAGGCC	12201988	76
2647	chr1	CAGACGGCGACGGCGCCAU	12167094	75
2648	chr1	CUCCAACACGACUUCAUCCA	12192497	75
2649	chr1	CUGGGGCCAUACUCCGGGUG	12192928	75
2650	chr1	CCUGGGGCCAUACUCCGGGU	12192929	75
2651	chr1	CACGUCCCCCACCGGAGUA	12192934	75
2652	chr1	GGGGUAAGUGUACUGCCCCU	12192946	75
2653	chr1	CCCACCCGGAGUAUGGCC	12192941	75
2654	chr1	CUUGCCUGCCGAUAAGGCC	12201989	75
2655	chr1	CUCGUCCUUCGGGGACUCCG	12206874	75
2656	chr1	CGUAGGGUGUAAAUGCCACC	12188797	74
2657	chr1	CCGCUGCGCAAGUGCCGCC	12191898	74
2658	chr1	AGGACUCAGCUACUCAUC	12192524	74
2659	chr1	GAUGGCCACCACGUUACAGC	12192864	74
2660	chr1	UGUAACGUGGUGGCCAUCCC	12192881	74
2661	chr1	CCCUGGGGCCAUACUCCGGG	12192930	74
2662	chr1	ACUUUUCUGCGACUUACCAAC	12193097	74
2663	chr1	GCCUUUAUCGGCAGGAAGUG	12201972	74
2664	chr1	GUACCCCGGGCCUUUAUCGGC	12201981	74
2665	chr1	CGAUAAGGCCGGGUACAC	12201998	74
2666	chr1	UGGACAGAAGGGCGCCACU	12202096	74
2667	chr1	AGCCCAGAGCUCCAGUCCGA	12167128	73
2668	chr1	CUCCCGGGCUCCGGGCGUA	12188813	73
2669	chr1	GCUCAGCGCGCAGUACCAGC	12191836	73
2670	chr1	GGCGACUUUCGCUUUCCAGU	12193094	73

FIG. 81

SEQ ID NO:	Chromosome	Protospacer (crRNA)	Cut site coordinate	Score
2671	chr11	GUGCACGAUCUGAUGCCCGG	102110999	82
2672	chr11	UCAGAUCGUGCACGUCCGCG	102111021	82
2673	chr11	GCACGAUCUGAUGCCCGGCG	102110997	81
2674	chr11	GGUCGGUCUCCGAGUCCCCG	102111024	81
2675	chr11	CAUCAGAUCGUGCACGUCCG	102111019	79
2676	chr11	UGUUGGUACUGGUACGCA	102223634	79
2677	chr11	GGGCAACGAGGUUACCUGUC	102111167	78
2678	chr11	GGGGCAACGAGGUUACCUGU	102111168	78
2679	chr11	GGUUGCCGGGUCCGGACGG	102110938	77
2680	chr11	ACGAGGUUACCUGUCGGGAG	102111162	77
2681	chr11	ACGAUCUGAUGCCC GGCGGG	102110995	76
2682	chr11	GGGCUCCGGGCGGUJUGAAGA	102111134	76
2683	chr11	GUGUUGGUACUGGUACGCG	102223635	76
2684	chr11	GGAAGUCAUCUGGGGUUCGA	102227537	76
2685	chr11	AUCAGAUCGUGCACGUCCGC	102111020	75
2686	chr11	CGGGGACUCGGAGACCGGACC	102111039	75
2687	chr11	AUGAACCUUUACCAAAACGA	102186126	75
2688	chr11	GAUGAACCUUUACCAAAACG	102186127	75
2689	chr11	GCCGGUUGCCGGGUCCGGA	102110941	74
2690	chr11	GCCGUCCGGACCCGGGCAAC	102110952	74
2691	chr11	UGCACGAUCUGAUGCCCGGC	102110998	74
2692	chr11	GACGUGCACGAUCUGAUGCC	102111002	74
2693	chr11	GGACUCGGAGACCGACCUGG	102111042	74
2694	chr11	CGAGGUUACCUUGUCGGGAGU	102111161	74
2695	chr11	ACCCGGCAACCGGCACCCG	102110961	73
2696	chr11	GCCCAAUCCACUCCGAC	102111165	73
2697	chr11	CGAUCAGACAACAACAUUGC	102162479	73
2698	chr11	AGCCCACAGGGAGGGCUAU	102205972	73
2699	chr11	GUUGAAGAGCGGCCUCCAGGU	102111041	72
2700	chr11	GGGGCACGUUGGCCGUUG	102111075	72
2701	chr11	ACAUCAUCAGACAACAACA	102162475	72
2702	chr11	AACGGUUCUGCUGUGAGGGC	102229729	72
2703	chr11	GUUGCCCGGGUCCGGACGGC	102110937	71
2704	chr11	UGGGGCACGUUGGCCGUUU	102111076	71
2705	chr11	GGGUGCCGGUUGCCCGGGUC	102110945	70
2706	chr11	CAGGGCCCGCCGUCCGGACC	102110944	70
2707	chr11	AGGGCCCGCCGUCCGGACCC	102110945	70
2708	chr11	CGACUCCUUICUUAAGCCGC	102111141	70
2709	chr11	UAAUAGGCCAGUACUGAUGC	102114155	70
2710	chr11	GGGGCUGUGACGUUCAUCU	102162511	70
2711	chr11	CACGAUCUGAUGCCGGCGG	102110996	69
2712	chr11	CCAAGGCUUGACCCUCGUUU	102186127	69
2713	chr11	GCCGCGGGUGCCGGUUGCCC	102110950	68
2714	chr11	CGUGCACGUCCGCGGGACU	102111027	68
2715	chr11	AAUAUGAUGAACUCGGCUUC	102162567	68
2716	chr11	CGCCGCGGGUGCCGGUUGCC	102110951	67
2717	chr11	GCUCAGAUCCUUUCCUUAAC	102223748	67
2718	chr11	CUGGGGCACGUUGGCCGUU	102111077	65

FIG. 82

Human IL1A (IL1A-001; GRCh38)		Target		On-target	Off-target	Precision	Frameshift	Combined score^	
#	cRNA sequence	Exon #	Strand score*	score**	score***	score****	score*****	score^	
sg235	CAGAGACAGATGATCAATGG	3	-	69.5	67	0.57	93.7	72.6	SEQ ID NO: 2719
sg236	GCCATAGCTTACATGATAGA	4	-	53.9	76.9	0.65	93.9	78.6	SEQ ID NO: 2720

Machine-Learning (inDelphi) Predictions									
Top genotype	Repair outcome†								
insA (35.3%)	TACCTGATTGAGAGACAGATGATCAA  {A} TGGAGGAACTGTCTTCATTTCATTA								
insT (56%)	ATGGAGTGGCCATAGCTTACATGAT  {T} AGAAGGATTCTGTGAGGAAGGAAA								

Empirical data: ICE, day 10 post-nucleofection									
KO score	Top genotype	inDelphi rank	Repair outcome						
99%	insA (99%)	1st	TACCTGATTGAGAGACAGATGATCAA  {A} TGGAGGAACTGTCTTCATTTCATTA						
48%	insT (32%)	1st	ATGGAGTGGCCATAGCTTACATGAT  {T} AGAAGGATTCTGTGAGGAAGGAAA						

**FIG. 83A**

**Human IL1B (IL1B-201; GRCh38)**

#	cRNA sequence	Exon #	Strand	On-target score*	Off-target score**	Precision score***	Frameshift	Combined score^
sg237	TGATGGCCCTAACAGATGA	3	+	60.9	66.7	0.57	80	67.9 SEQ ID NO: 2721
sg238	GGTGTGGAGATTCTGAGC	4	-	58.3	49.3	0.6	86.3	65.2 SEQ ID NO: 2722
sg248	ACCTATCTTCCTGGACAT	5	+	61.7	86.5	0.65	89.9	80.5 SEQ ID NO: 2723
sg249	CTTCGACACATGGGATAACG	5	+	68.2	93.8	0.52	75.3	73.7 SEQ ID NO: 2724
sg250	GTCGACTTCAGTGTACGTAC	5	-	64.4	91	0.48	83.9	74.3 SEQ ID NO: 2725

**Machine-Learning (inDelphi) Predictions**

Top genotype	Repair outcome†
insA (33.3%)	CTTTGAAGCTGATGGCCCTAACAGA   {A} TGAAGGTAAAGACTATGGTTAACTC
insT (51.5%)	TGCCTGTAAGTGGGGTGGAGATTCTG   {T} AGCTGGATGCCGCCATCCAGAGGGCA
insA (51.9%)	CATTCAAGAACCTATCTCTCGACA   {A} CATGGGATAACGGAGGCTATGTGCAC
insA (33.4%)	ACCTATCTCTCGACACATGGGATA   {A} ACGAGGCTTATGTGCACGATGCACCT
insT (18.3%)	CCCCGGAGGGTGGCAGTTCACTGATCG   {T} TACACGGTGCATCGTGCACATAAGCCT

**Empirical data: ICE, day 10 (\*day 3) post-nucleofection**

KO score	Top genotype	inDelphi rank	Repair outcome
66% insA (40%)		1st	CTTTGAAGCTGATGGCCCTAACAGA   {A} TGAAGGTAAAGACTATGGTTAACTC
0% wild-type		NA	TGCTGTAGTGGGGTGGAGATTCTG   AGCTGGATGCCGCCATCCAGAGGGCA
3%* CAdel (3%)*		2nd	CATTCAAGAACCTATCTCTCGACA   -- TGGGATAACGGAGCTATGTGCAC
33%* insA (22%)*		1st	ACCTATCTCTCGACACATGGGATA   {A} ACGAGGCTTATGTGCACGATGCACCT
81%* Gdel (55%)*		8th	TCCCGGGAGCGTGCAGTTCACTGATC   TACAGGTGCATCGTGCACATAAGCCT

**FIG. 83B**

**Canine IL1A (IL1A-201; CanFam3.1)**

#	crRNA sequence	Target		On-target score*	Off-target score**	Precision score***	Frameshift 8****	Combined score^
		Exon #	Strand					
sg239	TGACCATCTCTCTGAATC	3	+	49.5	57.7	0.55	78.3	63.7 SEQ ID NO: 2726
sg240	GACATCCCAGCTTACCTCA	4	+	43.8	41.8	0.61	87.2	63.3 SEQ ID NO: 2727
sg251	AGTATAAGTTGAAACAGG	5	+	72.1	85.1	0.49	77.6	70.6 SEQ ID NO: 2728
sg252	TCTGTAATGGAGGTATG	5	-	68.4	66.6	0.48	90.4	68.3 SEQ ID NO: 2729

**Machine-Learning (inDelphi) Predictions**

Top genotype	Repair outcome†
insA (35.2%)	TTCGAAATGACCATTCTCTGA   {A} ATCAGGTAAAGTAAATGACTGCAAATC
inst (52.7%)	AATCTCAAGACATCCAGCTTACCT   {T} TCAAGGAAATGTGGTAGTGGGGCA
insC (30.6%)	CTCAAATCAAAGTATAGTTCGACAAAC   {C} AGGAGGAAATTACCCAAAGACTGGCTG
insC (20.4%)	CATCCAAATTCCTGTAATGCAGCAGTC   {C} ATGAGGTAAATTCCCTGTTGTCG

**Empirical data: ICE, day 10 post-nucleofection**

KO score	Top genotype	inDelphi rank	Repair outcome
90%	insA (56%)	1st	TTCGAAATGACCATTCTCTGA   {A} ATCAGGTAAAGTAAATGACTGCAAATC
97%	insT (57%)	1st	AATCTCAAGACATCCAGCTTACCT   {T} TCAAGGAAATGTGGTAGTGGGGCA

**FIG. 83C**

**Canine IL1B (IL1B-201; CanFam3.1)**

#	cRNA sequence	Target		On-target score*	Off-target score**	Precision score***	Frameshift 8****	Combined score^
		Exon #	Strand					
sg241	TGATGGCCCTGAAATGTGA	3	+	66.1	51.5	0.53	91.2	65.2
sg242	ACTCTTGTACAGAGCTGGT	4	-	68.9	62	0.64	84	70

**Machine-Learning (inDelphi) Predictions**

Top genotype	Repair outcome†
insT (22.9%)	CTTTGAAGCTGATGGCCCTGGAAATG    {T} TGAAGGTGAGACCATTGGCTTAGCTC
insT (56.4%)	ATGCCCTCAGACCTCTTGTACAGAGCT    {T} GGTTGGAGACTTGAACTGGATGCC

**Empirical data: ICE, day 10 post-nucleofection**

KO score	Top genotype	inDelphi rank	Repair outcome
99% deITG (95%)		2nd	CTTTGAAGCTGATGGCCCTGGAAATG    --AAGGTGAGACCATTGGCTTAGCTC
99% insT (99%)		1st	ATGCCCTCAGACCTCTTGTACAGAGCT    {T} GGTTGGAGACTTGAACTGGATGCC

**FIG. 83D**

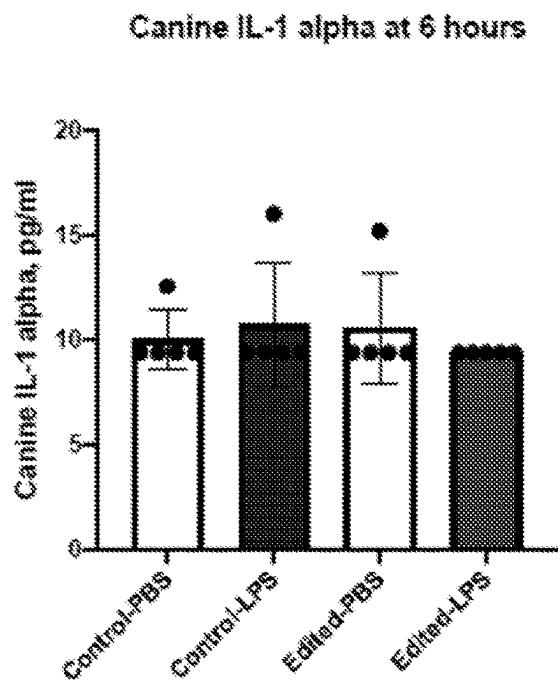


FIG. 84A

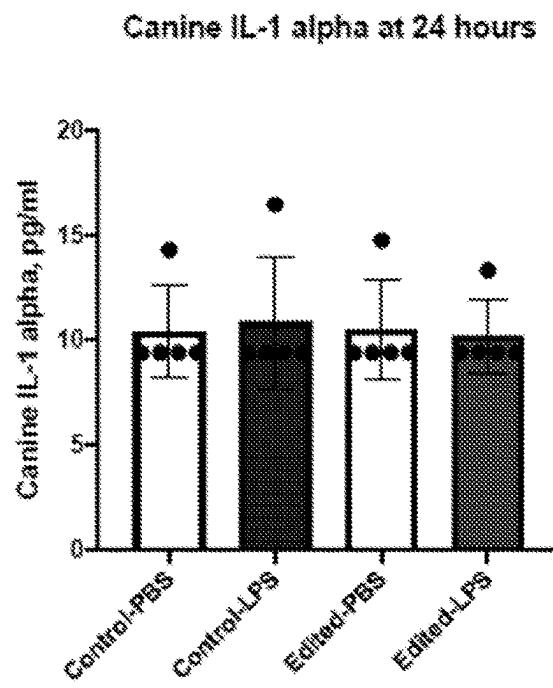


FIG. 84B

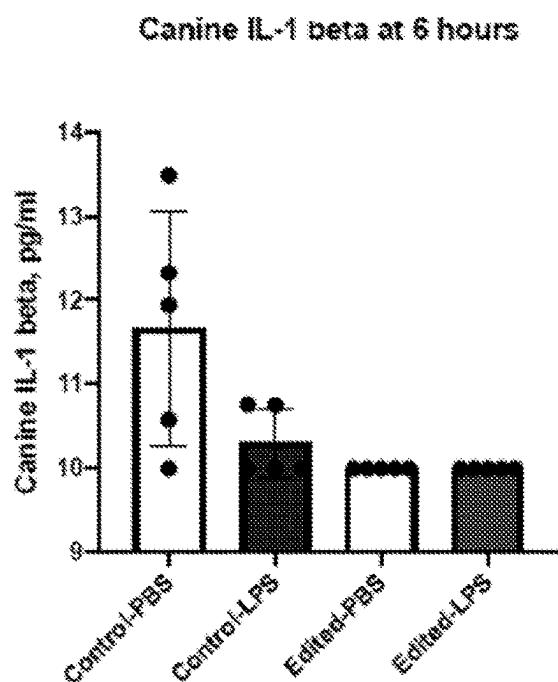


FIG. 84C

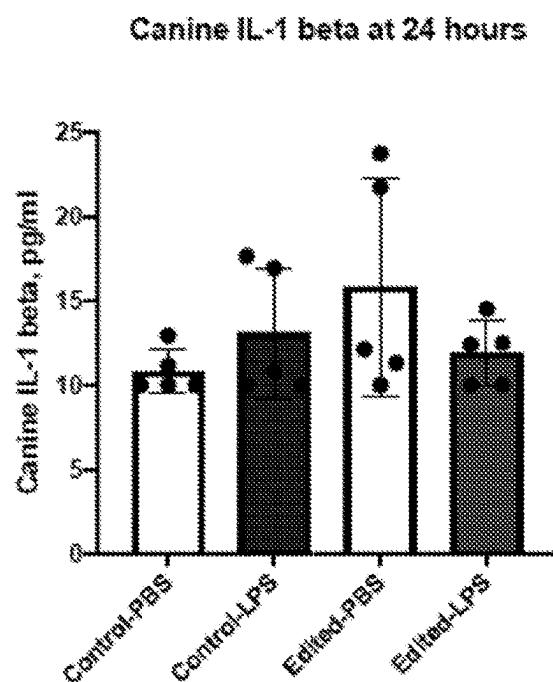
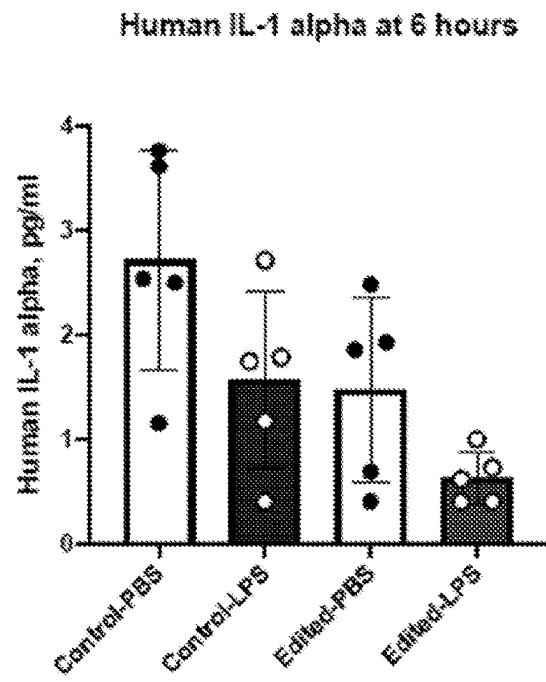
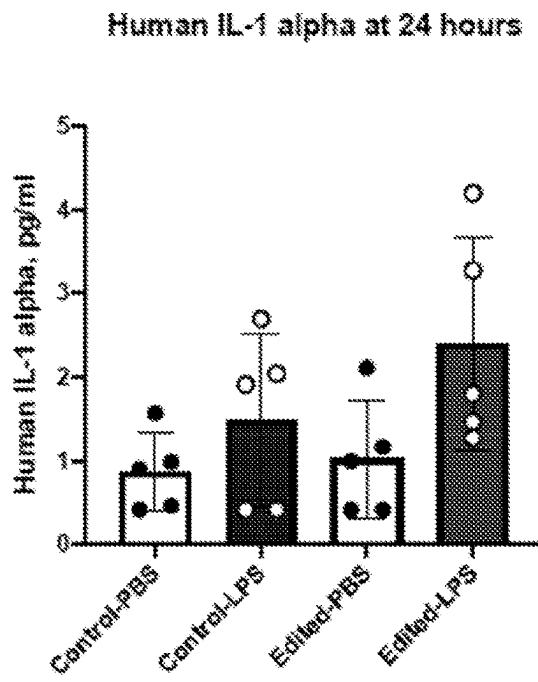


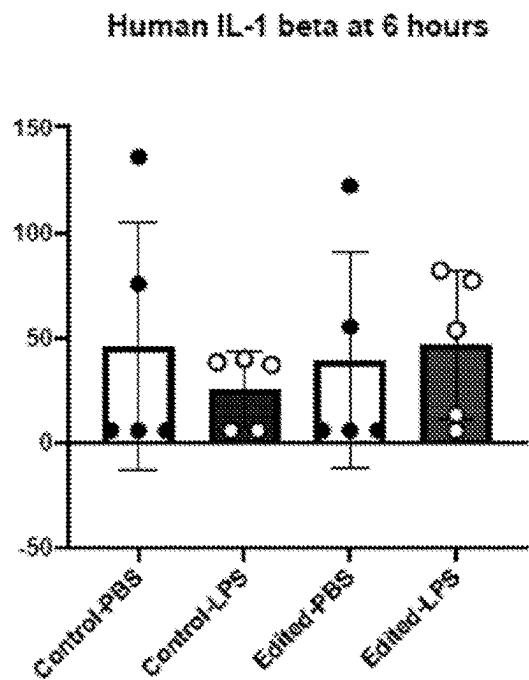
FIG. 84D



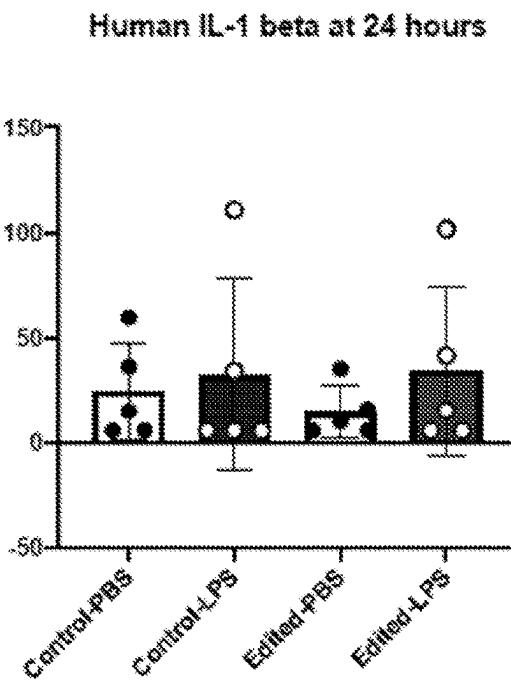
**FIG. 85A**



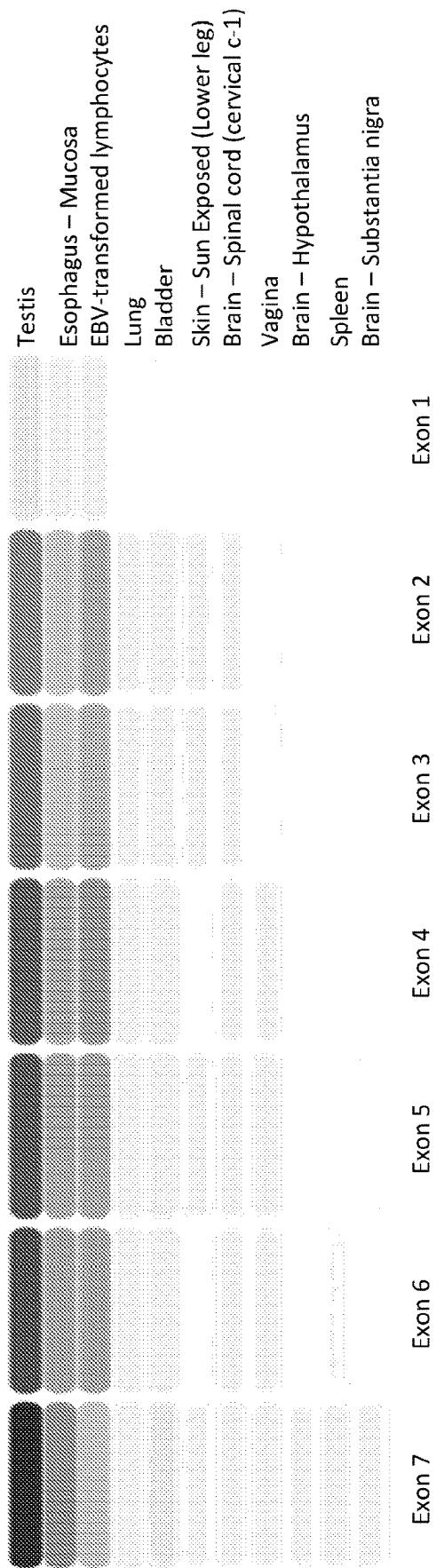
**FIG. 85B**



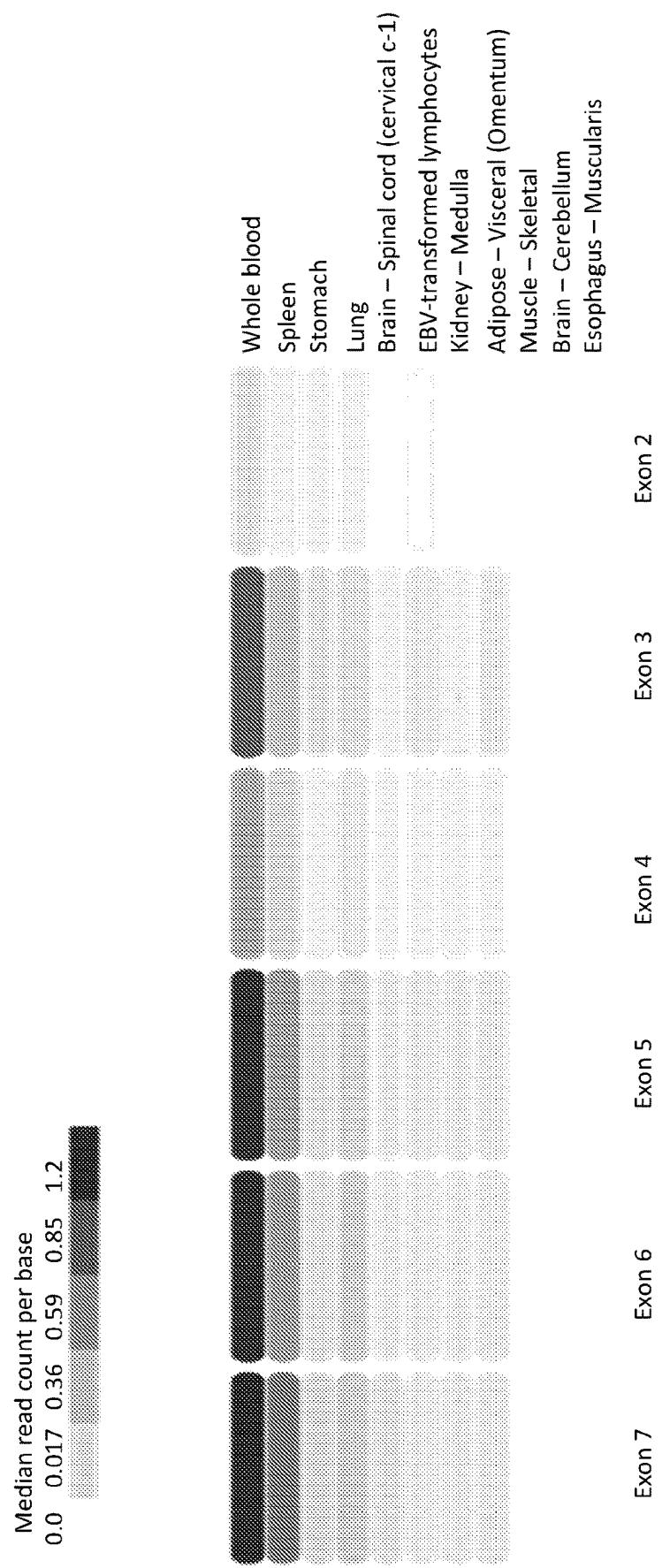
**FIG. 85C**



**FIG. 85D**



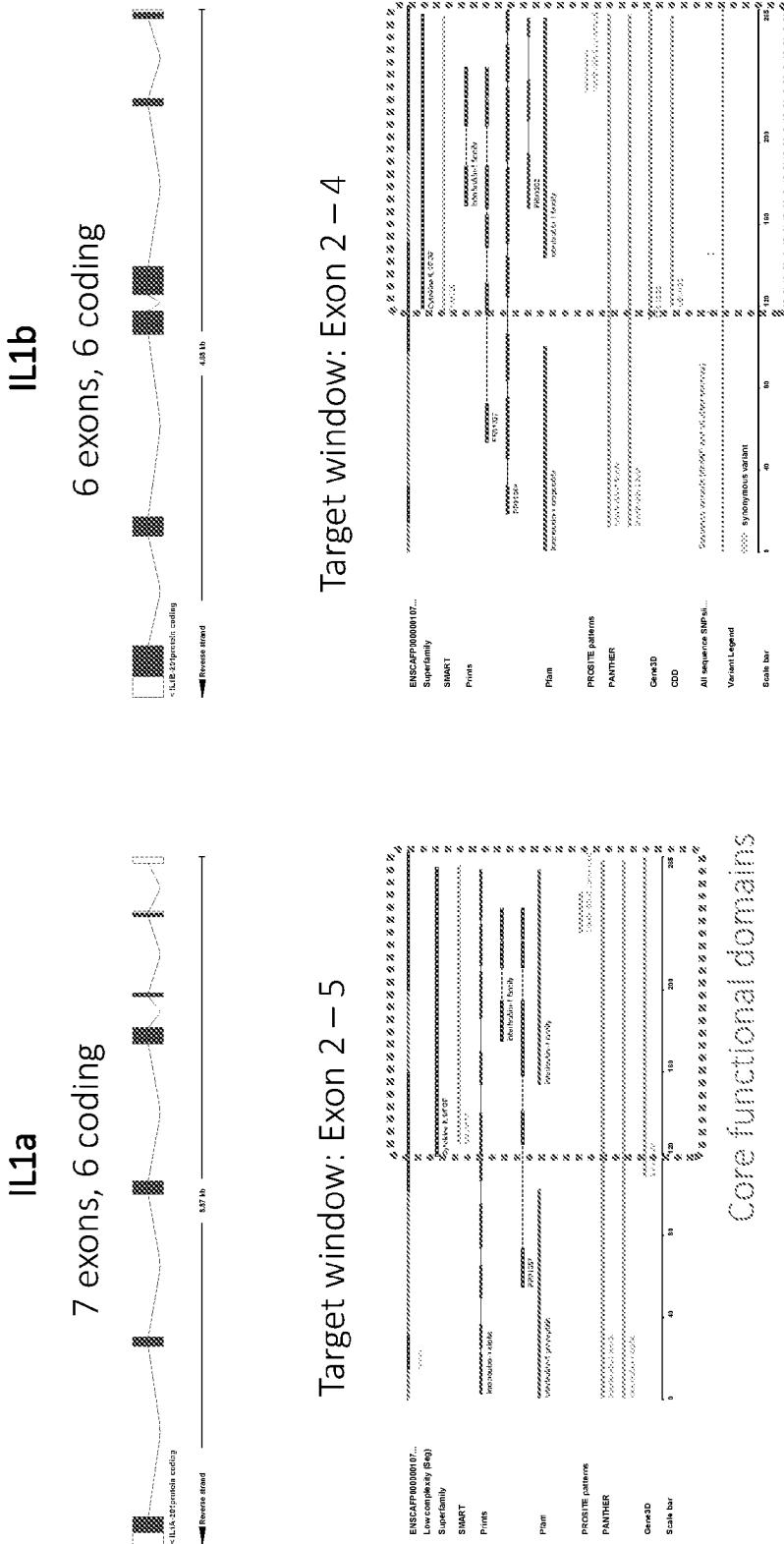
**FIG. 86**



**FIG. 87**

Single guide RNA (sgRNA)			Target			On-target		InDelphi Predictions		Off Targets	
#	Target sequence	Exon #	Strand	Domain	AA	Score	Precision	Frameshift	Indels	Other	
<b>Human IL-1A (IL1A-001; GRCh38)</b>											
235	CAGAGACAGATGATCAATGG	3	-	Interleukin-1 propeptide (1-109 aa)	25	0.68	0.57	94%	0.0.0-11.232 (0.0-0-2-1)		
236	GCCCATAGCTTACATGATAGA	4	-	Interleukin-1 propeptide (1-109 aa)	36	0.52	0.65	94%	0.0.0-14.133 (0.0-0-2-1)		
<b>Human IL-1B (IL1B-201; GRCh38)</b>											
237	TGATGGCCCTAACAGATGA	3	+	Interleukin-1 propeptide (1-103 aa)	32	0.55	0.57	80%	0.0.7-21-130 (0.0-2-7-2)		
238	GGGGTGGAGATTCTGTAGC	4	-	Interleukin-1 propeptide (1-103 aa)	50	0.57	0.6	86%	0.0.1-1.20 (0.0-0-0-0)		
248	ACCTATCTTCTTGACACAT	5	+		107	0.63	0.65	90%	0.0-0-15-140 (0.0-0-0-0)		
249	CTTCGACACATGGATAACG	5	+		110	0.66	0.52	75%	0.0-0-1-36 (0.0-0-0-0)		
250	GTGCAGTTCAAGTCGTAC	5	-		119/120	0.64	0.48	84%	0.0-0-2-27 (0.0-0-0-0)		
259	CATGCCACACAACTGACG	4	-	Interleukin-1 propeptide (1-103 aa)	66	0.67	0.46	79%	0.0-1-3-74 (0.0-0-0-2)		
260	AGGTCTGGAGGGACTG	4	-	Interleukin-1 propeptide (1-103 aa)	33/34	0.79	0.64	92%	0.0-14-4-377 (0.0-4-3-12)		

**FIG. 88**



**FIG. 89A**

**FIG. 89B**

Core functional domains

#	Target sequence	Single guide RNA (sgRNA)			Target			On-target	InDelphi Predictions	Off-targets
		Exon #	Strand	Domain	AA	Score	Precision			
<b>Canine IL-1A (IL1A-201; CanFam3.1)</b>										
239	TGACCATCTCTCTCTGAATC	3	+	Interleukin-1 propeptide (1-103 aa)		31	0.50	0.55	78%	0-0-2-24-451 (0-0-0-4-5)
240	CACATCCCAGCTTACCTTCA	4	+	Interleukin-1 propeptide (1-103 aa)		63	0.44	0.61	87%	0-0-0-12-123 (0-0-0-3-3)
251	AGTATAGTTGACAAACAGG	5	+	Cytokine IL1/FGF (122-260 aa)		144	0.72	0.49	78%	0-0-0-2-39 (0-0-0-0-0)
252	TCTGTAATTGCAGCAGTCATG	5	-	Cytokine IL1/FGF (122-260 aa)		150	0.68	0.48	90%	0-4-0-8-159 (0-0-0-0-4)
<b>Canine IL-1B (IL1B-201; CanFam3.1)</b>										
241	TGATGGCCCTGGAAATGTGA	3	+	Interleukin-1 propeptide (1-103 aa)		32	0.66	0.53	91%	0-0-1-29-183 (0-0-0-6-7)
242	ACTCTTGTATTACAGAGCTGGT	4	-	Interleukin-1 propeptide (1-103 aa)		56	0.69	0.64	84%	0-0-2-8-130 (0-0-1-3-3)

**FIG. 90**

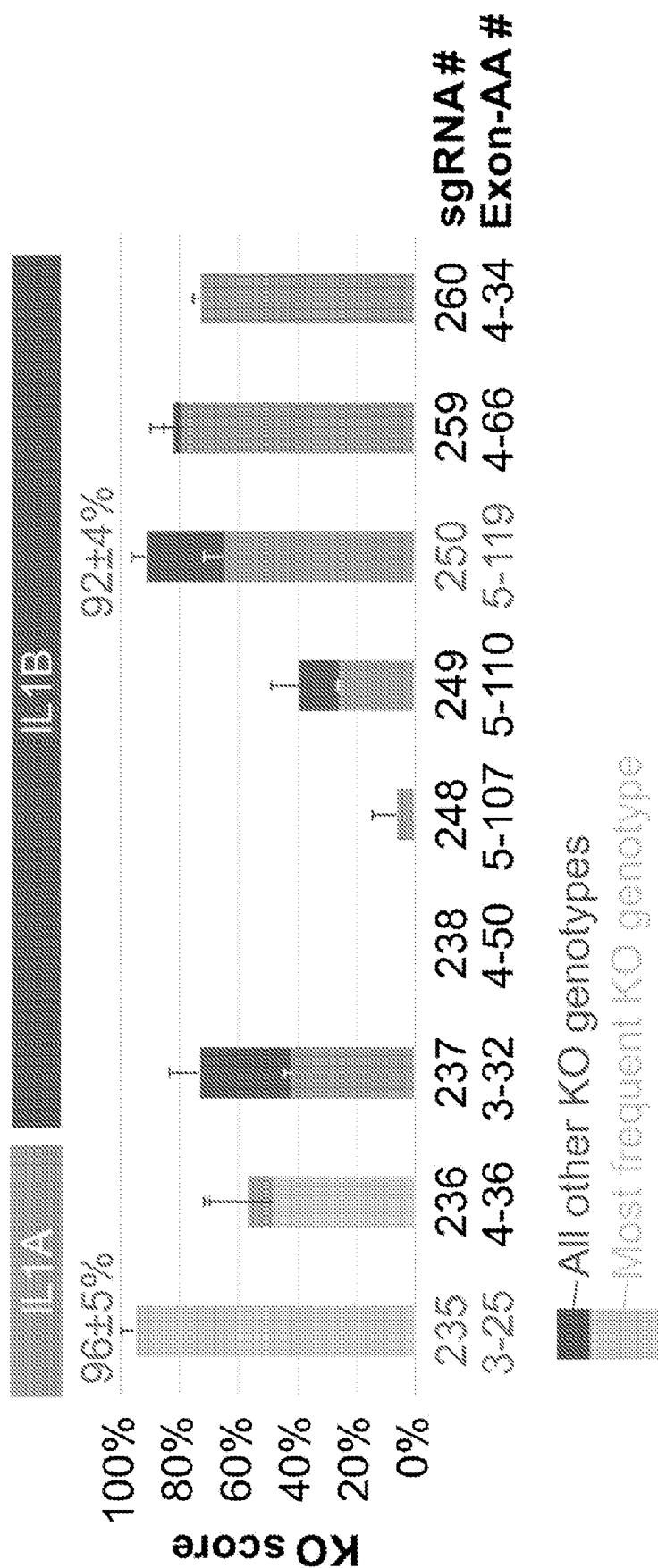
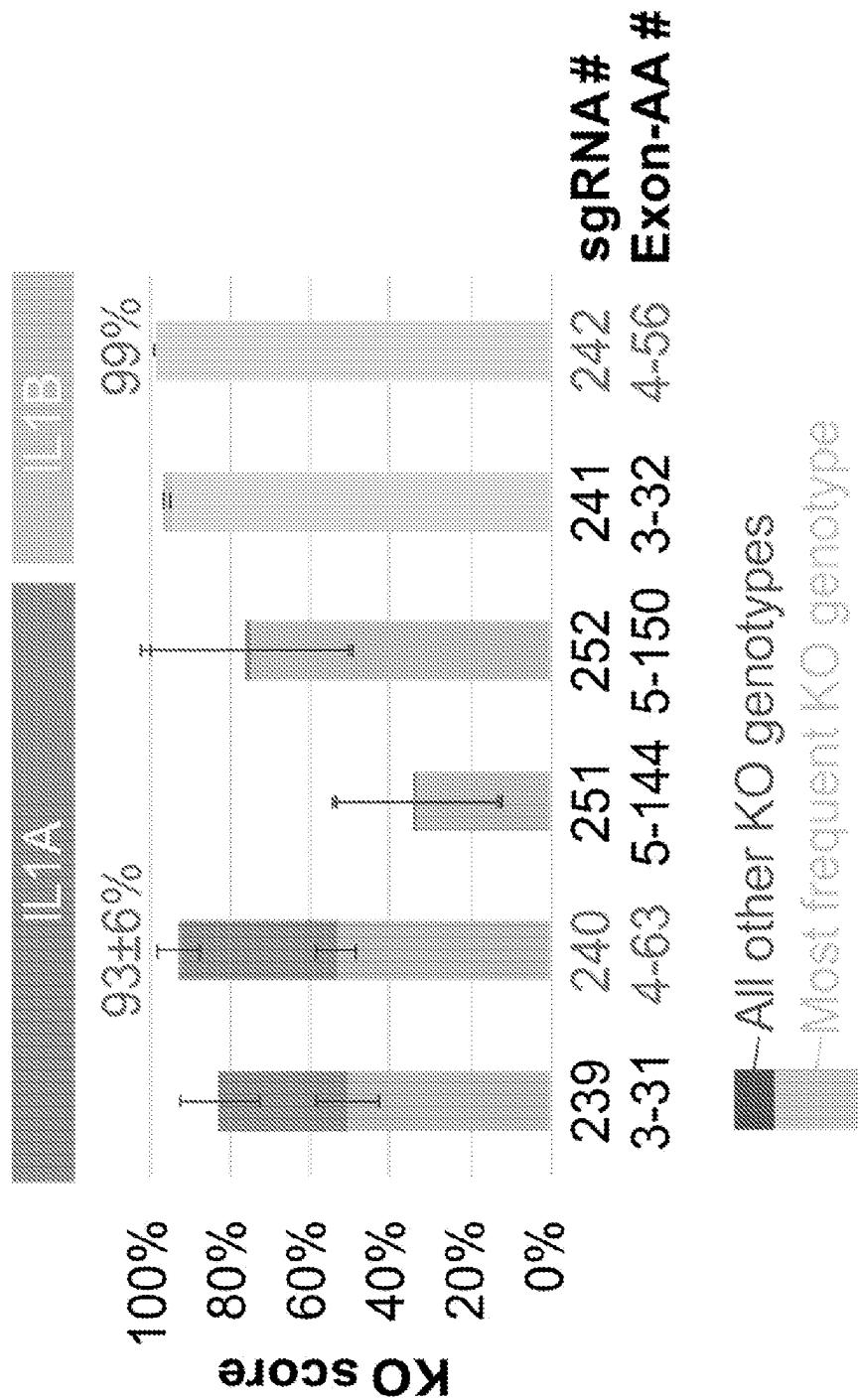


FIG. 91A



**FIG. 91B**

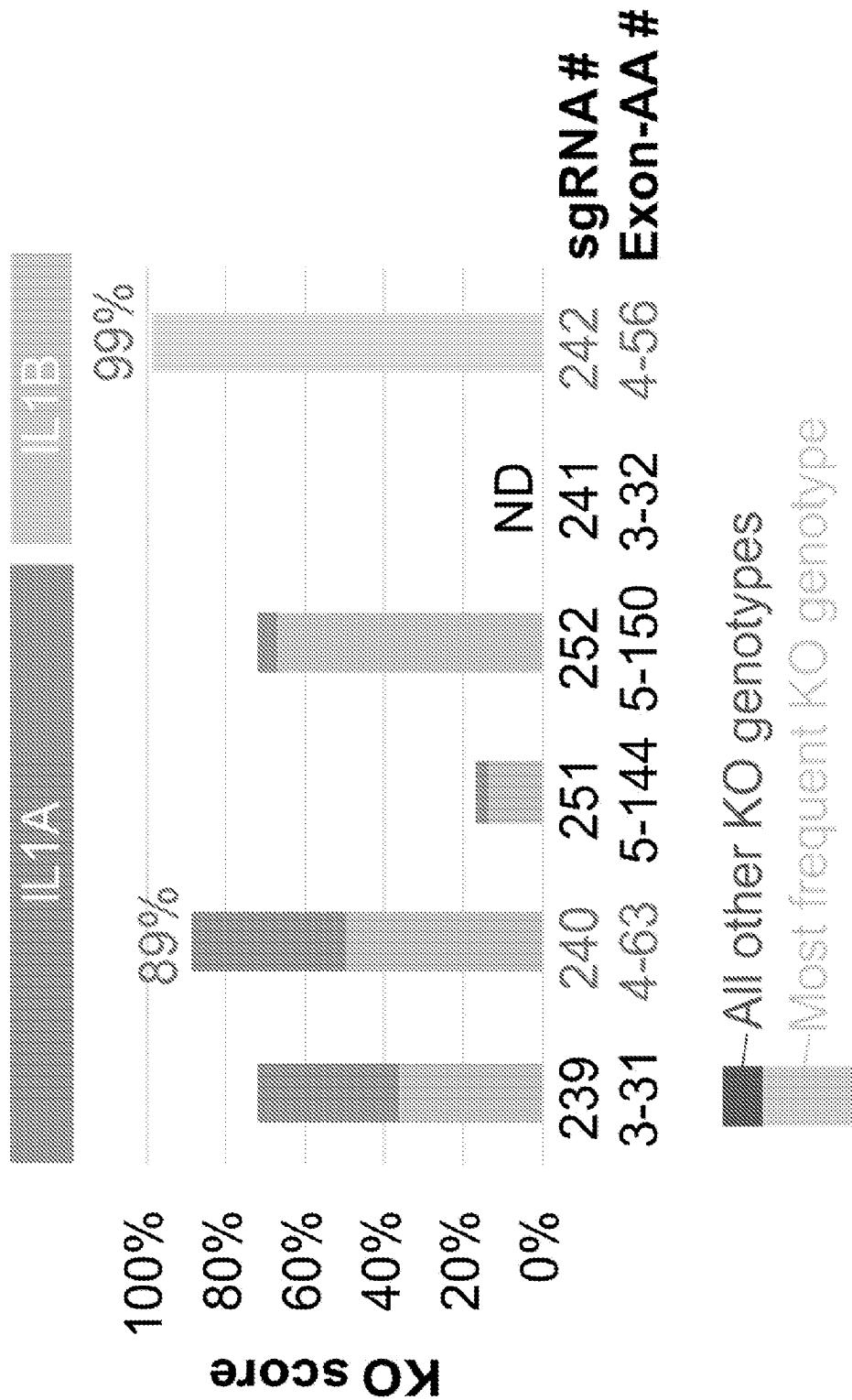


FIG. 91C

*In silico* predicted off-targets of sgRNA #240 in the dog (*Canis familiaris*) genome

Off-target	Sequence (Seed)	PAM	Score	Gene	Chromosome	Strand	Position	Mismatches	On-target	Editing efficiency
	GACATCCC <u>AGCTTACCTTC</u> A	AGG	100	IL1A	chr17	-1	36977152	0	TRUE	97%
#1	ZACATCCC <u>CCCTAACCTTC</u> A	CGG	1.67322207	lncRNA	chr27	1	34712401	3	FALSE	0%
#2	TAA <u>ATCCAGCTTACCTTC</u> A	CAG	1.54205034		chr35	1	18877859	3	FALSE	0%
#3	G <u>CCCCCCCAGCTTACCTTC</u> A	GAG	1.32551887		chr10	-1	26933301	4	FALSE	0%

*In silico* predicted off-targets of sgRNA #242 in the dog (*Canis familiaris*) genome

Off-target	Sequence (Seed)	PAM	Score	Gene	Chromosome	Strand	Position	Mismatches	On-target	Editing efficiency
	ACTCTTGTT <u>ACAGAGCTGGT</u>	GGG	100	IL1B	chr17	1	37022194	0	TRUE	99%
#1	ACT <u>TTTGTTCAGAGCTGGT</u>	CAG	6.16161972		chr33	-1	20234937	2	FALSE	0%
#2	CT <u>CTCATGTTACACAGCTGGT</u>	CCC	2.76564774		chr1	-1	47541563	3	FALSE	99%
#3	CT <u>CTCTTGTTCAGAGCTGGT</u>	GGG	2.32143742		chr16	1	32323843	3	FALSE	22%

**FIG. 92**

	On-target	Off-target #2	Off-target #3
Day 2	99%	0%	0%
Day 10	99%	0%	0%

**FIG. 93A**

	On-target	Off-target #2	Off-target #3
day 2	99%	99%	0%
day 10	99%	99%	6%

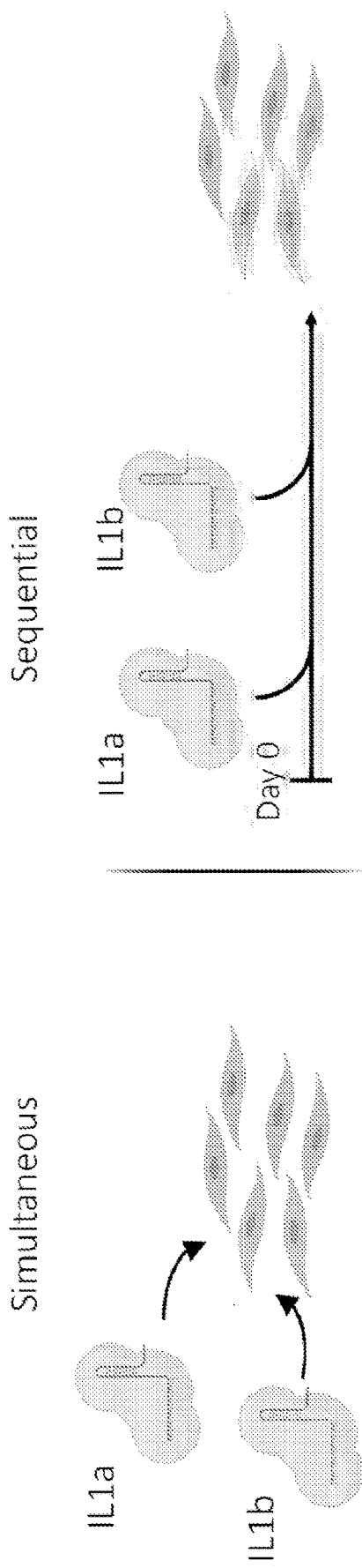
**FIG. 93B**

CHONDROCYTES						
Inference of CRISPR edits from Sanger traces (ICE), day 8-10 post-nucleofection						
#	Target sequence	Exon #	KO score	Top genotype	Predicted	Edit
<b>Human IL-1A (IL-1A-001; GRCh38)</b>						
235	CAGAGACAGATGATCAATGG	3	96±5%	insA	96±5%	ATTCAGAGACAGATGATCAA     TGG <u>A</u> CCACT
236	GCCATAGCTTACATGATAGA	4	57±13%	ins T	49±23%	TGCCCATAGCTTACATGAT     AGA <u>GG</u> ATT
<b>Human IL-1B (IL-1B-201; GRCh38)</b>						
237	TGATGGCCCTAACAGATGA	3	74±11%	insA	42±3%	AGCTGATGGCCCTAACAGA     TGA <u>AG</u> TTAAAG
238	GGTGGCGGGAGATTCGTAGC	4	0%	wt	0%	AGTGGTGGCGGGAGATTGT     AGCTGATGGCC
248	ACCTATCTCTTCGACACAT	5	6±8%	delCA	6±8%	AGAACCTATCTTCGACACA     TGGCATTAACCA
249	CTTCGACACATGGATAACC	5	40±9%	insA	26±0%	CTTCGACACATGGATA     ACC <u>GG</u> CTTAA
250	GTGCAGTTCACTGATCTAC	5	92±4%	delG	66±6%	AGCGTGGCACTGATC     TACAGCTTGCACTC
259	CATGGCACACAACTGACCG	4	83±8%	delG	80±6%	GTCCATGGCACACAACT-     ACGGGGCCTGCCT
260	AGGTCCTGGAAAGGAACTG	4	74±2%	insA	74±2%	TCCAGGTCCCTGGAAAGGAACTG     CTG <u>GG</u> AGAS

FIG. 94A

CHONDRIOCYTES						
Single guide RNA (sgRNA)		Inference of CRISPR edits from Sanger traces ((CE), day 8-10 post-nucleofection				
#	Target sequence	Exon #	KO score	Top genotype	Predicted	Edit
<b>Canine IL-1A (IL1A-201; CanFam3.1)</b>						
239	TGACCATTCTCTCTGAATC	3	83±10%	insA	51±8%	35%
240	GACATCCCAGCTAACCTCA	4	93±6%	insT/delT	54±5%	53%
251	AGTATACTTGACAAACAGG	5	34±21%	insC	33±21%	31%
252	TCTGTAAATGCCAGTCATG	5	77±26%	insC	75±25%	20%
<b>Canine IL-1B (IL1B-201; CanFam3.1)</b>						
241	TGA1GGCCCTGGAAATGTGA	3	99%	delTG	95%	17%
242	ACTCTTGTACAGAGCTGGT	4	99±0%	insT	99±0%	56%

FIG. 94B



	IL1A KO score	IL1B KO score
Simultaneous KO: wild-type cells + sgRNA #240 + sgRNA #242	69%	99%
Sequential KO: canine IL1B KO cells + sgRNA #240	68%	99%
Sequential KO: canine IL1A KO cells + sgRNA #242	97%	99%

**FIG. 95**

**IL1A sgRNAs**

Human	sgRNA #235 <<<<<<<<<<	hILIA GTTCCTCCATTGATCATCTGTCTCTGAAT eILIA ...TGAA....C....C....C. cILIA ...TGAA....C....C.... mILIA ...TG....C....C....
	sgRNA #236 <<<<<<<<<<	hILIA AATCCCTCTATCATGTAAAGCTATGGCCCA eILIA .....G...C.....A... cILIA .....G..A.G.....G..A.. mILIA .....G...C.....T..
	sgRNA #239 >>>>>>>>>>	cILIA AATTGACCCATCCTCTCTGAATCGGTAA hILIA C.....T...G..... eILIA .....C.. mILIA C.....
	sgRNA #240 >>>>>>>>>>	cILIA AAAGACATCCCAGCTTACCTTCAGGAAA hILIA T..A.....A.....G.. eILIA T.....A...G.A.....G.. mILIA ....TG...A.CT.C.....G.
	sgRNA #251 >>>>>>>>>>>	cILIA CAAAGTATACTTCGACAAACAGGAGGAAA hILIA .....A.....GCC.---AT.ATC. eILIA .....G..A.....G.C...TC...TC.. mILIA .....C...TA...AGG.TGTG.ACAC.C.
	sgRNA #252 <<<<<<<<<<	cILIA TTACCTCATGACTGCTGCATTACAGAATT hILIA G.....C.G.....T...C eILIA A..T..TGC.....A.T..C mILIA C..T...GC..CA..TGG...A..TG..CC
	sgRNA #086 >>>>>>>>>>>	mILIA ACAGTATCAGAACGGTCAAGCAACGGGAA hILIA GT...G..A..... eILIA CTG..GG.. cILIA GTG..GG...P....
	sgRNA #087 >>>>>>>>>>>	mILIA CAGTATCAGAACGGTCAAGCAACGGGAA hILIA T...G..A..... eILIA TG..GG.. cILIA TG..GG...T....
	sgRNA #096 <<<<<<<<<<	mILIA AGACCTTCACTGAAGATGACCTGCAGTCC hILIA ..AT..A.....T.....G..G.. eILIA ..TT.A...CA.T.....G.AG.. cILIA ..ATT.A...C..T.....G.AGA.
	sgRNA #101 <<<<<<<<<<	mILIA AATCCTTCTATGATGCAAGCTATGGCTCA hILIA .....C..T.....C.. eILIA .....A.C.. cILIA .....ATG...G..A.C..

**FIG. 96A**

## IL1B sgRNAs

	sgRNA #237 >>>>>>>>>>>>>	HIL1B	AGCTGATGCCCTAAACAGATGAGGTAA
	sgRNA #238 <<<<<<<<<	HIL1B	CATCCAGCTACGAATCTCGACCACACT
	sgRNA #246 >>>>>>>>>>>>	HIL1B	AGAACCTATCTTCTTCGACACATGGATA
	sgRNA #249 >>>>>>>>>>>>	HIL1B	CTTCTTCGACAATGGGATAACCGAGGCTT
Human	sgRNA #250 <<<<<<<<<	HIL1B	A...GT...AC...G.T....
	sgRNA #259 <<<<<<<<<	HIL1B	A...G.A.A...GCG...TAA...
	sgRNA #260 <<<<<<<<	HIL1B	C...GT...T...G.T...T.A.A
	sgRNA #241 >>>>>>>>>>	CIL1B	AGCTGATGCCCTGGAAATGTCAAGGTGA
Canine	sgRNA #242 <<<<<<<<<	HIL1B	.....AA.C.GA.....A.
	sgRNA #063 >>>>>>>>>>	HIL1B	G.A.G.....AAA.C.GA.....
Murine	sgRNA #074 <<<<<<<<<	HIL1B	..T...C...A...CCA...GA.....
	sgRNA #080 <<<<<<<<<	HIL1B	TGACCTGGCTGTCTT-CATCACACCATCC
	sgRNA #081 <<<<<<<<	HIL1B	G.....CT...CC...TG.....CGG.....
	sgRNA #082 <<<<<<<<	HIL1B	G.....CA...CCATGGGC...G.G.....
	sgRNA #083 <<<<<<<<	HIL1B	GA...ACA.T.C...TCGTA...G.....

FIG. 96B

## LIPID NANOPARTICLES FOR GENE EDITING SYSTEMS

### CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to U.S. Provisional Patent Application No. 63/334,476, filed Apr. 25, 2022, U.S. Provisional Patent Application No. 63/362,858, filed Apr. 12, 2022, U.S. Provisional Patent Application No. 63/342,471, filed May 16, 2022, and U.S. Provisional Patent Application No. 63/495,461, filed Apr. 11, 2023, the contents of which are hereby incorporated by reference herein, in their entireties, for all purposes.

### BACKGROUND OF THE DISCLOSURE

#### Joint Disorders

[0002] Treatment of osteoarthritis, degenerative joint disease, and other joint dysfunction is complex and there are few long term options for either symptomatic relief or restoring joint function. Osteoarthritis (OA) is the leading cause of disability due to pain. Neogi, *Osteoarthritis Cartilage* 2013; 21:1145-53. All mammal species are affected: working animals, domestic pets, and their owners all suffer OA-related discomfort, pain, and disability, depending on the degree of disease progression.

[0003] OA is a complex disease characterized by a progressive course of disability. Systemic inflammation is associated with OA and with OA disease progression. Inflammation is driven by increased levels of pro-inflammatory cytokines. New methods and compositions to treat this disease are acutely needed. Disclosed herein are compositions and methods useful for treating OA as well as other inflammatory joint disorders.

#### Back and Spine Disorders

[0004] Back or spine conditions or disorders, including low back pain, and pain or inflammation associated with discogenic disorders e.g., degenerative disc disease (DDD) or internal disc disruption (IDD), is a major cause of morbidity and disability worldwide for which few long-term options for amelioration currently exist. Andersson GB. Epidemiological features of chronic low-back pain. *Lancet*. 1999; 354:581-585. Presently available treatments include surgical or less invasive options that often fail to offer long-term palliation. Ju, et al. *Global Spine Journal* (2020): 2192568220963058. All vertebrate species are affected by back or spine conditions or disorders, including working animals, domestic pets, and their owners. All suffer from the associated discomfort, pain, and disability, depending on the degree of disease progression.

[0005] Back or spine conditions or disorders, such as low back pain, are complex diseases characterized by a multitude of inputs contributing to a progressive course of disability. Among these contributors are morphological irregularities (e.g., disc disruptions), inflammation, and changes in the localized cellular environment (e.g., vascularization and/or innervation). Peng, Bao-Gan. *World Journal of Orthopedics* 4.2 (2013): 42. Each contributing factor is driven by differential expression of various gene products, including at least pro-inflammatory cytokines, growth factors and other effector biomolecules. New methods and compositions to treat this disease are acutely needed.

### BRIEF SUMMARY OF THE DISCLOSURE

[0006] Provided herein are compositions and methods for treating synovial joint dysfunction, are described herein. In addition, compositions, and methods for treating or preventing localized nociception, inflammation, or morphological changes associated with back or spine conditions or disorders, are disclosed herein. Further, compositions and methods for the treating or preventing musculoskeletal disease and dysfunction, including fibrosis and/or scarring in, for example, post-operative subjects are herein described. Additionally, methods for gene-editing cells, including, but not limited to synovial cells and/or synoviocytes, chondrocytes, synovial macrophages, and synovial fibroblasts, and uses of gene-edited synovial cells and/or synoviocytes, chondrocytes, synovial macrophages, and synovial fibroblasts, in the treatment of diseases such as osteoarthritis are disclosed herein.

### BRIEF DESCRIPTION OF THE DRAWINGS

[0007] The presently disclosed embodiments will be further explained with reference to the attached drawings. The drawings shown are not necessarily to scale, with emphasis instead generally being placed upon illustrating the principles of the presently disclosed embodiments.

[0008] FIG. 1 illustrates SEQ ID NOs: 1-48, the crRNA sequences generated by the bioinformatic methods herein described that target human ADAM17 to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0009] FIG. 2 illustrates SEQ ID NOs: 49-96, the crRNA sequences generated by the bioinformatic methods herein described that target human ADAMTS1 to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0010] FIG. 3 illustrates SEQ ID NOs: 97-144, the crRNA sequences generated by the bioinformatic methods herein described that target human ADAMTS5 to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0011] FIG. 4 illustrates SEQ ID NOs: 145-192, the crRNA sequences generated by the bioinformatic methods herein described that target human ADM to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0012] FIG. 5 illustrates SEQ ID NOs: 193-240, the crRNA sequences generated by the bioinformatic methods herein described that target human ATP1A1 to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0013] FIG. 6 illustrates SEQ ID NOs: 241-281, the crRNA sequences generated by the bioinformatic methods herein described that target human BDNF to modify and/or ablate expression of its encoded products. Additional infor-









sembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0086] FIG. 79 illustrates SEQ ID NOs: 2558-2574, the crRNA sequences generated by the bioinformatic methods herein described that target human TNF to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0087] FIG. 80 illustrates SEQ ID NOs: 2575-2622, the crRNA sequences generated by the bioinformatic methods herein described that target human TNFRSF1A to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0088] FIG. 81 illustrates SEQ ID NOs: 2623-2670, the crRNA sequences generated by the bioinformatic methods herein described that target human TNFRSF1B to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0089] FIG. 82 illustrates SEQ ID NOs: 2671-2718, the crRNA sequences generated by the bioinformatic methods herein described that target human YAP1 to modify and/or ablate expression of its encoded products. Additional information includes the chromosomal genomic coordinates (assembly hg38) of the edit site and a score, summarizing several predicted performance metrics.

[0090] FIGS. 83A, 83B, 83C, and 83D collectively illustrate the results of cell-based and in-silico gene editing analysis of crRNA sequences targeting (A) hIL1A, (B) hIL1B, (C) cIL1A, and (D) cIL1B genes. “ $\circ$ ” denotes CRISPR cut position within the translation frame of amino acids (AA). “ $\ast$ ” denotes optimized score from Doench, Fusi et al. (2016). This score is optimized for 20 bp guides with an NGG PAM. Score spans from 0 to 100. Higher is better. “ $\ast\ast$ ” Specificity score from Hsu et al. (2013). Score spans from 0 to 100. Higher is better. “ $\ast\ast\ast$ ” This score is based on experiments in U2OS. A high precision score ( $>0.4$ ) implies that DNA repair outcomes are uniform and enriched for just a handful of unique genotypes. “ $\ast\ast\ast\ast$ ” This score is based on experiments in U2OS. A high ( $>80\%$ ) frameshift frequency will tend to knock a protein-coding gene out of frame. The typical genomic frameshift frequency is above 66% because 1-bp insertions and 1-2 bp deletions are particularly common repair outcomes. ^

[0091] Combined score=(Off-target score+Precision score\*100+Frameshift)/3. † Pipe symbol ‘|’ indicates CRISPR cut site. Curly braces ‘{ }’ indicate insertion. Hyphen ‘-’ indicates deletion. § Potential off-target sites. Scoring according to Hsu et al. (2013). The on-target site has a score of 100.

[0092] FIGS. 84A, 84B, 84C, and 84D collectively illustrate results of functional assays in edited or control canine chondrocytes measuring (A, B) cIL1A and (C,D) cIL1B release at 6 hours and 24 hours post-exposure to PBS or LPS.

[0093] FIGS. 85A, 85B, 85C, and 85D collectively illustrate results of functional assays in edited and control chondrocytes measuring (A, B) hIL1A and (C,D) cIL1B release from 6 hours and 24 hours after exposure to PBS or LPS.

[0094] FIG. 86 illustrates the results of a tissue-specific splicing and expression analysis of the hIL1A gene.

[0095] FIG. 87 illustrates the results of a tissue-specific splicing and expression analysis of hIL1B gene.

[0096] FIG. 88 illustrates the results of an in silico analysis of crRNAs targeting either hIL1A or hIL1B. On-target score (see Doench et al.) is optimized for 20-bp gRNA with NGG protospacer adjacent motif (PAM). Score spans from 0 to 1. Precision score is based on experiments in U2OS cells. A high precision score ( $>0.4$ ) implies that DNA repair outcomes are uniform and enriched for just a handful of unique genotypes. Frameshift percentage is based on experiments in U2OS cells. A high ( $>80\%$ ) frameshift frequency will tend to knock a protein-coding gene out of frame. The typical genomic frameshift frequency is above 66% because 1-bp insertions and 1-2 bp deletions are particularly common repair outcomes. Off-target score from CRISPR assess the number of matches in the genome with a given number of mismatches. Mismatches in Seed sequence have a more deleterious effect.

[0097] FIG. 89 illustrates results of splicing and functional analyses on cIL1A and cIL1B genes. The reference canine genome assembly (CanFam3.1) was used for these analyses.

[0098] FIG. 90 illustrates the results of an in silico analysis of crRNAs targeting either cIL1A or cIL1B genes.

[0099] FIGS. 91A, 91B, and 91C collectively illustrate knockdown efficacy of selected sgRNAs in (A) human chondrocytes, (B) canine chondrocytes and (C) canine synoviocytes.

[0100] FIG. 92 illustrates the results of an in silico analysis of the off-target effects for multiple sgRNAs in canine cells.

[0101] FIGS. 93A and 93B collectively illustrate (A) the efficacy of enhanced-specificity Cas9 (espCas9) to abrogate the off-target editing of the indicated sgRNA in canine cells as compared to (B) the effects with canonical spCas9.

[0102] FIGS. 94A and 94B collectively illustrate summaries of editing activity for crRNAs targeting IL1A and IL1B in (A) humans and (B) canine chondrocytes.

[0103] FIG. 95 illustrates results of co-administrating multiple sgRNAs in canine cells either simultaneously or sequentially.

[0104] FIGS. 96A and 96B collectively illustrate sequence alignments of (A) IL1A and (B) IL1B genes for disparate mammalian species (human, horse, mouse and dog).

## DETAILED DESCRIPTION OF THE DISCLOSURE

### I. Introduction

[0105] Provided herein are compositions and methods for silencing the translation of one or more proteins in an animal in need thereof to treat a disease, illness or condition associated with pain.

[0106] In some embodiments, receptor signaling is silenced by CRISPR editing of the gene encoding the receptor. In some embodiments, the CRISPR editing results in ablation of a transmembrane domain (i.e., generation of a soluble decoy receptor). In some embodiments, the CRISPR editing results in ablation of a cytoplasmic domain (i.e., generation of a membrane-bound decoy receptor). In particular embodiments, compositions and methods are provided to gene-edit FGF2, CCN2, ADAMTS5, MMP1 and/or NGF.

## II. Definitions

**[0107]** Unless defined otherwise, all technical and scientific terms used herein have the same meaning as is commonly understood by one of skill in the art to which this invention belongs. All patents and publications referred to herein are incorporated by reference in their entireties.

**[0108]** In some embodiments, pain is ameliorated by silencing of a nociception signaling protein (or its cognate receptor) via CRISPR editing of the gene encoding the protein (or receptor). In some embodiments, the CRISPR editing results in ablation of a transmembrane domain of a pain receptor (i.e., generation of a soluble decoy receptor). In some embodiments, the CRISPR editing results in ablation of the cytoplasmic domain of a pain receptor (i.e., generation of a membrane-bound decoy receptor). In particular embodiments, compositions and methods are provided to gene-edit (i) one or more growth factors or growth factor receptors (e.g., FGF2, CCN2, NGF, NTF3, NTF4, BDNF, FGFR1, NGFR, NTRK1, or NTRK2), (ii) one or more metalloproteases or regulators thereof (e.g., ADAM17, ADAMTS1, ADAMTS5, MMP1, MMP2, MMP3, MMP7, MMP8, MMP10, MMP12, MMP13, TIMP1, or TIMP3), (iii) one or more cytokines, chemokines or cytokine/chemokine receptors (e.g., CXCL1, CXCL2, CXCL3, CXCL5, CXCL6, CXCL8, CCL2, CCL3, CCL5, CCL7, CCL20, IL1A, IL1B, IL4, IL6, IL10, IL13, IL17A, IL18, TNF, CXCR1, CXCR2, CCR7, TNFRSF1A, TNFRSF1B, IL1R1, IL1RAP, IL4R, IL6R, IL10RA, IL10RB, IL13RA1, IL13RA2, IL17RA, IL18R1, or IL18RAP), (iv) one or more regulators of neuronal signaling (e.g., SCN1A, SCN2A, SCN3A, SCN4A, SCN5A, SCN8A, SCN9A, SCN10A, SCN11A, TAC1, TAC3, TACR1, TACR2, TACR3, or ATP1A1), (v) one or more other regulators of cell signaling (e.g., CALCA, CALCB, CALCRL, RAMP1, ADM, CRCP, YAP1, MRGPRX2), or (vi) a combination of any genes of (i)-(v) to ameliorate pain.

### Definitions

**[0109]** Unless defined otherwise, all technical and scientific terms used herein have the same meaning as is commonly understood by one of skill in the art to which this disclosure belongs. All patents and publications referred to herein are incorporated by reference in their entireties.

**[0110]** The term “FGF2 gene” refers to a mammalian gene encoding a Fibroblast growth factor 2 polypeptide. Non-limiting examples of FGF2 genes include: NCBI Gene ID: 2247 [human], NCBI Gene ID: 403857 [canine], NCBI Gene ID: 100033955 [equine], NCBI Gene ID: 100135772 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an FGF2 gene include: UniProt: P09038; NP\_001348594.1 [human], XP\_038421156.1 [canine], NP\_001182150.1 [equine], XP\_044911834.1 [feline]), as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above act as ligands for the FGF receptors FGFR1, FGFR2, FGFR3 and FGFR4 in addition to strongly binding heparin and integrins. Additionally, FGF2 signaling is thought to impact localized nociception via at least its pro-angiogenic activity and has been implicated in pain perception related to at least IVD degeneration and at joint lesions. In some instances, and merely for the sake of disambiguation, a

prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

**[0111]** The term “FGFR1 gene” refers to a mammalian gene encoding a Fibroblast Growth Factor Receptor 1 polypeptide. Non-limiting examples of FGFR1 genes include: NCBI Gene ID: 2260 [human], NCBI Gene ID: 100057614 [equine], NCBI Gene ID: 101086055 [feline] as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an FGFR1 gene include: UniProt: P11362; NP\_001167534.1 [human], XP\_038545782.1 [canine], XP\_023486323.1 [equine], XP\_011279822.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above are tyrosine-protein kinases that act as cell-surface receptor for fibroblast growth factors. In that role, they play an essential role in the regulation of embryonic development, cell proliferation, differentiation and migration. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

**[0112]** The term “CCN2 gene” refers to a mammalian gene encoding a Cellular Communication Network Factor 2 polypeptide. Non-limiting examples of CCN2 genes include: NCBI Gene ID: 1490 [human], NCBI Gene ID: 476202 [canine], NCBI Gene ID: 100073098 [equine], NCBI Gene ID: 101094598 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CCN2 gene include: UniProt: P29279; NP\_001892.2 [human], XP\_038321343.1 [canine], XP\_023506869.1 [equine], XP\_023110145.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above are mitogens secreted by vascular endothelial cells and are related to chondrocyte proliferation and differentiation, cell adhesion in many cell types. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

**[0113]** The term “ADAMTS5 gene” refers to a mammalian gene encoding an ADAM Metallopeptidase with Thrombospondin Type 1 Motif 5 polypeptide. Non-limiting examples of ADAMTS5 genes include: NCBI Gene ID: 11096 [human], NCBI Gene ID: 487713 [canine], NCBI Gene ID: 100066005 [equine], NCBI Gene ID: 101085063 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an ADAMTS5 gene include: UniProt: Q9UNA0; NP\_008969.2 [human], XP\_038299214.1 [canine], XP\_023485737.1 [equine], XP\_023094603.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, members of the family share several distinct protein modules, including a propeptide region, a metalloproteinase domain, a disintegrin-like domain, and a thrombospondin type 1 (TS) motif with individual members of the family differing in the number of C-terminal TS motifs. ADAMTS5 has two unique C-terminal domains. Once proteolytically

processed to generate the mature enzyme. ADAMTS5 functions as an aggrecanase that cleaves aggrecan, a major proteoglycan of cartilage, and may mediate cartilage destruction in osteoarthritis. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0114] The term “ADAMTS1 gene” refers to a mammalian gene encoding an ADAM Metallopeptidase with Thrombospondin Type 1 Motif 1 polypeptide. Non-limiting examples of ADAMTS1 genes include: NCBI Gene ID: 9510 [human], NCBI Gene ID: 100686153 [canine], NCBI Gene ID: 791251 [equine], NCBI Gene ID: 101085309 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an ADAMTS1 gene include: UniProt: Q9UH18; NP\_008919.3 [human], XP\_038374156.1 [canine], XP\_023485736.1 [equine], XP\_019695041.3 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, members of the family share several distinct protein modules, including a propeptide region, a metalloproteinase domain, a disintegrin-like domain, and a thrombospondin type 1 (TS) motif with individual members of the family differing in the number of C-terminal TS motifs. ADAMTS1 contains two disintegrin loops and three C-terminal TS motifs. The protein has anti-angiogenic activity and functions as an aggrecanase that cleaves aggrecan, a major proteoglycan of cartilage, and may be involved in its turnover and has been associated with various inflammatory processes. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0115] The term “MMP1 gene” refers to a mammalian gene encoding a Matrix Metallopeptidase 1 polypeptide. Non-limiting examples of MMP1 genes include: NCBI Gene ID: 4312 [human], NCBI Gene ID: 489428 [canine], NCBI Gene ID: 100033896 [equine], NCBI Gene ID: 101084217 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an MMP1 gene include: UniProt: P03956; NP\_001139410.1 [human], XP\_038521018.1 [canine], NP\_001075316.1 [equine], XP\_003992365.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, MMP1 is proteolytically processed from a preproprotein to generate the mature protease. This secreted protease breaks down the interstitial collagens, including types I, II, and III. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0116] The term “MMP2 gene” refers to a mammalian gene encoding a Matrix Metallopeptidase 2 polypeptide. Non-limiting examples of MMP2 genes include: NCBI Gene ID: 4313 [human], NCBI Gene ID: 403733 [canine], NCBI Gene ID: 100033948 [equine], NCBI Gene ID: 101098838 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an MMP2 gene include: UniProt: P08253; NP\_001121363.1 [human],

XP\_038515255.1 [canine], XP\_023492775.1 [equine], XP\_003998091.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene belongs to the broader family of zinc-dependent enzymes that cleave components of the extracellular matrix. MMP2 is a gelatinase A, type IV collagenase, that contains three fibronectin type II repeats in its catalytic site that allow binding of denatured type IV and V collagen and elastin. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0117] The term “MMP3 gene” refers to a mammalian gene encoding a Matrix Metallopeptidase 3 polypeptide. Non-limiting examples of MMP3 genes include: NCBI Gene ID: 4314 [human], NCBI Gene ID: 403733 [canine], NCBI Gene ID: 100034195 [equine], NCBI Gene ID: 493666 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an MMP3 gene include: UniProt: P08254; NP\_002413.1 [human], NP\_001002967.1 [canine], NP\_001075964.1 [equine], XP\_003992356.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene belongs to the broader family of zinc-dependent enzymes that cleave components of the extracellular matrix. MMP3 is an enzyme that degrades fibronectin, laminin, collagens III, IV, IX, and X, and cartilage proteoglycans and is thought to be involved in wound repair, progression of atherosclerosis, and tumor initiation. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0118] The term “MMP7 gene” refers to a mammalian gene encoding a Matrix Metallopeptidase 7 polypeptide. Non-limiting examples of MMP7 genes include: NCBI Gene ID: 4316 [human], NCBI Gene ID: 489432 [canine], NCBI Gene ID: 100068985 [equine], NCBI Gene ID: 727698 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an MMP7 gene include: UniProt: P09237; NP\_002414.1 [human], NP\_001229655.1 [canine], XP\_001498859.1 [equine], XP\_003992352.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene belongs to the broader family of zinc-dependent enzymes that cleave components of the extracellular matrix. MMP7 is proteolytically processed to generate the mature protease, which breaks down proteoglycans, fibronectin, elastin and casein in addition to activating procollagenase. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0119] The term “MMP8 gene” refers to a mammalian gene encoding a Matrix Metallopeptidase 8 polypeptide. Non-limiting examples of MMP8 genes include: NCBI Gene ID: 4317 [human], NCBI Gene ID: 489429 [canine], NCBI Gene ID: 100069005 [equine], NCBI Gene ID: 101080995 [feline], as well as synonymous and non-syn-

onymous sequence variants thereof. Non-limiting examples of gene products encoded by an MMP8 gene include: UniProt: P22894; NP\_001291370.1 [human], XP\_038521019.1 [canine], XP\_005611595.1 [equine], XP\_003992354.3 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene belongs to the broader family of zinc-dependent enzymes that cleave components of the extracellular matrix. MMP8 is an enzyme that degrades interstitial collagens. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0120] The term “MMP10 gene” refers to a mammalian gene encoding a Matrix Metallopeptidase 10 polypeptide. Non-limiting examples of MMP10 genes include: NCBI Gene ID: 4319 [human], NCBI Gene ID: 100146442 [equine], NCBI Gene ID: 101081247 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an MMP10 gene include: UniProt: P09238; NP\_002416.1 [human], XP\_005614947.1 [equine], XP\_003992355.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene belongs to the broader family of zinc-dependent enzymes that cleave components of the extracellular matrix. MMP10 is an enzyme that degrades fibronectin, and type I, III, IV, and V gelatins. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0121] The term “MMP12 gene” refers to a mammalian gene encoding a Matrix Metallopeptidase 12 polypeptide. Non-limiting examples of MMP12 genes include: NCBI Gene ID: 4321 [human], NCBI Gene ID: 611789 [canine], NCBI Gene ID: 100069047 [equine], NCBI Gene ID: 101084472 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an MMP12 gene include: UniProt: P39900; NP\_002417.2 [human], NP\_001274067.1 [canine], XP\_001498924.2 [equine], XP\_003992366.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene belongs to the broader family of zinc-dependent enzymes that cleave components of the extracellular matrix. MMP12 is an enzyme with significant elastolytic activity and may be involved in tissue injury and remodeling. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0122] The term “MMP13 gene” refers to a mammalian gene encoding a Matrix Metallopeptidase 13 polypeptide. Non-limiting examples of MMP13 genes include: NCBI Gene ID: 4322 [human], NCBI Gene ID: 403763 [canine], NCBI Gene ID: 100009711 [equine], NCBI Gene ID: 493679 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an MMP13 gene include: UniProt: P45452; NP\_002418.1 [human], XP\_038521017.1 [canine], NP\_001075273.1 [equine], XP\_023094811.2 [feline],

as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene belongs to the broader family of zinc-dependent enzymes that cleave components of the extracellular matrix. MMP13 is an enzyme that degrades various types of collagen and has been implicated in wound healing, tissue remodeling, cartilage degradation, bone development, bone mineralization and ossification. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0123] The term “TIMP1 gene” refers to a mammalian gene encoding a TIMP Metallopeptidase Inhibitor 1 polypeptide. Non-limiting examples of TIMP1 genes include: NCBI Gene ID: 7076 [human], NCBI Gene ID: 403816 [canine], NCBI Gene ID: 100034220 [equine], NCBI Gene ID: 101095886 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TIMP1 gene include: UniProt: P01033; NP\_003245.1 [human], NP\_001003182.1 [canine], XP\_023488949.1 [equine], XP\_023105059.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene functions by forming one to one complexes with target metalloproteinases, such as collagenases, irreversibly inactivating through binding to their catalytic zinc cofactor. TIMP1 acts on MMP1, MMP2, MMP3, MMP7, MMP8, MMP9, MMP10, MMP11, MMP12, MMP13 and MMP16, but not on MMP14 and has been shown to act as a growth factor regulating cell differentiation, migration and cell death. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0124] The term “TIMP3 gene” refers to a mammalian gene encoding a TIMP Metallopeptidase Inhibitor 3 polypeptide. Non-limiting examples of TIMP3 genes include: NCBI Gene ID: 7078 [human], NCBI Gene ID: 481289 [canine], NCBI Gene ID: 100033947 [equine], NCBI Gene ID: 101091215 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TIMP3 gene include: UniProt: P35625; NP\_000353.1 [human], NP\_001271368.1 [canine], NP\_001075339.1 [equine], XP\_003989265.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene complexes with metalloproteinases (such as collagenases) to irreversibly inactivate them by binding to their catalytic zinc cofactor. TIMP3 is known to act on MMP1, MMP2, MMP3, MMP7, MMP9, MMP13, MMP14 and MMP15. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0125] The term “CXCL1 gene” refers to a mammalian gene encoding a C-X-C Motif Chemokine Ligand 1 polypeptide. Non-limiting examples of CXCL1 genes include: NCBI Gene ID: 2919 [human], NCBI Gene ID: 100034121 [equine], NCBI Gene ID: 102901432 [feline], as well as

synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CXCL1 gene include: UniProt: P09341; NP\_001502.1 [human], NP\_001296409.1 [equine], XP\_023108817.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene has chemotactic activity for neutrophils and may play a role in inflammation. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0126] The term "CXCL2 gene" refers to a mammalian gene encoding a C-X-C Motif Chemokine Ligand 2 polypeptide. Non-limiting examples of CXCL2 genes include: NCBI Gene ID: 2920 [human], NCBI Gene ID: 100233237 [equine], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CXCL2 gene include: UniProt: P19875, Q9UPB8; NP\_002080.1 [human], NP\_001137427.1 [equine], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene has antimicrobial function via its regulation of inflammatory and immunoregulatory processes. CXCL2 is expressed at the site of inflammation and has been shown to suppress proliferation of hematopoietic progenitor cells. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0127] The term "CXCL3 gene" refers to a mammalian gene encoding a C-X-C Motif Chemokine Ligand 3 polypeptide. Non-limiting examples of CXCL3 genes include: NCBI Gene ID: 2921 [human] NCBI Gene ID: 100056258 [equine], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CXCL3 gene include: UniProt: P19876, Q4W5H9; NP\_002081.2 [human], NP\_001137265.1 [equine], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is a secreted growth factor that signals through the G-protein coupled receptor, CXCR2 and plays a role in inflammation and as a chemoattractant for neutrophils. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0128] The term "CXCL5 gene" refers to a mammalian gene encoding a C-X-C Motif Chemokine Ligand 5 polypeptide. Non-limiting examples of CXCL5 genes include: NCBI Gene ID: 6374 [human], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CXCL5 gene include: UniProt: P19876, Q4W5H9; NP\_002081.2 [human], UniProt: P97885 [rat], UniProt: P50228 [mouse] as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is thought to interact with the G-protein coupled receptor, CXCR2 to promote angiogenesis, remodel connective tissues and recruit neutrophils. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a

particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0129] The term "CXCL6 gene" refers to a mammalian gene encoding a C-X-C Motif Chemokine Ligand 6 polypeptide. Non-limiting examples of CXCL6 genes include: NCBI Gene ID: 6372 [human], NCBI Gene ID: 106557449 [canine], NCBI Gene ID: 100033988 [equine], NCBI Gene ID: 101094593 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CXCL6 gene include: UniProt: P80162; NP\_002984.1 [human], XP\_038541813.1 [canine], NP\_001075355.2 [equine], XP\_003985379.3 [feline] as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is a chemotactic factor for neutrophils and exhibits antibacterial activity. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0130] The term "CXCL8 gene" refers to a mammalian gene encoding a C-X-C Motif Chemokine Ligand 8 polypeptide. Non-limiting examples of CXCL8 genes include: NCBI Gene ID: 3576 [human], NCBI Gene ID: 403850 [canine], NCBI Gene ID: 100037400 [equine], NCBI Gene ID: 493836 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CXCL8 gene include: UniProt: P10145; NP\_000575.1 [human], NP\_001003200.1 [canine], NP\_001077420.2 [equine], NP\_001009281.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is secreted by mononuclear macrophages, neutrophils, eosinophils, T lymphocytes, epithelial cells, and fibroblasts and functions as a chemotactic factor that guides neutrophils to the site of infection. CXCL8 also participates with other cytokines in the proinflammatory signaling cascade. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0131] The term "CCL2 gene" refers to a mammalian gene encoding a C-C Motif Chemokine Ligand 2 polypeptide. Non-limiting examples of CCL2 genes include: NCBI Gene ID: 6347 [human], NCBI Gene ID: 403981 [canine], NCBI Gene ID: 100034136 [equine], NCBI Gene ID: 100127112 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CCL2 gene include: UniProt: P13500; NP\_002973.1 [human], NP\_001003297.1 [canine], NP\_001075400.1 [equine], XP\_003996605.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene acts as a ligand for CCR2, which induces chemotactic activity for monocytes and basophils (but not neutrophils or eosinophils). In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0132] The term "CCL3 gene" refers to a mammalian gene encoding a C-C Motif Chemokine Ligand 3 polypeptide.

Non-limiting examples of CCL3 genes include: NCBI Gene ID: 6348 [human], NCBI Gene ID: 448787 [canine], NCBI Gene ID: 100057909 [equine], NCBI Gene ID: 100302540 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CCL3 gene include: UniProt: P10147; NP\_002974.1 [human], NP\_001005251.2 [canine], NP\_001108413.1 [equine], NP\_001157129.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene plays a role in inflammatory responses through binding to the receptors CCR1, CCR4 and CCR5. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0133] The term “CCL5 gene” refers to a mammalian gene encoding a C-C Motif Chemokine Ligand 5 polypeptide. Non-limiting examples of CCL5 genes include: NCBI Gene ID: 6352 [human], NCBI Gene ID: 403522 [canine], NCBI Gene ID: 100033925 [equine], NCBI Gene ID: 493689 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CCL5 gene include: UniProt: P13501; NP\_001265665.1 [human], NP\_001003010.1 [canine], NP\_001075332.1 [equine], NP\_001009827.1 [feline]) as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene functions as a chemoattractant for blood monocytes, memory T helper cells and eosinophils, induces the release of histamine from basophils, and activates eosinophils. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0134] The term “CCL7 gene” refers to a mammalian gene encoding a C-C Motif Chemokine Ligand 7 polypeptide. Non-limiting examples of CCL7 genes include: NCBI Gene ID: 6354 [human], NCBI Gene ID: 491148 [canine], NCBI Gene ID: 100071714 [equine], NCBI Gene ID: 101096931 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CCL7 gene include: UniProt: P80098; NP\_006264.2 [human], NP\_001010960.1 [canine], XP\_005597638.1 [equine], XP\_044900774.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is a secreted chemokine which attracts macrophages during inflammation and metastasis and is an in vivo substrate of MMP2. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0135] The term “CCL20 gene” refers to a mammalian gene encoding a C-C Motif Chemokine Ligand 20 polypeptide. Non-limiting examples of CCL20 genes include: NCBI Gene ID: 6364 [human], NCBI Gene ID: 448790 [canine], NCBI Gene ID: 100629808 [equine], NCBI Gene ID: 101089032 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CCL20 gene include: Uni-

Prot: P78556; NP\_001123518.1 [human], NP\_001005254.1 [canine], XP\_003365179.2 [equine], XP\_003991274.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is involved in inflammatory processes and displays chemotactic activity for lymphocytes. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0136] The term “CXCR1 gene” refers to a mammalian gene encoding a C-X-C Motif Chemokine Receptor 1 polypeptide. Non-limiting examples of CXCR1 genes include: NCBI Gene ID: 3577 [human], NCBI Gene ID: 478906 [canine], NCBI Gene ID: 100058291 [equine], NCBI Gene ID: 101085650 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CXCR1 gene include: UniProt: P25024; NP\_000625.1 [human], XP\_038303849.1 [canine], XP\_001491062.1 [equine], XP\_011283865.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is a receptor for IL8 and transduces signaling to mediate neutrophil migration to sites of inflammation, among other activities. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0137] The term “CXCR2 gene” refers to a mammalian gene encoding a C-X-C Motif Chemokine Receptor 2 polypeptide. Non-limiting examples of CXCR2 genes include: NCBI Gene ID: 3579 [human], NCBI Gene ID: 478905 [canine], NCBI Gene ID: 100055552 [equine], NCBI Gene ID: 101085396 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CXCR2 gene include: e.g., UniProt: P25025; NP\_001161770.1 [human], NP\_001003151.2 [canine], XP\_005610662.1 [equine], XP\_044890398.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is a receptor for IL8 and transduces signaling to mediate neutrophil migration to sites of inflammation, among other activities. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0138] The term “CCR7 gene” refers to a mammalian gene encoding a C-C Motif Chemokine Receptor 7 polypeptide. Non-limiting examples of CCR7 genes include: NCBI Gene ID: 1236 [human], NCBI Gene ID: 491011 [canine], NCBI Gene ID: 100067673 [equine], NCBI Gene ID: 101084327 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CCR7 gene include: UniProt: P32248; NP\_001288643.1 [human], XP\_038403305.1 [canine], XP\_001500231.1 [equine], XP\_003996882.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this

gene controls the migration of memory T cells to inflamed tissues, as well as stimulate dendritic cell maturation. Signals mediated by this receptor may also function in chronic inflammation pathogenesis. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0139] The term “ADAM17 gene” refers to a mammalian gene encoding an ADAM Metallopeptidase Domain 17 polypeptide. Non-limiting examples of ADAM17 genes include: NCBI Gene ID: 6868 [human], NCBI Gene ID: 475662 [canine], NCBI Gene ID: 100072496 [equine], NCBI Gene ID: 101089004 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a ADAM17 gene include: UniProt: P78536; NP\_001369706.1 [human], NP\_001273795.1 [canine], NP\_001295481.1 [equine], XP\_003984558.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is proteolytically processed to generate a mature protease, which functions by shedding the ectodomain of tumor necrosis factor-alpha, thereby releasing soluble tumor necrosis factor-alpha from its membrane-bound precursor. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0140] The term “TNF gene” refers to a mammalian gene encoding a Tumor Necrosis Factor polypeptide. Non-limiting examples of TNF genes include: NCBI Gene ID: 7124 [human], NCBI Gene ID: 403922 [canine], NCBI Gene ID: 100033834 [equine], NCBI Gene ID: 493755 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TNF gene include: UniProt: P01375; NP\_000585.2 [human], NP\_001003244.4 [canine], NP\_001075288.2 [equine], NP\_001009835.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a multifunctional proinflammatory cytokine that is mainly secreted by macrophages and can bind (and therefore function through) its receptors TNFRSF1A and TNFRSF1B. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0141] The term “TNFRSF1A gene” refers to a mammalian gene encoding a Tumor Necrosis Factor Receptor 1 polypeptide. Non-limiting examples of TNFRSF1A genes include: NCBI Gene ID: 7132 [human], NCBI Gene ID: 403634 [canine], NCBI Gene ID: 100059548 [equine], NCBI Gene ID: 493957 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TNFRSF1A gene include: UniProt: P19438; NP\_001056.1 [human], XP\_038295153.1 [canine], XP\_023498787.1 [equine], NP\_001009361.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above are transmembrane receptor proteins capable of binding Tumor Necrosis Factor Alpha (TNFA) or

lymphotoxin alpha (LTA), its principal ligand. Upon binding to TNFA, the receptor trimerizes and is activated, transmitting intracellular signaling cascades with role in various processes, including apoptosis and inflammation. See generally, Ward-Kavanagh, L. K., et al. (2016). *Immunity*, 44(5), 1005-1019. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0142] The term “TNFRSF1B gene” refers to a mammalian gene encoding a Tumor Necrosis Factor Receptor 2 polypeptide. Non-limiting examples of TNFRSF1B genes include: NCBI Gene ID: 7133 [human], NCBI Gene ID: 487437 [canine], NCBI Gene ID: 100055840 [equine], NCBI Gene ID: 101080392 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TNFRSF1B gene include: UniProt: P20333; XP\_011540362.1 [human], XP\_038387905.1 [canine], XP\_023491528.1 [equine], XP\_023113905.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above are transmembrane receptor proteins capable of binding TNFA or LTA and are implicated in pro-survival pathways through downstream activation of NFkB pathway. See generally, Ward-Kavanagh, L. K., et al. (2016). *Immunity*, 44(5), 1005-1019. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0143] The terms “IL4 gene” refers to a mammalian gene encoding an Interleukin 4 polypeptide. Non-limiting examples of IL4 genes include: NCBI Gene ID: 3565 [human], NCBI Gene ID: 403785 [canine], NCBI Gene ID: 100034225 [equine], NCBI Gene ID: 751514 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL4 gene include: UniProt: P05112; NP\_000580.1 [human], NP\_001003159.1 [canine], NP\_001075988.1 [equine], NP\_001036804.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a pleiotropic cytokine produced by activated T cells and is considered an important cytokine for tissue repair, counterbalancing the effects of proinflammatory type 1 cytokines, though it also promotes allergic airway inflammation and mediates acute inflammation, among other activities. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0144] The terms “IL4R gene” refers to a mammalian gene encoding an Interleukin 4 Receptor polypeptide. Non-limiting examples of IL4R genes include: NCBI Gene ID: 3566 [human], NCBI Gene ID: 489957 [canine], NCBI Gene ID: 791252 [equine], NCBI Gene ID: 101096277 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL4R gene include: UniProt: P24394; NP\_000409.1 [human], NP\_001003159.1 [canine], XP\_005598791.2 [equine], XP\_023102076.2 [feline], as

well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a type I transmembrane protein that can bind interleukin 4 and interleukin 13 to regulate IgE production and promote differentiation of Th2 cells, among other activities. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0145] The terms "IL6 gene" refers to a mammalian gene encoding an Interleukin 6 polypeptide. Non-limiting examples of IL6 genes include: NCBI Gene ID: 3569 [human], NCBI Gene ID: 403985 [canine], NCBI Gene ID: 100034196 [equine], NCBI Gene ID: 493687 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL6 gene include: UniProt: P05231; NP\_000591.1 [human], NP\_001003301.1 [canine], NP\_001075956.2 [equine], NP\_001009211.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a cytokine that functions in inflammation and the maturation of B cells that is primarily produced at sites of acute and chronic inflammation, where it is secreted into the serum and induces a transcriptional inflammatory response through interleukin 6 receptor. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0146] The term "IL6R gene" refers to a mammalian gene encoding an Interleukin-6 Receptor polypeptide. Non-limiting examples of IL6R genes include: NCBI Gene ID: 3560 [human], NCBI Gene ID: 612271 [canine], NCBI Gene ID: 102148787 [equine], NCBI Gene ID: 101085689 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL6R gene include: UniProt: P08887; CAA41231.1 [human], XP\_038527979.1 [canine], XP\_023496854.1 [equine], XP\_023103841.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above are transmembrane proteins capable of binding to interleukin-6, its native ligand. This binding event triggers intracellular signaling events that result in pro-inflammatory responses. See generally, Wolf, J., et al. (2014). *Cytokine*, 70(1), 11-20. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0147] The terms "IL6ST gene" refers to a mammalian gene encoding an Interleukin-6 Cytokine Family Signal Transducer polypeptide. Non-limiting examples of IL6ST genes include: NCBI Gene ID: 3572 [human], NCBI Gene ID: 403545 [canine], NCBI Gene ID: 100051700 [equine], NCBI Gene ID: 101089832 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an IL6ST gene include: UniProt: P40189; NP\_001177910.1 [human], NP\_001273950.1 [canine], XP\_023481030.1 [equine], XP\_011281205.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various gly-

coforms thereof. Canonically, the proteins encoded by the genes listed above are signal transducers shared by many cytokines, including interleukin 6 (IL6), ciliary neurotrophic factor (CNTF), leukemia inhibitory factor (LIF), and oncostatin M (OSM) and function as a part of the cytokine receptor complex. Activation of this protein is dependent upon the binding of cytokines to their receptors (e.g., IL6 to IL6R). Knockout studies in mice suggest that this gene plays a critical role in regulating myocyte apoptosis. See generally, Martinez-Perez, C., et al. (2021). *Journal of Personalized Medicine*, 11(7), 618. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0148] The terms "IL10 gene" refers to a mammalian gene encoding an Interleukin 10 polypeptide. Non-limiting examples of IL10 genes include: NCBI Gene ID: 3586 [human], NCBI Gene ID: 403628 [canine], NCBI Gene ID: 100034187 [equine], NCBI Gene ID: 493683 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL10 gene include: UniProt: P22301; NP\_000563.1 [human], NP\_001003077.1 [canine], NP\_001075959.1 [equine], NP\_001009209.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a pleiotropic cytokine that regulates inflammation and acts on many immune cell types through binding to its heterodimeric receptor composed of IL10RA and IL10RB, thereby activating downstream signaling cascades, such as the JAK-STAT pathway. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0149] The term "IL10RA gene" refers to a mammalian gene encoding a Interleukin 10 Receptor Alpha polypeptide. Non-limiting examples of IL10RA genes include: NCBI Gene ID: 3587 [human], NCBI Gene ID: 610823 [canine], NCBI Gene ID: 100071172 [equine], NCBI Gene ID: 101087601 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL10RA gene include: UniProt: Q13651; NP\_001549.2 [human], XP\_038520677.1 [canine], XP\_014596783.1 [equine], XP\_003992449.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is, upon forming a heterodimer with IL10RB, a regulator of pro-inflammatory signaling through the binding of its ligand IL-10. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0150] The term "IL10RB gene" refers to a mammalian gene encoding an Interleukin 10 Receptor Beta polypeptide. Non-limiting examples of IL10RB genes include: NCBI Gene ID: 3588 [human], NCBI Gene ID: 478404 [canine], NCBI Gene ID: 100052549 [equine], NCBI Gene ID: 101090038 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL10RB gene include:

UniProt: Q08334; NP\_000619.3 [human], XP\_038299308.1 [canine], XP\_023485821.1 [equine], XP\_003991512.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is, upon forming a heterodimer with IL10RA, a regulator of pro-inflammatory signaling through the binding of its ligand IL-10. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0151] The term "IL13 gene" refers to a mammalian gene encoding an Interleukin 13 polypeptide. Non-limiting examples of IL13 genes include: NCBI Gene ID: 3596 [human], NCBI Gene ID: 442990 [canine], NCBI Gene ID: 100034113 [equine], NCBI Gene ID: 101084678 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL13 gene include: UniProt: P35225; NP\_001341920.1 [human], NP\_001003384.1 [canine], NP\_001137263.1 [equine], NP\_001009209.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes regulates the production of pro-inflammatory cytokines and chemokines. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0152] The term "IL13RA1 gene" refers to a mammalian gene encoding an Interleukin 13 Receptor Alpha 1 polypeptide. Non-limiting examples of IL13RA1 genes include: NCBI Gene ID: 3597 [human], NCBI Gene ID: 403623 [canine], NCBI Gene ID: 100055312 [equine], NCBI Gene ID: 101091351 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL13RA1 gene include: UniProt: P78552; NP\_001551.1 [human], XP\_038306633.1 [canine], XP\_023490026.1 [equine], XP\_023104651.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a low affinity binding partner of IL13 and comprises a functional receptor once associated with of IL13RA2. Once bound to IL13, the receptor complex stimulates the production of pro-inflammatory cytokines and chemokines. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0153] The term "IL13RA2 gene" refers to a mammalian gene encoding an Interleukin 13 Receptor Alpha 2 polypeptide. Non-limiting examples of IL13RA2 genes include: NCBI Gene ID: 3598 [human], NCBI Gene ID: 403622 [canine], NCBI Gene ID: 100057673 [equine], NCBI Gene ID: 101100114 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL13RA2 gene include: UniProt: Q14627; NP\_000631.1 [human], NP\_001003075.1 [canine], XP\_023489189.1 [equine], XP\_044906881.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these

genes is a high affinity binding partner of IL13 but lacks a cytoplasmic domain. Along with IL13RA1, it forms a functional receptor that stimulates the production of pro-inflammatory cytokines and chemokines. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0154] The term "IL17A gene" refers to a mammalian gene encoding an Interleukin 17A polypeptide. Non-limiting examples of IL17A genes include: NCBI Gene ID: 3605 [human], NCBI Gene ID: 481837 [canine], NCBI Gene ID: 100034142 [equine], NCBI Gene ID: 101095339 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL17A gene include: UniProt: Q16552; NP\_002181.1 [human], NP\_001159350.1 [canine], NP\_001137264.1 [equine], XP\_006931878.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is an inflammatory cytokine that activates the NF kappa B signaling pathway through interactions with its heterodimeric receptor complex of IL17RA and IL17RC, thereby activating transcription of various chemokines, cytokines and other factors. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0155] The term "IL17RA gene" refers to a mammalian gene encoding an Interleukin 17 Receptor A polypeptide. Non-limiting examples of IL17RA genes include: NCBI Gene ID: 23765 [human], NCBI Gene ID: 486759 [canine], NCBI Gene ID: 100055511 [equine], NCBI Gene ID: 101095588 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL17RA gene include: UniProt: Q96F46; NP\_001276834.1 [human], XP\_038295433.1 [canine], XP\_005610881.1 [equine], XP\_023112364.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a transmembrane protein that binds to IL17A with low affinity as part of a multimeric receptor complex. With its ligand, IL17RA is implicated in many inflammatory conditions. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0156] The term "IL18 gene" refers to a mammalian gene encoding an Interleukin 18 polypeptide. Non-limiting examples of IL18 genes include: NCBI Gene ID: 3606 [human], NCBI Gene ID: 403796 [canine], NCBI Gene ID: 100034216 [equine], NCBI Gene ID: 493688 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL18 gene include: UniProt: Q14116; NP\_001230140.1 [human], XP\_038520002.1 [canine], XP\_005611483.1 [equine], NP\_001009213.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a pro-inflammatory cytokine that regulates inflammatory signaling through the NF kappa B pathway

when engaged with its receptor and co-receptor, IL18R1 and IL18RAP. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0157] The term “IL18R1 gene” refers to a mammalian gene encoding an Interleukin 18 Receptor 1 polypeptide. Non-limiting examples of IL18R1 genes include: NCBI Gene ID: 8809 [human], NCBI Gene ID: 611438 [canine], NCBI Gene ID: 100058269 [equine], NCBI Gene ID: 493938 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL18R1 gene include: UniProt: Q13478; NP\_001269328.1 [human], XP\_038536128.1 [canine], XP\_023474273.1 [equine], NP\_001009863.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is an essential component for transducing IL18-mediated pro-inflammatory signaling. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0158] The term “IL18RAP gene” refers to a mammalian gene encoding an Interleukin 18 Receptor Accessory Protein polypeptide. Non-limiting examples of IL18RAP genes include: NCBI Gene ID: 8807 [human], NCBI Gene ID: 481327 [canine], NCBI Gene ID: 100050212 [equine], NCBI Gene ID: 101084868 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL18RAP gene include: UniProt: Q53TU5; NP\_001380415.1 [human], XP\_038536125.1 [canine], XP\_014586460.1 [equine], XP\_019682529.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is an accessory protein that enhances the signal transduction of IL18-mediated pro-inflammatory signaling. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0159] The term “NGF gene” refers to a mammalian gene encoding a Nerve Growth Factor polypeptide. Non-limiting examples of NGF genes include: NCBI Gene ID: 4803 [human], NCBI Gene ID: 403402 [canine], NCBI Gene ID: 100065669 [equine], NCBI Gene ID: 100144611 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a NGF gene include: UniProt: P01138; NP\_002497.2 [human], XP\_038546347.1 [canine], XP\_001496237.2 [equine], XP\_044889256.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, this secreted protein forms a functional homodimer that is incorporated into a larger complex and has nerve growth stimulating activity. The complex is also involved in the regulation of growth and the differentiation of sympathetic and certain sensory neurons. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0160] The term “NGFR gene” refers to a mammalian gene encoding a Nerve Growth Factor Receptor polypeptide. Non-limiting examples of NGFR genes include: NCBI Gene ID: 4804 [human], NCBI Gene ID: 491071 [canine], NCBI Gene ID: 100069694 [equine], NCBI Gene ID: 101101519 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a NGFR gene include: UniProt: P08138; NP\_002498.1 [human], XP\_038531049.1 [canine], XP\_023508464.1 [equine], XP\_023099534.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes contains four 40-amino acid repeats within its extracellular domain with 6 cysteine residues at conserved positions followed by a serine/threonine-rich region. This cysteine-rich region contains the nerve growth factor binding domain and allows for signal transduction once bound. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0161] The term “NTF3 gene” refers to a mammalian gene encoding a Neurotrophin-3 polypeptide. Non-limiting examples of NTF3 genes include: NCBI Gene ID: 4908 [human], NCBI Gene ID: 493963 [canine], NCBI Gene ID: 100051839 [equine], NCBI Gene ID: 486731 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a NTF3 gene include: UniProt: P20783; NP\_001096124.1 [human], XP\_038293846.1 [canine], XP\_023498780.1 [equine], NP\_001009367.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes controls survival and differentiation of neurons. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0162] The term “NTF4 gene” refers to a mammalian gene encoding a Neurotrophin-4 polypeptide. Non-limiting examples of NTF4 genes include: NCBI Gene ID: 4909 [human], NCBI Gene ID: 611987 [canine], NCBI Gene ID: 100054859 [equine], NCBI Gene ID: 101100428 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a NTF4 gene include: UniProt: P34130; NP\_001382418.1 [human], NP\_001177358.2 [canine], XP\_023505846.1 [equine], XP\_023101354.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is proteolytically processed to a mature form, which can promote survival of neurons through binding of its cognate receptor. Dysregulation of this protein is observed in various neurological disorders. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0163] The term “NTRK1 gene” refers to a mammalian gene encoding a Neurotrophic Receptor Tyrosine Kinase 1 polypeptide. Non-limiting examples of NTRK1 genes include: NCBI Gene ID: 4914 [human], NCBI Gene ID: 490404 [canine], NCBI Gene ID: 100064594 [equine],

NCBI Gene ID: 101081603 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a NTRK1 gene include: UniProt: P04629; NP\_001007793.1 [human], XP\_038527745.1 [canine], XP\_023496742.1 [equine], XP\_023103311.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a membrane-bound receptor that binds neurotrophin and signals through the MAPK pathway to regulate cell differentiation, among other functions. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0164] The term “NTRK2 gene” refers to a mammalian gene encoding a Neurotrophic Receptor Tyrosine Kinase 2 polypeptide. Non-limiting examples of NTRK2 genes include: NCBI Gene ID: 4915 [human], NCBI Gene ID: 484147 [canine], NCBI Gene ID: 100061700 [equine], NCBI Gene ID: 101101347 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a NTRK2 gene include: UniProt: Q16620; NP\_001007098.1 [human], XP\_038510982.1 [canine], XP\_023482906.1 [equine], XP\_023097987.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a membrane-bound receptor that binds neurotrophin and signals through the MAPK pathway to regulate cell differentiation, among other functions. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0165] The term “BDNF gene” refers to a mammalian gene encoding a Brain-Derived Neurotrophic Factor polypeptide. Non-limiting examples of BDNF genes include: NCBI Gene ID: 627 [human], NCBI Gene ID: 403461 [canine], NCBI Gene ID: 100009689 [equine], NCBI Gene ID: 493690 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a BDNF gene include: UniProt: P23560; NP\_001137277.1 [human], NP\_001002975.1 [canine], NP\_001075256.1 [equine], NP\_001009828.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is proteolytically processed to a mature form, which can promote survival of neurons through binding of its cognate receptor. Dysregulation of this protein is observed in various neurological disorders. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0166] The terms “SCN1A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 1 polypeptide. Non-limiting examples of SCN1A genes include: NCBI Gene ID: 6323 [human], NCBI Gene ID: 478775 [canine], NCBI Gene ID: 100052059 [equine], NCBI Gene ID: 101081823 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN1A gene

include: UniProt: P35498; NP\_001159435.1 [human], XP\_038302870.1 [canine], XP\_023478839.1 [equine], XP\_019693764.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes mediates the voltage-dependent sodium ion permeability of excitable membranes and is involved in sensory perception of mechanical pain (i.e., activation in somatosensory neurons has been shown to induce pain without neurogenic inflammation). In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0167] The terms “SCN2A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 2 polypeptide. Non-limiting examples of SCN2A genes include: NCBI Gene ID: 6326 [human], NCBI Gene ID: 478773 [canine], NCBI Gene ID: 100051816 [equine], NCBI Gene ID: 101080472 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN2A gene include: UniProt: Q99250; NP\_001035232.1 [human], XP\_038302857.1 [canine], XP\_023478830.1 [equine], XP\_023115179.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes mediates the voltage-dependent sodium ion permeability of excitable membranes. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0168] The term “SCN3A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 3 polypeptide. Non-limiting examples of SCN3A genes include: NCBI Gene ID: 6328 [human], NCBI Gene ID: 478772 [canine], NCBI Gene ID: 100061941 [equine], NCBI Gene ID: 101082587 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN3A gene include: UniProt: Q9NY46; NP\_001075145.1 [human], XP\_038302852.1 [canine], XP\_023478823.1 [equine], XP\_019693750.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a subunit of voltage-gated sodium channels and is responsible for propagation of action potentials in neurons and muscle tissue. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0169] The terms “SCN4A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 4 polypeptide. Non-limiting examples of SCN4A genes include: NCBI Gene ID: 6328 [human], NCBI Gene ID: 119873250 [canine], NCBI Gene ID: 100049793 [equine], NCBI Gene ID: 101098669 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN4A gene include: UniProt: Q9NY46; NP\_001075145.1 [human], XP\_038531923.1 [canine], NP\_001075230.2 [equine], XP\_006940553.1 [feline], as well as sequence variants,

isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a subunit of voltage-gated sodium channels and is responsible for propagation of action potentials in neurons and muscle tissue. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0170] The terms “SCN5A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 5 polypeptide. Non-limiting examples of SCN5A genes include: NCBI Gene ID: 6331 [human], NCBI Gene ID: 403497 [canine], NCBI Gene ID: 100034027 [equine], NCBI Gene ID: 101100994 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN5A gene include: UniProt: Q14524; NP\_000326.2 [human], NP\_001002994.1 [canine], NP\_001157367.1 [equine], XP\_044893792.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a subunit of voltage-gated sodium channels and is found primarily in cardiac muscle and is responsible for the initial upstroke of the action potential in an electrocardiogram. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0171] The term “SCN8A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 8 polypeptide. Non-limiting examples of SCN8A genes include: NCBI Gene ID: 6335 [human], NCBI Gene ID: 477604 [canine], NCBI Gene ID: 100052777 [equine], NCBI Gene ID: 101096578 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN8A gene include: UniProt: Q9UQD0; NP\_001171455.1 [human], XP\_038294063.1 [canine], XP\_023499351.1 [equine], XP\_023112849.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is the ion pore subunit of the voltage-gated sodium channel and is essential for rapid membrane depolarization during neuronal action potentials. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0172] The term “SCN9A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 9 polypeptide. Non-limiting examples of SCN9A genes include: NCBI Gene ID: 6335 [human], NCBI Gene ID: 100855710 [canine], NCBI Gene ID: 100052120 [equine], NCBI Gene ID: 101082841 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN9A gene include: UniProt: Q15858; NP\_001352465.1 [human], XP\_038302872.1 [canine], XP\_023478844.1 [equine], XP\_044889827.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a voltage-dependent sodium ion channel that has been associated with various pain disorders, especially in the

development of inflammatory pain. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0173] The term “SCN10A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 10 polypeptide. Non-limiting examples of SCN10A genes include: NCBI Gene ID: 6336 [human], NCBI Gene ID: 477026 [canine], NCBI Gene ID: 100055493 [equine], NCBI Gene ID: 101085569 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN10A gene include: UniProt: Q9Y5Y9; NP\_001280235.2 [human], NP\_001003203.1 [canine], XP\_014587037.1 [equine], XP\_044893784.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a membrane-spanning subunit of voltage-dependent sodium channels that may be involved in the onset of pain associated with neuropathies. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0174] The term “SCN11A gene” refers to a mammalian gene encoding a Sodium Voltage-Gated Channel Alpha 11 polypeptide. Non-limiting examples of SCN11A genes include: NCBI Gene ID: 11280 [human], NCBI Gene ID: 485593 [canine], NCBI Gene ID: 100068480 [equine], NCBI Gene ID: 101085312 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a SCN11A gene include: UniProt: Q9UI33; NP\_001336182.1 [human], XP\_038426400.1 [canine], XP\_001916634.3 [equine], XP\_044893782.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a membrane-spanning subunit of voltage-dependent sodium channels and is highly expressed in nociceptive neurons of dorsal root ganglia and trigeminal ganglia. Mutations in the SCN11A gene have been associated with various pain disorders. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0175] The terms “TAC1 gene” refers to a mammalian gene encoding a Tachykinin Precursor 1 polypeptide. Non-limiting examples of TAC1 genes include: NCBI Gene ID: 6863 [human], NCBI Gene ID: 475239 [canine], NCBI Gene ID: 100052324 [equine], NCBI Gene ID: 101095481 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TAC1 gene include: UniProt: P20366; NP\_003173.1 [human], XP\_038541905.1 [canine], XP\_014594521.1 [equine], XP\_003982840.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is a precursor for four products of the tachykinin peptide hormone family-substance P, neuropeptide A, neuropeptide K and neuropeptide gamma. These hormones are thought to function as neurotransmitters that interact with nerve receptors and smooth

muscle cells. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0176] The terms “TAC3 gene” refers to a mammalian gene encoding a Tachykinin Precursor 3 polypeptide. Non-limiting examples of TAC3 genes include: NCBI Gene ID: 6866 [human], NCBI Gene ID: 607315 [canine], NCBI Gene ID: 100052722 [equine], NCBI Gene ID: 101089368 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TAC3 gene include: UniProt: Q9UHF0; NP\_001171525.1 [human], UniProt: A0A8I3N7Z8; NP\_001362511.2 canine], XP\_023499603.1 [equine], XP\_019690663.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is proteolytically processed to generate a mature peptide, which is primarily expressed in the central and peripheral nervous systems and functions as a neurotransmitter. This peptide is the ligand for the neurokinin-3 receptor. These hormones are thought to function as neurotransmitters that interact with nerve receptors and smooth muscle cells. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0177] The term “TACR1 gene” refers to a mammalian gene encoding a Tachykinin Receptor 1 polypeptide. Non-limiting examples of TACR1 genes include: NCBI Gene ID: 6869 [human], NCBI Gene ID: 403815 [canine], NCBI Gene ID: 100053491 [equine], NCBI Gene ID: 101090094 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TACR1 gene include: UniProt: P25103; NP\_001049.1 [human], NP\_001012637.1 canine], XP\_001499730.1 [equine], XP\_003984209.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is the receptor for the tachykinin substance P, also referred to as neurokinin 1. TACR1 activates a phosphatidylinositol-calcium second messenger system and can also bind substance K and neuromedin-K with less affinity. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0178] The term “TACR2 gene” refers to a mammalian gene encoding a Tachykinin Receptor 2 polypeptide. Non-limiting examples of TACR2 genes include: NCBI Gene ID: 6865 [human], NCBI Gene ID: 489020 [canine], NCBI Gene ID: 100034168 [equine], NCBI Gene ID: 101094541 [feline]], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TACR2 gene include: UniProt: P21452; NP\_001048.2 [human], NP\_001012635.1 [canine], XP\_001502752.2 [equine], XP\_044896003.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is the receptor for the tachykinin substance K, also referred to as neurokinin A.

TACR2 activates a phosphatidylinositol-calcium second messenger system and can also bind neuromedin-K and substance P with less affinity. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0179] The term “TACR3 gene” refers to a mammalian gene encoding a Tachykinin Receptor 3 polypeptide. Non-limiting examples of TACR3 genes include: NCBI Gene ID: 6870 [human], NCBI Gene ID: 403814 [canine], NCBI Gene ID: 100073088 [equine], NCBI Gene ID: 101093603 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a TACR3 gene include: UniProt: P29371; NP\_001050.1 [human], NP\_001091010.1 [canine], XP\_023492571.1 [equine], XP\_003985169.3 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is the receptor for the tachykinin neurokinin 3, also referred to as neurokinin B or neuromedin-K. TACR3 activates a phosphatidylinositol-calcium second messenger system and can also bind substance K and substance P with less affinity. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0180] The term “MRGPRX2 gene” refers to a mammalian gene encoding a MAS related GPR family member X2 polypeptide. Non-limiting examples of MRGPRX2 genes include: NCBI Gene ID: 117194 [human], NCBI Gene ID: 485410 [canine], NCBI Gene ID: 100071950 [equine], NCBI Gene ID: 101097092 [feline]) or an encoded gene product (e.g., UniProt: Q96LB1; NP\_001290544.1 [human], XP\_032825538.1 [canine], XP\_023501936.1 [equine], XP\_003993155.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes enables G protein-coupled receptor activity and neuropeptide binding activity and is involved in mast cell degranulation and positive regulation of cytokinesis. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0181] The term “ATP1A1 gene” refers to a mammalian gene encoding a ATPase Na+/K+ transporting subunit alpha 1 polypeptide. Non-limiting examples of ATP1A1 genes include: NCBI Gene ID: 476 [human], NCBI Gene ID: 403992 [canine], NCBI Gene ID: 100034139 [equine], NCBI Gene ID: 101083695 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a ATP1A1 gene include: UniProt: P05023; NP\_000692.2 [human], NP\_001376153.1 [canine], NP\_001108004.2 [equine], XP\_011283388.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is an integral membrane protein subunit of the complex responsible for establishing and maintaining the electrochemical gradients of Na and K ions across a plasma membrane, which is essential for osmoregulation and electrical excitability of nerve and muscle. In some instances,

and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0182] The term “CALCA gene” refers to a mammalian gene encoding a Calcitonin Related Polypeptide Alpha polypeptide. Non-limiting examples of CALCA genes include: NCBI Gene ID: 796 [human], NCBI Gene ID: 403946 [canine], NCBI Gene ID: 100033906 [equine], NCBI Gene ID: 101095582 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CALCA gene include: UniProt: P01258; NP\_001029124.1 [human], NP\_001300719.1 [canine], NP\_001075323.1 [equine], XP\_019667660.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, this gene encodes multiple gene products, such as calcitonin, calcitonin gene-related peptide and katacalcin, through tissue-specific alternative RNA splicing of the gene transcripts and cleavage of inactive precursor proteins. The proteins are involved in calcium regulation, regulate phosphorus metabolism, and function as a vasodilator, among other functions. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0183] The term “CALCB gene” refers to a mammalian gene encoding a Calcitonin Related Polypeptide Beta polypeptide. Non-limiting examples of CALCB genes include: NCBI Gene ID: 797 [human], NCBI Gene ID: 403415 [canine], NCBI Gene ID: 100034126 [equine], NCBI Gene ID: 101094539 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CALCB gene include: UniProt: P10092; NP\_000719.1 [human], NP\_001002948.1 [canine], NP\_001075397.1 [equine], XP\_044894937.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes acts as a vasodilator and a neurotransmitter, among other functions. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0184] The terms “CALCRL gene” refers to a mammalian gene encoding a Calcitonin Receptor Like Receptor polypeptide. Non-limiting examples of CALCRL genes include: NCBI Gene ID: 10203 [human], NCBI Gene ID: 488438 [canine], NCBI Gene ID: 100054281 [equine], NCBI Gene ID: 101086333 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CALCRL gene include: UniProt: Q16602; NP\_001258680.1 [human], XP\_038303202.1 [canine], XP\_023477941.1 [equine], XP\_011283721.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes comprises the receptor for CGRP (with RAMP1) and receptor for ADM (with RAMP2/3) and activates adenylyl cyclase. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the

protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0185] The term “RAMP1 gene” refers to a mammalian gene encoding a Receptor Activity Modifying Protein 1 polypeptide. Non-limiting examples of RAMP1 genes include: NCBI Gene ID: 10267 [human], NCBI Gene ID: 607163 [canine], NCBI Gene ID: 100066550 [equine], NCBI Gene ID: 101092133 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a RAMP1 gene include: UniProt: 060894; NP\_005846.1 [human], XP\_038291846.1 [canine], XP\_023498460.1 [equine], XP\_044890618.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by these genes is required to transport calcitonin-receptor-like receptor (CRLR) to the plasma membrane and, with CRLR, functions as a CGRP receptor. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0186] The term “ADM gene” refers to a mammalian gene encoding an Adrenomedullin polypeptide. Non-limiting examples of ADM genes include: NCBI Gene ID: 133 [human], NCBI Gene ID: 403817 [canine], NCBI Gene ID: 100033857 [equine], NCBI Gene ID: 101087095 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a ADM gene include: UniProt: P35318; NP\_001115.1 [human], NP\_001003183.1 [canine], NP\_001157351.1 [equine], XP\_044894880.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is a 52 aa peptide with several functions, including vasodilation, regulation of hormone secretion, promotion of angiogenesis. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0187] The term “CRCP gene” refers to a mammalian gene encoding a CGRP Receptor Component polypeptide. Non-limiting examples of CRCP genes include: NCBI Gene ID: 27297 [human], NCBI Gene ID: 479705 [canine], NCBI Gene ID: 100061681 [equine], NCBI Gene ID: 101084503 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a CRCP gene include: UniProt: 075575; NP\_001035737.1 [human], XP\_038523718.1 [canine], XP\_001493592.3 [equine], XP\_044903465.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is an accessory protein for the CGRP receptor that modulates CGRP responsiveness in a variety of tissues. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0188] The term “YAP1 gene” refers to a mammalian gene encoding a Yes1-Associated Protein polypeptide. Non-limiting examples of YAP1 genes include: NCBI Gene ID:

10413 [human], NCBI Gene ID: 479465 [canine], NCBI Gene ID: 100068834 [equine], NCBI Gene ID: 101101408 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a YAP1 gene include: UniProt: P46937; NP\_001123617.1 [human], XP\_038521022.1 [canine], XP\_023500466.1 [equine], XP\_044894121.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by this gene is involved in development, growth, repair and homeostasis. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0189] The term “IL1RAP gene” refers to a mammalian gene encoding an Interleukin 1 Receptor Accessory Protein polypeptide. Non-limiting examples of IL1RAP genes include: NCBI Gene ID: 3556 [human], NCBI Gene ID: 488126 [canine], NCBI Gene ID: 100068726 [equine], NCBI Gene ID: 101094125 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL1RAP gene include: UniProt: Q9NPH3; NP\_002173.1 [human], XP\_038318680.1 [canine], XP\_001498597.2 [equine], XP\_044893081.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above are capable of associating with IL1R1 bound to IL1 to form the high affinity interleukin-1 receptor complex that mediates interleukin-1-dependent activation of NF-kappa-B and other signaling pathways through the recruitment of adapter molecules such as TOLL1P, MYD88, and IRAK1 or IRAK2 via TIR-TIR interactions with the cytoplasmic domains of receptor/coreceptor subunits. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f, referring to the human, canine, equine, and feline forms, respectively).

[0190] The term “IL1R1 gene” refers to a mammalian gene encoding an Interleukin 1 receptor type 1 polypeptide. Non-limiting examples of IL1R1 genes include: NCBI Gene ID: 3554 [human], NCBI Gene ID: 481328 [canine], NCBI Gene ID: 100009699 [equine], NCBI Gene ID: 101080705 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a ILR1 gene include: UniProt: P14778; NP\_001307909.1 [human], XP\_038536135.1 [canine], NP\_001075263.2 [equine], XP\_023107327.2 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above are capable of binding all forms of the pro-inflammatory cytokine interleukin 1 (IL1 or IL1) to mediate interleukin-1-dependent activation of NF-kappa-B, MAPK and other signaling pathways. This intracellular signaling involves the recruitment of adapter molecules such as TOLLIP, MYD88, and IRAK1 or IRAK2 via TIR-TIR interactions with the cytoplasmic domains of receptor/coreceptor subunits. IL1R1 can also bind the Interleukin 1 receptor antagonist (IL1Ra or IL1Ra or IL1RN), which prevents association with IL1RAP to form a signaling-competent complex. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species

(with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0191] The term “IL1A gene” refers to a mammalian gene encoding a Interleukin 1 Alpha polypeptide. Non-limiting examples of IL1A genes include: NCBI Gene ID: 3552 [human], NCBI Gene ID: 403782 [canine], NCBI Gene ID: 100064969 [equine], NCBI Gene ID: 493944 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by a IL1A gene include: UniProt: P01583; NP\_000566.3 [human], NP\_001003157.2 [canine], NP\_001075969.2 [equine], NP\_001009351.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the proteins encoded by the genes listed above are pro-inflammatory cytokines that signal through interaction with IL1R1 and IL1RAP to activate various pathways, including MAPK, JNK and NF-kappa B. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0192] The terms “IL1B gene” refers to a mammalian gene encoding an Interleukin 1 Beta polypeptide. Non-limiting examples of IL1B genes include: NCBI Gene ID: 3553 [human], NCBI Gene ID: 403974 [canine], NCBI Gene ID: 100034237 [equine], NCBI Gene ID: 768274 [feline], as well as synonymous and non-synonymous sequence variants thereof. Non-limiting examples of gene products encoded by an IL1B gene include: UniProt: P01584; NP\_000567.1 [human], NP\_001033060.1 [canine], NP\_001075995.1 [equine], NP\_001070882.1 [feline], as well as sequence variants, isoforms encoded by alternative splicing, and various glycoforms thereof. Canonically, the protein encoded by the genes listed above is a major mediator of the inflammatory response and pyrogen that signals through interaction with IL1R1 and IL1RAP. In the central nervous system (CNS) IL1B has been shown to contribute to inflammatory pain hypersensitivity, among other pathologies. In some instances, and merely for the sake of disambiguation, a prefix is added when referring to the protein or gene of a particular species (with h, c, e, and f referring to the human, canine, equine, and feline forms, respectively).

[0193] The term “treatment” refers to obtaining a desired pharmacologic and/or physiologic effect. The effect may be prophylactic in terms of completely or partially preventing a disease or symptom thereof and/or may be therapeutic in terms of a partial or complete cure for a disease and/or adverse effect attributable to the disease. For example, a composition, method, or system of the present disclosure may be administered as a prophylactic treatment to a subject that has a predisposition for a given condition (e.g., arthritis). “Treatment”, as used herein, covers any treatment of a disease in a mammal, particularly in a human, canine, feline, or equine, and includes: (a) preventing the disease from occurring in a subject which may be predisposed to the disease but has not yet been diagnosed as having it; (b) inhibiting the disease, i.e., arresting its development or progression; and (c) relieving the disease, i.e., causing regression of the disease and/or relieving one or more disease symptoms.

[0194] “Treatment” is also meant to encompass delivery of an agent in order to provide for a pharmacologic effect,

even in the absence of a disease or condition. For example, “treatment” encompasses delivery of a composition that can elicit an immune response or confer immunity in the absence of a disease condition, e.g., in the case of a vaccine. It is understood that compositions and methods of the present disclosure are applicable to treat all mammals, including, but not limited to human, canine, feline, equine, and bovine subjects.

[0195] The term “therapeutically effective” refers to the amount of a composition or combination of compositions as described herein that is sufficient to effect the intended application including, but not limited to, disease treatment. A therapeutically effective amount may vary depending upon the intended application (in vitro or in vivo), or the subject and disease condition being treated (e.g., the weight, age and gender of the subject), the severity of the disease condition, or the manner of administration. The term also applies to a dose that will induce a particular response in target cells (e.g., the reduction of platelet adhesion and/or cell migration). The specific dose will vary depending on the particular composition(s) chosen, the dosing regimen to be followed, whether the composition is administered in combination with other compositions or compounds, timing of administration, the tissue to which it is administered, and the physical delivery system in which the composition is carried.

[0196] A “spinal condition or disorder” includes, but is not limited to, low back pain, neck pain, discogenic disorders, adolescent idiopathic scoliosis, adult degenerative scoliosis, cervical degenerative disc disease, cervical disc herniation, cervical myelopathy, cervical stenosis, compression fractures, degenerative spondylolisthesis, isthmic spondylolisthesis, low back sprains and strains, lumbar degenerative disc disease, lumbar disc herniation, lumbar stenosis, neck sprain (whiplash) and strain, neck strain, osteoporosis, and whiplash. Generally, such disorders or conditions contribute to or cause localized nociception, inflammation, or morphological changes (e.g., fibrosis, degeneration, osteolysis, osteogenesis) at the cervical, thoracic, lumbar or sacral spine, or surrounding tissues.

[0197] “Low back pain” is defined as measurable or discernible pain or discomfort (either chronic or sporadic) in a given subject, encompassing at least the lumbar-spinal region of a mammal. The pain may present as being localized to the lower back (e.g., muscle ache) or as shooting, burning, stinging, and/or radiating sensations throughout the subject’s back and/or extremities. The pain may be idiopathic or may be associated with one or more (diagnosed or undiagnosed) underlying conditions including, but not limited to degenerative disc disease, chronic inflammation, arthritis, osteoporosis, trauma (e.g., post-surgical), infection (e.g., discospondylitis), neuropathies, musculo-skeletal abnormalities (e.g., slipped discs or spinal stenosis or spondylolisthesis), herniated nucleus pulposus (HNP), annular ligament tears, facet joint arthritis, radicular nerve compression, and/or other degenerative disorders.

[0198] “Neck pain” is defined as measurable or discernible pain or discomfort associated with the cervical spine or adjacent ligaments, muscles, and/or tendons. The pain may manifest as localized pain in the neck or shooting, stinging, burning, and/or radiating sensations throughout the back or extremities, including, but not limited to, the subject’s head, shoulders, arms, legs, and/or back. Neck pain may be idiopathic or associated with one or more (diagnosed or

undiagnosed) underlying conditions, including, but not limited to, degenerative disc disease, rheumatoid arthritis, osteoporosis, fibromyalgia, chronic inflammation, infection (e.g., discospondylitis), herniated disc, spondylosis, spinal stenosis, cervical compressive myelopathy, whiplash, and/or other disorders.

[0199] The terms “polynucleotide,” “nucleotide,” and “nucleic acid” are used interchangeably herein to refer to all forms of nucleic acid, oligonucleotides, including deoxyribonucleic acid (DNA) and ribonucleic acid (RNA). Polynucleotides include genomic DNA, cDNA and antisense DNA, and spliced or unspliced mRNA, rRNA, tRNA, lncRNA, RNA antagonists, and inhibitory DNA or RNA (RNAi, e.g., small or short hairpin (sh)RNA, microRNA (miRNA), aptamers, small or short interfering (si)RNA, trans-splicing RNA, or antisense RNA). Polynucleotides also include non-coding RNA, which include for example, but are not limited to, RNAi, miRNAs, lncRNAs, RNA antagonists, aptamers, and any other non-coding RNAs known to those of skill in the art. Polynucleotides include naturally occurring, synthetic, and intentionally altered or modified polynucleotides as well as analogues and derivatives. The term “polynucleotide” also refers to a polymeric form of nucleotides of any length, including deoxyribonucleotides or ribonucleotides, or analogs thereof, and is synonymous with nucleic acid sequence. A polynucleotide may comprise modified nucleotides, such as methylated nucleotides and nucleotide analogs, and may be interrupted by non-nucleotide components. If present, modifications to the nucleotide structure may be imparted before or after assembly of the polymer. The term polynucleotide, as used herein, refers interchangeably to double- and single-stranded molecules. Unless otherwise specified or required, any embodiment as described herein encompassing a polynucleotide encompasses both the double-stranded form and each of two complementary single-stranded forms known or predicted to make up the double-stranded form. Polynucleotides can be single, double, or triplex, linear or circular, and can be of any length. In discussing polynucleotides, a sequence or structure of a particular polynucleotide may be described herein according to the convention of providing the sequence in the 5' to 3' direction.

[0200] The term “gene” or “nucleotide sequence encoding a polypeptide” refers to the segment of DNA involved in producing a polypeptide chain. The DNA segment may include regions preceding and following the coding region (leader and trailer) involved in the transcription/translation of the gene product and the regulation of the transcription/translation, as well as intervening sequences (introns) between individual coding segments (exons). For example, a gene includes a polynucleotide containing at least one open reading frame capable of encoding a particular protein or polypeptide after being transcribed and translated.

[0201] The terms “extracellular domain” and “ectodomain” may be used interchangeably and, when referring to transmembrane cellular receptors, is defined as the portion of the protein that is exposed to the extracellular environment and is able to engage with and/or bind a ligand.

[0202] The terms “cytoplasmic domain” and “intracellular domain” may be used interchangeably and, when referring to transmembrane receptors, define the portion of the protein that is exposed to the cytoplasm. In many instances, these portions of the proteins comprise signaling domains to recruit and associate with various intracellular factors. Fol-

lowing engagement with a ligand via the extracellular domain, the interaction effects changes that may result in new association, dissociation or recruitment of various cytoplasmic factors that aid in transducing a signal.

**[0203]** The term “transmembrane domain,” which may be abbreviated as “TM,” as it refers to transmembrane receptors, is defined as the portion of the protein is embedded within the plasma membrane (i.e., not exposed to either the extracellular environment or the cytosol). Transmembrane domains are generally of a more hydrophobic character than either the extracellular or cytoplasmic portions and often adopt higher order helical structures. Though its primary role is an anchor, ligand-induced conformational changes to particular receptors have been shown to impact the transmembrane domain such that it is integral to the subsequent intracellular signaling.

**[0204]** The term “receptor” refers to a protein capable of binding another cognate protein (i.e., its ligand) with high affinity. This receptor-ligand interaction may be 1:1, or result in multimerization, wherein numerous proteins aggregate to bind one or more ligands. Receptors are generally present at the cell surface, such that they may most efficiently encounter a ligand and initiate intracellular signaling.

**[0205]** The term “intracellular signaling” refers to cellular changes that result due to events occurring at the cell surface. Typically, a soluble ligand binds its receptor at the cell surface, which can induce changes in the receptor, such that associated intracellular factors are also affected. These factors may then impact others within the cell, and this cascade continues until, in many cases, a particular factor is able to alter gene expression in the nucleus in response to the stimulus at the surface.

**[0206]** The term “RNA-guided nuclease” refers to an enzyme capable of breaking the backbone of, for example, a DNA molecule. The activity of RNA-guided nucleases is directed by a nucleic acid molecule (i.e., guide RNA). Once properly oriented to form a functional ribonucleoprotein complex, the enzyme locates a specific position within a target nucleic acid (e.g., a gene or locus) via sequence complementarity with a portion of the guide RNA. Non-exhaustive examples of RNA-guided nucleases include Cas9, Cas12 and Cas12a (previously known as Cpf1).

**[0207]** The term “Cas9” refers to an RNA-guided, double-stranded DNA-binding nuclease protein or nickase protein, or a variant thereof and may be used to refer to either naturally-occurring or recombinant Cas9 nucleases variants (e.g., ES-Cas9, HF-Cas9, PE-Cas9, and AR-Cas9). The wildtype Cas9 nuclease has two functional domains, e.g., RuvC and HNH, that simultaneously cut both strands of double stranded DNA, resulting in a double-strand break. Cas9 enzymes described herein may comprise a HNH or HNH-like nuclease domain and/or a RuvC or RuvC-like nuclease domain without impacts on the ability to induce double-strand breaks in genomic DNA (e.g., at a target locus) when both functional domains are active. The Cas9 enzyme may comprise one or more catalytic domains of a Cas9 protein derived from bacteria belonging to the group consisting of *Corynebacter*, *Sutterella*, *Legionella*, *Treponema*, *Filifactor*, *Eubacterium*, *Streptococcus*, *Lactobacillus*, *Mycoplasma*, *Bacteroides*, *Flavivola*, *Flavobacterium*, *Sphaerochaeta*, *Azospirillum*, *Gluconacetobacter*, *Neisseria*, *Roseburia*, *Parvibaculum*, *Staphylococcus*,

*Nitratifractor*, and *Campylobacter*. In some embodiments, the two catalytic domains are derived from different bacteria species.

**[0208]** As used herein, “PAM” refers to a Protospacer Adjacent Motif and is necessary for an RNA-guided nuclease to bind a target nucleic acid. In many instances, the PAM directly abuts the complementary sequence in the target. Naturally occurring Cas9, for example, molecules recognize specific PAM sequences (see, e.g., Table 1). In some embodiments, a Cas9 molecule has the same PAM specificities as a naturally occurring Cas9 molecule. In other embodiments, a Cas9 molecule has a PAM specificity not associated with a naturally occurring Cas9 molecule. In other embodiments, a Cas9 molecule’s PAM specificity is not associated with the naturally occurring Cas9 molecule to which it has the closest sequence homology. For example, a naturally occurring Cas9 molecule can be altered such that the PAM sequence recognition is altered to decrease off target sites, improve specificity, or eliminate a PAM recognition requirement. In an embodiment, a Cas9 molecule may be altered (e.g., to lengthen a PAM recognition sequence, improve Cas9 specificity to high level of identity, to decrease off target sites, and/or increase specificity). In an embodiment, the length of the PAM recognition sequence is at least 4, 5, 6, 7, 8, 9, 10 or 15 amino acids in length. In some embodiments, a Cas9 molecule may be altered to ablate PAM recognition.

**[0209]** The terms “guide RNA,” “gRNA” or “sgRNA” may be used interchangeably and refer to an RNA molecule, preferably a synthetic RNA molecule, composed of a targeting (crRNA) sequence and scaffold. These molecules, once loaded onto a functional RNA-guided nuclease can direct sequence-specific cleavage of a target nucleic acid.

**[0210]** An sgRNA can be administered or formulated, e.g., as a synthetic RNA, or as a nucleic acid comprising a sequence encoding the gRNA, which is then expressed in the target cells. As would be evident to one of ordinary skill in the art, various tools may be used in the design and/or optimization of an sgRNA in order to, for example, increase specificity and/or precision of genomic editing at a particular site.

**[0211]** In general, candidate sgRNAs may be designed and identified by first locating suitable PAMs within a genomic sequence. Then additional calculations may be utilized to predict on-target and off-target efficiencies. Available web-based tools to aid in the initial design and modeling of candidate sgRNAs include, without limitation, CRISPR-seek, CRISPR Design Tool, Cas-OFFinder, E-CRISP, ChopChop, CasOT, CRISPR direct, CRISPOR, BREAKING-CAS, CrispRGold, and CCTop. See, e.g., Safari, F. et al. (2017). Current Pharmaceutical Biotechnology, 18(13): 1038-54, which is incorporated by reference herein in its entirety for all purposes. Such tools are also described, for example, in PCT Publication No. WO2014093701A1 and Liu, G. et al. (2020). Computational approaches for effective CRISPR guide RNA design and evaluation. Computational and Structural Biotechnology Journal, 18: 35-44, each of which is incorporated by reference herein in its entirety for all purposes. Candidate sgRNAs may be further assessed by experimental screening or other methodologies.

**[0212]** The terms “CRISPR RNA” or “crRNA” refer to the portion of an sgRNA molecule with complementarity to the target nucleic acid.

[0213] The phrase “pharmaceutically acceptable” refers to those compounds, materials, compositions, and/or dosage forms that are, within the scope of sound medical judgment, suitable for use in contact with the tissues of human beings and animals without excessive toxicity, irritation, allergic response, or other problems or complications commensurate with a reasonable benefit/risk ratio.

[0214] The terms “pharmaceutically acceptable carrier” or “pharmaceutically acceptable excipient” are intended to include any and all solvents, dispersion media, coatings, antibacterial and antifungal agents, isotonic and absorption delaying agents, and inert ingredients. The use of such pharmaceutically acceptable carriers or pharmaceutically acceptable excipients for active pharmaceutical ingredients is well known in the art. Except insofar as any conventional pharmaceutically acceptable carrier or pharmaceutically acceptable excipient is incompatible with the active pharmaceutical ingredient, its use in the therapeutic compositions of the disclosure is contemplated. Additional active pharmaceutical ingredients, such as other drugs, can also be incorporated into the described compositions and methods.

[0215] The term “pharmaceutically acceptable excipient” is intended to include vehicles and carriers capable of being co-administered with a compound to facilitate the performance of its intended function. The use of such media for pharmaceutically active substances is well known in the art. Examples of such vehicles and carriers include solutions, solvents, dispersion media, delay agents, emulsions and the like. Any other conventional carrier suitable for use with the multi-binding compounds also falls within the scope of the present disclosure.

[0216] As used herein, the term “a”, “an”, or “the” generally is construed to cover both the singular and the plural forms.

[0217] The terms “about” and “approximately” mean within a statistically meaningful range of a value. Such a range can be within an order of magnitude, preferably within 50%, more preferably within 20%, more preferably still within 10%, and even more preferably within 5% of a given value or range. The allowable variation encompassed by the terms “about” or “approximately” depends on the particular system under study, and can be readily appreciated by one of ordinary skill in the art. Moreover, as used herein, the terms “about” and “approximately” mean that compositions, amounts, formulations, parameters, shapes and other quantities and characteristics are not and need not be exact, but may be approximate and/or larger or smaller, as desired, reflecting tolerances, conversion factors, rounding off, measurement error and the like, and other factors known to those of skill in the art. In general, a dimension, size, formulation, parameter, shape or other quantity or characteristic is “about” or “approximate,” whether or not expressly stated to be such. It is noted that embodiments of very different sizes, shapes and dimensions may employ the described arrangements.

[0218] The term “substantially” as used herein can refer to a majority of, or mostly, as in at least about 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, 99%, 99.5%, 99.9%, 99.99%, or at least about 99.999% or more.

[0219] The transitional terms “comprising,” “consisting essentially of,” and “consisting of,” when used in the appended claims, in original and amended form, define the claim scope with respect to what unrecited additional claim elements or steps, if any, are excluded from the scope of the

claim(s). The term “comprising” is intended to be inclusive or open-ended and does not exclude any additional, unrecited element, method, step or material. The term “consisting of” excludes any element, step or material other than those specified in the claim and, in the latter instance, impurities ordinary associated with the specified material(s). The term “consisting essentially of” limits the scope of a claim to the specified elements, steps or material(s) and those that do not materially affect the basic and novel characteristic(s) of the claimed methods and compositions. All compositions, methods, and kits described herein that embody the present disclosure can, in alternate embodiments, be more specifically defined by any of the transitional terms “comprising,” “consisting essentially of,” and “consisting of.”

[0220] As used herein, the term “delivering” means providing an entity to a destination. For example, delivering a therapeutic and/or prophylactic to a subject may involve administering a nanoparticle composition including the therapeutic and/or prophylactic to the subject (e.g., by an intravenous, intramuscular, intradermal, subcutaneous, intraarticular, or intradiscal route). Administration of a nanoparticle composition to a mammal or mammalian cell may involve contacting one or more cells with the nanoparticle composition.

[0221] As used herein, “naturally occurring” means existing in nature without artificial aid.

[0222] As used herein, a “PEG lipid” or “PEGylated lipid” refers to a lipid comprising a polyethylene glycol component. These lipids may also be referred to a PEG-modified lipids.

[0223] As used herein, a “phospholipid” is a lipid that includes a phosphate moiety and one or more carbon chains, such as unsaturated fatty acid chains. A phospholipid may include one or more multiple (e.g., double or triple) bonds (e.g., one or more unsaturations). Particular phospholipids may facilitate fusion to a membrane. For example, a cationic phospholipid may interact with one or more negatively charged phospholipids of a membrane (e.g., a cellular or intracellular membrane). Fusion of a phospholipid to a membrane may allow one or more elements of a lipid-containing composition to pass through the membrane permitting, e.g., delivery of the one or more elements to a cell.

[0224] Any of the compositions disclosed herein can be administered to a non-human subject, such as a laboratory or farm animal. Non-limiting examples of a non-human subject include laboratory or research animals, pets, wild or domestic animals, farm animals, etc., e.g., a dog, a goat, a guinea pig, a hamster, a mouse, a pig, a non-human primate (e.g., a gorilla, an ape, an orangutan, a lemur, a baboon, etc.), a rat, a sheep, a horse, a cow, or the like. As used herein, a “lipid component” is that component of a nanoparticle composition that includes one or more lipids. For example, the lipid component may include one or more cationic/ionizable, PEGylated, structural, or other lipids, such as phospholipids.

### III. Methods

#### A. CRISPR

[0225] Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. Although methods and materials similar or equivalent to those described herein may be used in the

practice or testing of the present disclosure, suitable methods and materials are described below. All publications, patent applications, patents, and other references mentioned herein are incorporated by reference in their entirety. In addition, the materials, methods, and examples are illustrative only and not intended to be limiting.

[0226] In one aspect, the present disclosure encompasses compositions relating to clustered regularly interspaced short palindromic repeats (CRISPR) and CRISPR-associated RNA-guided nucleases and associated methods, components, and compositions (hereafter, CRISPR/Cas systems). Such systems minimally require at least one isolated or non-naturally occurring RNA-guided nuclease (e.g., a Cas9 protein) and at least one isolated or non-naturally occurring guide RNA (e.g., an sgRNA) to effectuate augmentation of a nucleic acid sequence (e.g., genomic DNA).

[0227] In some embodiments, a CRISPR/Cas system effectuates the alteration of a targeted gene or locus in a eukaryotic cell by effecting an alteration of the sequence at a target position (e.g., by creating an insertion or deletion (collectively, an indel) resulting in loss-of-function of (i.e., knocking out) the affected gene or allele; e.g., a nucleotide substitution resulting in a truncation, nonsense mutation, or other type of loss-of-function of an encoded product of, for example, (i) one or more growth factors or growth factor receptors (e.g., FGF2, CCN2, NGF, NTF3, NTF4, BDNF, FGFR1, NGFR, NTRK1, NTRK2), (ii) one or more metalloproteases or regulators thereof (e.g., ADAM17, ADAMTS1, ADAMTS5, MMP1, MMP2, MMP3, MMP7, MMP8, MMP10, MMP12, MMP13, TIMP1, TIMP3), (iii) one or more cytokines, chemokines or cytokine/chemokine receptors (e.g., CXCL1, CXCL2, CXCL3, CXCL5, CXCL6, CXCL8, CCL2, CCL3, CCL5, CCL7, CCL20, IL1A, IL1B, IL4, IL6, IL10, IL13, IL17A, IL18, TNF, CXCR1, CXCR2, CCR7, TNFRSF1A, TNFRSF1B, IL1R1, IL1RAP, IL4R, IL6R, IL10RA, IL10RB, IL13RA1, IL13RA2, IL17RA, IL18R1, IL18RAP), (iv) one or more regulators of neuronal signaling (e.g., SCN1A, SCN2A, SCN3A, SCN4A, SCN5A, SCN8A, SCN9A, SCN10A, SCN11A, TAC1, TAC3, TACR1, TACR2, TACR3, ATP1A1), (v) one or more other regulators of cell signaling (e.g., CALCA, CALCB, CALCRL, RAMP1, ADM, CRCP, YAP1, MRGPRX2), and (vi) combinations of any genes of (i)-(v), i.e., mRNA or protein; a deletion of one or more nucleotides resulting in a truncation, nonsense mutation, or other type of loss-of-function of an encoded product of, for example, one or more FGF2, CCN2, ADAMTS5, MMP1, or NGF gene; e.g., loss-of-function of the encoded mRNA or protein by a single nucleotide, double nucleotide, or other frame-shifting deletion, or a deletion resulting in a premature stop codon; or an insertion resulting in a truncation, nonsense mutation, or other type of loss-of-function of an encoded gene product, such as an encoded gene product of, for example, one or FGF2, CCN2, ADAMTS5, MMP1, or NGF gene (i.e., mRNA or protein); e.g., a single nucleotide, double nucleotide, or other frame-shifting insertions, or an insertion resulting in a premature stop codon. In some embodiments, a CRISPR/Cas system of the present disclosure provides for the alteration of a gene and/or encoded product of a gene, such that the altered product has a resultant loss-of-function and becomes a dominant negative or decoy (e.g., a transmembrane receptor incapable of initiating intracellular signaling or a soluble receptor). In some embodiments, a CRISPR/Cas system of the present disclosure is encapsu-

lated in an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0228] In one aspect, CRISPR/Cas systems effectuate changes to the sequence of a nucleic acid through nuclease activity. For example, in the case of genomic DNA, the RNA-guided-nuclease locates a target position within a targeted gene or locus by sequence complementarity with the target genomic sequence (e.g., CRISPR RNA (crRNA) or a complementary component of a synthetic single guide RNA (sgRNA)) and cleaves the genomic DNA upon recognition of a particular, nuclease-specific motif called the protospacer adjacent motif (PAM). See generally, Collias, D., & Beisel, C. L. (2021). *Nature Communications*, 12(1), 1-12.

[0229] Nuclease activity (i.e., cleavage) induces a double-strand break (DSB) in the case of genomic DNA. Endogenous cellular mechanisms of DSB repair, namely non-homologous end joining (NHEJ), microhomology-mediated end joining (MMEJ), and homologous recombination, result in erroneous repair at a given target position with some calculable frequency as a result of interference from said components of the CRISPR/Cas system, thereby introducing substitutions or indels into the genomic DNA. See generally Scully, R., et al. (2019). *Nature Reviews Molecular Cell Biology*, 20(11), 698-714. At some frequency, these indels and/or substitutions may result in frameshifts, nonsense mutations (i.e., early stop codons) or truncations that impact the availability of gene products, such as mRNA and/or protein. In certain embodiments, the CRISPR/Cas system may induce a homology-directed repair (HDR) mechanism leading to insertions of non-random sequences at a target position through the use of templates (e.g., an HDR template) provided to the cell as part of the system along with the nuclease and gRNA. See Bloh, K., & Rivera-Torres, N. (2021). *International Journal of Molecular Sciences*, 22(8), 3834.

[0230] In general, the minimum requirements of the CRISPR/Cas system will be dependent upon the nuclease (i.e., Cas protein) provided therewith. To this extent, these bacterially derived nucleases have been functionally divided into Types I, III, and V, which all fall into Class 1 and Types II, IV, and VI that are grouped into Class 2.

#### Class 1 CRISPR/Cas Systems:

[0231] The exact components, compositions, and methods for effectuating a change in a targeted nucleic acid sequence using a Class 1 CRISPR/Cas system will vary, but should minimally include: a nuclease (selected from at least Types I, and III), at least one guide RNA selected from 1) sgRNA or 2) a combination of crRNA and tracrRNA. These CRISPR/Cas systems have been categorized together as Class 1 CRISPR/Cas systems due to their similarities in requirements and mode of action within a eukaryotic cell. To this end, compositions, components, and methods among Class 1 constituents may be considered functionally interchangeable, and the following details, provided merely for exemplary purposes, do not represent an exhaustive list of class members. In some embodiments, a CRISPR/Cas system of the present disclosure is encapsulated in an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0232] Cas3 (see Table 1) is the prototypical Type I DNA nuclease that functions as the effector protein as part of a larger complex (the Cascade complex comprising Cse1,

Cse2,), that is capable of genome editing. See generally He, L., et al. (2020). *Genes*, 11(2), 208. Unlike other CRISPR/Cas systems, Type I systems localize to the DNA target without the Cas3 nuclease via the Cascade complex, which then recruits Cas3 to cleave DNA upon binding and locating the 3' PAM. The Cascade complex is also responsible for processing crRNAs such that they can be used to guide it to the target position. Because of this functionality, Cascade has the ability to process multiple arrayed crRNAs from a single molecule. See. Luo, M. (2015). *Nucleic Acids Research*, 43(1), 674-681. As such, Type I system may be used to edit multiple targeted genes or loci from a single molecule.

[0233] Because the natural Cas3 substrate is ssDNA, its function in genomic editing is thought to be as a nickase; however, when targeted in tandem, the resulting edit is a result of blunt end cuts to opposing strands to approximate a blunt-cutting endonuclease, such as Cas9. See Pickar-Oliver, A., & Gersbach, C. A. (2019). *Nature Reviews Molecular Cell Biology*, 20(8), 490-507.

[0234] Like Type I nucleases, the Type III system relies upon a complex of proteins to effect nucleic acid cleavage. Particularly, Cas10 possesses the nuclease activity to cleave ssDNA in prokaryotes. See Tamulaitis, G. *Trends in Microbiology*, 25(1), 49-61 (2017). Interestingly, this CRISPR/Cas system, native to archaea, exhibits dual specificity and targets both ssDNA and ssRNA. Aside from this change, the system functions much like Type I in that the crRNA targets an effector complex (similar to Cascade) in a sequence-dependent manner. Similarly, the effector complex processes crRNAs prior to association. The dual nature of this nuclease makes its applications to genomic editing potentially more powerful, as both genomic DNA and, in some cases, mRNAs with the same sequence may be targeted to silence particular targeted genes.

#### Class 2 CRISPR/Cas Systems:

[0235] The exact components, compositions, and methods for effectuating a change in a targeted nucleic acid sequence using a Class 2 CRISPR/Cas system will vary but should minimally include: a nuclease (selected from at least Types II, and V), at least one guide RNA selected from 1) sgRNA or 2) a combination of crRNA and tracrRNA. These CRISPR/Cas systems have been categorized together as Class 2 CRISPR/Cas systems due to their similarities in requirements and mode of action within a eukaryotic cell. To this end, compositions, components, and methods among Class 2 constituents may be considered functionally interchangeable, and the following details, provided merely for exemplary purposes, do not represent an exhaustive list of class members:

[0236] Type II nucleases are the best-characterized CRISPR/Cas systems, particularly the canonical genomic editing nuclease Cas9 (see Table 1). Multiple Cas9 proteins, derived from various bacterial species, have been isolated. The primary distinction between these nucleases is the PAM, a required recognition site within the targeted dsDNA. After association with a gRNA molecule, the crRNA (or targeting domain of a sgRNA) orients the nuclease at the proper

position, but the protein's recognition of the PAM is what induces a cleavage event near that site, resulting in a blunt DSB.

[0237] In addition to the naturally derived Cas9 proteins, several engineered variants have similarly been reported. These range from Cas9 with enhanced specific (i.e., less off-target activity), such as espCas9. Others have been catalytically modified via point mutations in the RuvC (e.g., D10A) and HNH (e.g., H840A) domains such that they induce only single-strand breaks (i.e., Cas9 nickases). See Frock, R. et al. (2015). *Nature Biotechnology*, 33(2), 179-186. These have also been shown to be less error-prone in editing. Such mitigation of off-target effects becomes paramount when selecting for a desired insertion (i.e., a knock in mutation, in which a desired nucleotide sequence is introduced into a target nucleic acid molecule) rather than a deletion. Indeed, less off-target effects may aid in the preferred DNA repair mechanism (HDR, in most instances for knock in mutations). See generally Naeem, M., et al. (2020). *Cells*, 9(7), 1608.

[0238] Additional exemplary further engineered variants of canonical Cas proteins (e.g., mutants, chimeras, and include the following (each of which are hereby incorporated by reference in their entireties for all purposes): WO2015035162A2, WO2019126716A1, WO2019126774A1, WO2014093694A1, WO2014150624A1, US20190225955A1, U.S. patent Ser. No. 11/427,818, U.S. patent Ser. No. 11/242,542, U.S. patent Ser. No. 11/098,297, U.S. patent Ser. No. 10/876,100, U.S. patent Ser. No. 10/767,193, U.S. patent Ser. No. 10/494,621, and U.S. patent Ser. No. 10/100,291.

[0239] For the avoidance of doubt, SpCas9 collectively refers to any one of the group consisting of espCas9 (also referred to herein as ES-Cas9 or esCas9), HF-Cas9, PE-Cas9, ARCas9 (also referred to as AR-Cas9), SpCas9-D1135E, SpCas9-HF1, HypaCas9, HiFiCas9, xCas9-3.6, xCas9-3.7, Sniper-Cas9, evoCas9, SpartaCas, LZ3Cas9, miCas9, and SuperFi-Cas9. Additional examples of Cas9 variants disclosed in the following are hereby incorporated by reference in their entireties for all purposes: Huang, X., et al. (2022). *Cells*, 11(14), 2186.

[0240] Like the canonical Cas9 systems, Type V nucleases only require a synthetic sgRNA with a targeting domain complementary to a genomic sequence to carry out genomic editing. These nucleases contain a RuvC domain but lack the HNH domain of Type II nucleases. Further, Cas12, for example, leaves a staggered cut in the dsDNA substrate distal to the PAM, as compared to Cas9's blunt cut next to the PAM. Both Cas12a, also known as Cpf1, and Cas12b, also known as C2cl (see Table 1), act as part of larger complex of two gRNA-associated nucleases that acts on dsDNA as a quaternary structure, nicking each strand simultaneously. See Zetsche, B. et al. (2015). *Cell*, 163(3):759-771; see also Liu, L. et al. (2017). *Molecular Cell*, 65(2): 310-322. Additionally, Cas12b (C2cl) is a highly accurate nuclease with little tolerance for mismatches. See Yang, H. et al. (2016). *Cell*, 167(7):1814-1828.e12.

TABLE 1

Exemplary list of Cas nucleases and their requirements			
Nuclease (Species)	PAM (5' → 3')	Type of end generated (nucleic acid target)	Spacer length (nt)
Cas9 ( <i>S. pyogenes</i> )	NGG	Blunt (dsDNA)	20
Cas9 ( <i>S. aureus</i> )	NNGRRT	Blunt (dsDNA)	20
Cas9 ( <i>C. jejuni</i> )	NNNNRYAC	Blunt (dsDNA)	22
Cas9 ( <i>S. thermophilus</i> )	NNAGAAW	Blunt (dsDNA)	20
Cas9 ( <i>N. meningitidis</i> )	NNNNGATT	Blunt (dsDNA)	24
Cas9 ( <i>F. novicida</i> )	NGG	Blunt (dsDNA)	21
Cas12a ( <i>L. bacterium</i> )	TTTV	5' staggered (dsDNA/ssDNA)	23-25
Cas12a ( <i>Acidaminococcus</i> sp.)	TTTN	5' staggered (dsDNA/ssDNA)	24
Cas3 ( <i>E. coli</i> )	CTT/CCT/CAT/CTC	None/blunt (ssDNA)	32

See generally Wang, J., Zhang, C., & Feng, B. (2020). Journal of Cellular and Molecular Medicine, 24(6), 3256-3270, where N = any nucleotide; R = any purine (A or G); Y = any pyrimidine (C or T); W = A or T; V = A, C or G.

[0241] In one aspect, the CRISPR/Cas system of the present disclosure comprises at least one RNA-guided nuclease (e.g. a Cas protein) derived from one or more of the following selected bacterial genera: *Corynebacterium*, *Sutterella*, *Legionella*, *Treponema*, *Fiifactor*, *Eubacterium*, *Streptococcus*, *Lactobacillus*, *Mycoplasma*, *Bacteroides*, *Flavobacterium*, *Spirochaeta*, *Azospirillum*, *Gluconacetobacter*, *Neisseria*, *Roseburia*, *Parvibaculum*, *Nitratifractor*, *Campylobacter*, *Pseudomonas*, *Streptomyces*, *Staphylococcus*, *Francisella*, *Acidaminococcus*, *Lachnospiraceae*, *Lepotrichia*, and *Prevotella*. In some embodiments, the Cas protein is derived from Deltaproteobacteria or Planctomycetes bacterial species.

[0242] Some aspects of the present disclosure provide strategies, methods, compositions, and treatment modalities for altering a targeted sequence within a gene locus (e.g., altering the sequence of wild type and/or of a mutant sequence within a cell or within a mammal) by insertion or deletion of one or more nucleotides mediated by an RNA-guided nuclease and one or more guide RNAs (gRNAs), resulting in loss of function of the targeted gene product. In some embodiments, the loss of function results in “knocking out” the gene of interest (i.e., generation of a “knock out”) by ablating gene expression. In some embodiments, the loss function results in a non-functional gene product (i.e., a gene product without all functionality of the wildtype gene product). In some embodiments, the loss of function results in expression of gene product with different characteristics (e.g., different binding affinity or different cellular localization).

[0243] In certain embodiments, the targeted gene is selected from (i) one or more growth factors or growth factor receptors (e.g., FGF2, CCN2, NGF, NTF3, NTF4, BDNF, FGFR1, NGFR, NTRK1, NTRK2), (ii) one or more metalloproteases or regulators thereof (e.g., ADAM17, ADAMTS1, ADAMTS5, MMP1, MMP2, MMP3, MMP7, MMP8, MMP10, MMP12, MMP13, TIMP1, TIMP3), (iii) one or more cytokines, chemokines or cytokine/chemokine

receptors (e.g., CXCL1, CXCL2, CXCL3, CXCL5, CXCL6, CXCL8, CCL2, CCL3, CCL5, CCL7, CCL20, IL1A, IL1B, IL4, IL6, IL10, IL13, IL17A, IL18, TNF, CXCR1, CXCR2, CCR7, TNFRSF1A, TNFRSF1B, IL1R1, IL1RAP, IL4R, IL6R, IL10RA, IL10RB, IL13RA1, IL13RA2, IL17RA, IL18R1, IL18RAP), (iv) one or more regulators of neuronal signaling (e.g., SCN1A, SCN2A, SCN3A, SCN4A, SCN5A, SCN8A, SCN9A, SCN10A, SCN11A, TAC1, TAC3, TACR1, TACR2, TACR3, ATP1A1), (v) one or more other regulators of cell signaling (e.g., CALCA, CALCB, CALCRL, RAMP1, ADM, CRCP, YAP1, MRGPRX2), and (vi) combinations of any genes of (i)-(v). In some embodiments, any region of the targeted gene (e.g., a promoter region, a 5' untranslated region, a 3' untranslated region, an exon, an intron, or an exon/intron border) is targeted by an RNA-guided nuclease to alter the gene. In some embodiments, a non-coding region of the targeted gene (e.g., an enhancer region, a promoter region, an intron, 5' UTR, 3' UTR, polyadenylation signal) is targeted to alter the gene.

#### CRISPR Guide RNAs:

[0244] In one aspect, the CRISPR/Cas system of the present disclosure further provides a gRNA molecule (e.g., an isolated or non-naturally occurring RNA molecule) that interacts with the RNA-guided nuclease. In certain embodiments, the gRNA is an sgRNA comprising a crRNA sequence comprising a nucleotide sequence which is complementary to a sequence in a target nucleic acid. In some embodiments, the sgRNA further comprises an RNA scaffolding portion (i.e., tracrRNA) that interacts with the RNA-guided nuclease, such that the crRNA is positioned to scan a target nucleic acid for complementarity. In some embodiments, the system is further, optionally, comprised of an oligonucleotide—an HDR template with homology to either side of the target position. See Bloh, K., & Rivera-Torres, N. (2021). International Journal of Molecular Sciences, 22(8):3834.

**[0245]** In an embodiment, the RNA-guided nuclease and sgRNA are configured to orient an associated nuclease such that a cleavage event, (e.g., a double strand break or a single strand break) occurs sufficiently close to a complementary sequence in the targeted nucleic acid, thereby facilitating an alteration in the nucleic acid sequence. In some embodiments, the crRNA is 20 nucleotides in length. In some embodiments, the crRNA is 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, or 30 nucleotides in length.

**[0246]** In some embodiments, the crRNA orients the RNA-guided nuclease such that a cleavage event occurs within 1, 2, 3, 4, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 125, 150, 175, or 200 nucleotides away from the complementary sequence in the targeted nucleic acid. The double- or single-strand break may be positioned upstream or downstream of the complementary sequence in the targeted nucleic acid. In some embodiments, the cleavage event occurs within a targeted gene. In some embodiments, the cleavage event occurs upstream of a targeted gene.

**[0247]** In certain embodiments, a second gRNA molecule, comprising a second crRNA orients a second RNA-guided nuclease, such that a cleavage event occurs sufficiently close to a complementary sequence in the targeted nucleic acid, thereby facilitating an alteration in the nucleic acid sequence. In some embodiments, the first gRNA and the second gRNA promote a cleavage event within a single targeted gene. In some embodiments, the first gRNA and the second gRNA promote a cleavage event within different targeted genes. In some embodiments, the second crRNA is 20 nucleotides in length. In some embodiments, the second crRNA is 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, or 30 nucleotides in length.

**[0248]** In some embodiments, the second crRNA orients the RNA-guided nuclease such that a cleavage event occurs within 1, 2, 3, 4, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 125, 150, 175, or 200 nucleotides away from the complementary sequence in the targeted nucleic acid. The double- or single-strand break may be positioned upstream or downstream of the complementary sequence in the targeted nucleic acid. In some embodiments, the cleavage event occurs within a targeted gene. In some embodiments, the cleavage event occurs upstream of a targeted gene.

**[0249]** In some embodiments, the targeting domains of the first gRNA and the second gRNA are configured such that a cleavage event is positioned, independently for each of the gRNA molecules, within 1, 2, 3, 4, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 125, 150, 175, or 200 nucleotides of the others cleavage event. In some embodiments, the first gRNA and the second gRNA molecules alter the targeted nucleic acid sequences simultaneously. In some embodiments, the first gRNA and the second gRNA molecules alter the targeted nucleic acid sequences sequentially.

**[0250]** In some embodiments, a single-strand break is accompanied by a second single-strand break, positioned by the crRNA of a first gRNA and a second gRNA, respectively. For example, the crRNA may orient the associated RNA-guided nucleases such that a cleavage event, (e.g., the two single-strand breaks), are positioned within 1, 2, 3, 4, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 125, 150, 175, or 200 nucleotides of one another. In some embodiments, a first crRNA and a second crRNA are

configured to orient associated RNA-guided nucleases such that, for example, two single-strand breaks occur at the same position, or within 1, 2, 3, 4, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50 nucleotides of one another, on opposing strands of genomic DNA, thereby essentially approximating a double strand break.

**[0251]** In some embodiments a nucleic acid encodes a second sgRNA molecule. In some embodiments, a nucleic acid encodes a third sgRNA molecule. In some embodiments, a nucleic acid encodes a fourth sgRNA molecule.

**[0252]** In certain embodiments, a nucleic acid may comprise (a) a sequence encoding a first sgRNA, comprising a crRNA that is complementary with a sequence in a targeted gene, (b) a sequence encoding a second sgRNA, comprising a crRNA that is complementary with a sequence in a second targeted gene, and (c) a sequence encoding an RNA-guided nuclease (e.g., Cas9). Optionally, (d) and (e) are sequences encoding a third sgRNA and a fourth sgRNA, respectively. In some embodiments, the second targeted gene is the same as the first targeted gene. In other embodiments, the second targeted gene is different from the first targeted gene. In some embodiments, (a), (b), and (c) are encoded within the same nucleic acid molecule (e.g., the same vector). In some embodiments, (a) and (b) are encoded within the same nucleic acid molecule. In some embodiments, (a), (b) and (d) are encoded within the same nucleic acid molecule. In some embodiments, (a), (b) and (e) are encoded within the same nucleic acid molecule. In some embodiments, (a), (b), (d) and (e) are encoded within the same nucleic acid molecule. In some embodiments, (a), (b), and (c) are encoded within separate nucleic acid molecules. When more than two sgRNAs are used, any combination of (a), (b), (c), (d) and (e) may be encoded within a single or separate nucleic acid molecules.

**[0253]** In one aspect, the nucleic acid molecules (i.e., those encoding (a), (b), (c), (d) or (e)) are delivered to a target cell (i.e., any combination of the encoded RNA-guided nuclease of (c) and at least one encoded gRNA molecule of (a), (b), (d), or (e) contact a target cell). In some embodiments, said nucleic acid molecules are delivered to a target cell in vivo. In other embodiments, said nucleic acid molecules are delivered to a target cell ex vivo. In some embodiments, said nucleic acid molecules are delivered to a target cell in vitro. In certain embodiments, said nucleic acid molecules are delivered to a target cell as DNA. In other embodiments, said nucleic acid molecules are delivered to a target cell as RNA (e.g., mRNA). In some embodiments, the products of said nucleic acid molecules are delivered as an assembled ribonucleoprotein (RNP).

**[0254]** In some embodiments, contacting a target cell comprises delivering said RNA-guided nuclease of (c), as a protein with at least one said nucleic acid molecules selected from (a), (b), (d), and (e). In some embodiments, contacting a target cell comprises delivering said encoded RNA-guided nuclease of (c), as DNA with at least one said nucleic acid molecules selected from (a), (b), (d), and (e). In some embodiments, contacting a target cell comprises delivering said encoded RNA-guided nuclease of (c), as mRNA with at least one said nucleic acid molecules selected from (a), (b), (d), and (e).

**[0255]** In certain embodiments, CRISPR components are delivered to a target cell via nanoparticles. Exemplary nanoparticles that may be used with all CRISPR/Cas systems disclosed herein include, at least, lipid nanoparticles or

liposomes, hydrogel nanoparticles, metalorganic nanoparticles, gold nanoparticles, magnetic nanoparticles and virus-like particles. See generally Xu, C. F. et al. (2021). Advanced Drug Delivery Reviews, 168:3-29. In some embodiments, CRISPR components of the present disclosure are encapsulated in an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

#### B. TALEN

[0256] In one aspect, the present disclosure contemplates use of methods, components, and compositions relating to Transcription Activator-Like Effector Nucleases (TALENs) to effectuate augmentation of a ‘nucleic acid sequence (e.g., a targeted gene).

[0257] TALE stands for “Transcription Activator-Like Effector” proteins, which include TALENs (“Transcription Activator-Like Effector Nucleases”). A method of using a TALE system for gene editing may also be referred to herein as a TALE method. TALEs are naturally occurring proteins from the plant pathogenic bacteria genus *Xanthomonas*, and contain DNA-binding domains composed of a series of 33-35-amino-acid repeat domains that each recognizes a single base pair. TALE specificity is determined by two hypervariable amino acids that are known as the repeat-variable di-residues (RVDs). Modular TALE repeats are linked together to recognize contiguous DNA sequences. A specific RVD in the DNA-binding domain recognizes a base in the target locus, providing a structural feature to assemble predictable DNA-binding domains. The DNA binding domains of a TALE are fused to the catalytic domain of a type IIS FokI endonuclease to make a targetable TALE nuclease. To induce site-specific mutation, two individual TALEN arms, separated by a 14-20 base pair spacer region, bring FokI monomers in close proximity to dimerize and produce a targeted double-strand break.

[0258] Several large, systematic studies utilizing various assembly methods have indicated that TALE repeats can be combined to recognize virtually any user-defined sequence. Custom-designed TALE arrays are also commercially available through Cellectis Bioresearch (Paris, France), Transposagen Biopharmaceuticals (Lexington, KY, USA), and Life Technologies (Grand Island, NY, USA). TALE and TALEN methods suitable for use in the present disclosure are described in U.S. Patent Application Publication Nos. US 2011/0201118 A1; US 2013/0117869 A1; US 2013/0315884 A1; US 2015/0203871 A1 and US 2016/0120906 A1, the disclosures of which are incorporated by reference herein.

[0259] Non-limiting examples of genes that may be silenced or inhibited by permanently gene-editing via a TALE method include (i) one or more growth factors or growth factor receptors (e.g., FGF2, CCN2, NGF, NTF3, NTF4, BDNF, FGFR1, NGFR, NTRK1, NTRK2), (ii) one or more metallproteases or regulators thereof (e.g., ADAM17, ADAMTS1, ADAMTS5, MMP1, MMP2, MMP3, MMP7, MMP8, MMP10, MMP12, MMP13, TIMP1, TIMP3), (iii) one or more cytokines, chemokines or cytokine/chemokine receptors (e.g., CXCL1, CXCL2, CXCL3, CXCL5, CXCL6, CXCL8, CCL2, CCL3, CCL5, CCL7, CCL20, IL1A, IL1B, IL4, IL6, IL10, IL13, IL17A, IL18, TNF, CXCR1, CXCR2, CCR7, TNFRSF1A, TNFRSF1B, IL1R1, IL1RAP, IL4R, IL6R, IL10RA, IL10RB, IL13RA1, IL13RA2, IL17RA, IL18R1,

IL18RAP), (iv) one or more regulators of neuronal signaling (e.g., SCN1A, SCN2A, SCN3A, SCN4A, SCN5A, SCN8A, SCN9A, SCN10A, SCN11A, TAC1, TAC3, TACR1, TACR2, TACR3, ATP1A1), (v) one or more other regulators of cell signaling (e.g., CALCA, CALCB, CALCRL, RAMP1, ADM, CRCP, YAP1, MRGPRX2), and (vi) combinations of any genes of (i)-(v). In an aspect, the disclosure provides compositions for up-regulation of protein receptors (including wildtype or genetically edited), including those that bind to anti-inflammatory cytokines via a TALE method.

[0260] Examples of systems, methods, and compositions for altering the expression of a target gene sequence by a TALE method, and which may be used in accordance with embodiments of the present disclosure, are described in U.S. Pat. No. 8,586,526, which is incorporated by reference herein.

#### C. Zinc-Finger Nucleases (ZFN)

[0261] In one aspect, the present disclosure contemplates use of methods, components, and compositions relating to zinc-finger nucleases (ZFNs) to effectuate augmentation of a ‘nucleic acid sequence (e.g., a targeted gene).

[0262] An individual zinc finger contains approximately 30 amino acids in a conserved  $\beta\beta\alpha$  configuration. Several amino acids on the surface of the  $\alpha$ -helix typically contact 3 bp in the major groove of DNA, with varying levels of selectivity. Zinc fingers have two protein domains. The first domain is the DNA binding domain, which includes eukaryotic transcription factors and contain the zinc finger. The second domain is the nuclease domain, which includes the FokI restriction enzyme and is responsible for the catalytic cleavage of DNA.

[0263] The DNA-binding domains of individual ZFNs typically contain between three and six individual zinc finger repeats and can each recognize between 9 and 18 base pairs. If the zinc finger domains are specific for their intended target site then even a pair of 3-finger ZFNs that recognize a total of 18 base pairs can, in theory, target a single locus in a mammalian genome. One method to generate new zinc-finger arrays is to combine smaller zinc-finger “modules” of known specificity. The most common modular assembly process involves combining three separate zinc fingers that can each recognize a 3 base pair DNA sequence to generate a 3-finger array that can recognize a 9 base pair target site. Alternatively, selection-based approaches, such as oligomerized pool engineering (OPEN) can be used to select for new zinc-finger arrays from randomized libraries that take into consideration context-dependent interactions between neighboring fingers. Engineered zinc fingers are available commercially; Sangamo Biosciences (Richmond, CA, USA) has developed a proprietary platform (CompoZr®) for zinc-finger construction in partnership with Sigma-Aldrich (St. Louis, MO, USA).

[0264] Non-limiting examples of genes that may be silenced or inhibited by permanently gene-editing via a zinc finger method include (i) one or more growth factors or growth factor receptors (e.g., FGF2, CCN2, NGF, NTF3, NTF4, BDNF, FGFR1, NGFR, NTRK11, NTRK2), (ii) one or more metallproteases or regulators thereof (e.g., ADAM17, ADAMTS1, ADAMTS5, MMP1, MMP2, MMP3, MMP7, MMP8, MMP10, MMP12, MMP13, TIMP1, TIMP3), (iii) one or more cytokines, chemokines or cytokine/chemokine receptors (e.g., CXCL1, CXCL2,

CXCL3, CXCL5, CXCL6, CXCL8, CCL2, CCL3, CCL5, CCL7, CCL20, IL1A, IL1B, IL4, IL6, IL10, IL13, IL17A, IL18, TNF, CXCR1, CXCR2, CCR7, TNFRSF1A, TNFRSF1B, IL1R1, IL1RAP, IL4R, IL6R, IL10RA, IL10RB, IL13RA1, IL13RA2, IL17RA, IL18R1, IL18RAP), (iv) one or more regulators of neuronal signaling (e.g., SCN1A, SCN2A, SCN3A, SCN4A, SCN5A, SCN8A, SCN9A, SCN10A, SCN11A, TAC1, TAC3, TACR1, TACR2, TACR3, ATP1A1), (v) one or more other regulators of cell signaling (e.g., CALCA, CALCB, CALCRL, RAMP1, ADM, CRCP, YAP1, MRGPRX2), and (vi) combinations of any genes of (i)-(v). Non-limiting examples of genes that may be augmented such that their resultant products function as decoys or dominant negatives by permanently gene-editing via a zinc finger method include. In an aspect, the disclosure provides compositions for up-regulation of protein receptors (including wildtype or genetically edited), including those that bind to anti-inflammatory cytokines via a zinc finger method.

[0265] Examples of systems, methods, and compositions for altering the expression of a target gene sequence by a zinc finger method, which may be used in accordance with embodiments of the present disclosure, are described in U.S. Pat. Nos. 6,534,261, 6,607,882, 6,746,838, 6,794,136, 6,824,978, 6,866,997, 6,933,113, 6,979,539, 7,013,219, 7,030,215, 7,220,719, 7,241,573, 7,241,574, 7,585,849, 7,595,376, 6,903,185, and 6,479,626, which are incorporated by reference herein.

[0266] Other examples of systems, methods, and compositions for altering the expression of a target gene sequence by a zinc finger method, which may be used in accordance with embodiments of the present disclosure, are described in Beane, et al., *Mol. Therapy*, 2015, 23 1380-1390, the disclosure of which is incorporated by reference herein.

#### IV. Joint Disease or Illness

##### A. Introduction

[0267] As described herein, embodiments of the present disclosure provide compositions and methods for improving joint function and treating joint disease. In particular embodiments, compositions and methods are provided to gene-edit synovial fibroblasts, synoviocytes, chondrocytes, tissue (resident) macrophages, or other cells to reduce pro-inflammatory signaling mediated by the binding of inflammatory cytokines—including, but not limited to, IL1 $\alpha$ , IL1 $\beta$ , TNF $\alpha$ , IL6, IL8, IL18, IL33, matrix metalloproteinases (MMPs), TGF $\beta$ 1, TGF $\beta$ 2, and combinations thereof—to their cognate receptor(s). Some embodiments are used for treating various forms of arthritis and other inflammatory joint diseases. Some embodiments are further useful for treating canine lameness due to osteoarthritis. Some embodiments are further useful for treating equine lameness due to joint disease. Some embodiments are further useful for treating feline lameness due to joint disease. Some embodiments are also useful for treating post-traumatic arthritis, gout, pseudogout, psoriatic arthritis, and other inflammation-mediated or immune-mediated joint diseases. Some embodiments are further useful as it relates to encapsulation in an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0268] Treatment of osteoarthritis, degenerative joint disease, and other joint dysfunctions is complex, and few

long-term options exist for either symptomatic relief or restoring joint function. Osteoarthritis (OA) is the leading cause of disability due to pain. See, Neogi, T. (2013). *Osteoarthritis Cartilage*, 21(9):1145-53. OA and similar diseases impact all mammal species, including working animals, domestic pets, and their owners. The common mechanistic thread among joint diseases is the presence of acute or chronic inflammation, which is driven by increased levels of pro-inflammatory cytokine signaling. Joint diseases tend to take a progressive course that encompasses discomfort, pain, and—especially in the case of OA—disability, depending on the degree of disease progression.

[0269] Psoriatic arthritis (PsA) is another chronic inflammatory joint disease, in which the joint symptoms are accompanied by skin lesions, such as those commonly associated with psoriasis. See, Boehncke, W. et al. (2014). *British Journal of Dermatology*, 170(4):772-786. Like other forms of arthritis, such as OA, PsA is caused by pro-inflammatory signaling of a host of cytokines, including IL1. Indeed, PsA morbidity has been shown to correlate with single nucleotide polymorphisms (SNPs) that impact the activity of the IL1 gene locus. See, Rahman, P. et al. (2006). *Arthritis and Rheumatism*, 54(7):2321-2325. These studies also implicate inflammatory cytokine signaling, in general, and IL1 more specifically, in disease progression.

[0270] Gout is a chronic inflammatory condition that affects joints. The underlying cause is monosodium urate (MSU) crystal deposition and the resultant host response, particularly in joint structures (as well as subcutaneous tissues and other sites). See, Dalbeth, N., & Stamp, L. (2014). *Annals of the Rheumatic Diseases*, 73(9):1598-1600. The clinical manifestations include recurrent acute flares of severe inflammatory arthritis and tendinobursitis. IL1 and other pro-inflammatory mediators are a major contributor to this host response. See, Dinarello, C. A. (2014). *Molecular Medicine*, 20(1):S43-S58. To this end, effective blockade of these signaling pathways may provide relief to gout patients.

[0271] The current standard of care for many joint disease patients includes anti-inflammatory medications (e.g., NSAIDs) or anti-rheumatics (e.g., methotrexate [inhibitor of AICAR] or adalimumab [anti-TNF alpha monoclonal antibody]). See, Friedman, B., & Cronstein, B. (2019). *Joint Bone Spine*, 86(3):301-307. All of these treatments require repeated dosing for continued effectiveness, which may lead to toxicity issues or tolerance over time. As such, new methods and compositions to treat joint disease and illness are acutely needed to treat these chronic conditions.

[0272] In one aspect, the compositions and methods herein described are directed to treat joint disease or illness in a mammal in need thereof. In some embodiments, the joint disease or illness is osteoarthritis. In some embodiments, the joint disease or illness is psoriatic arthritis. In some embodiments, the joint disease or illness is gout.

[0273] Among the advantages of the present disclosure over treatments currently available for mammals afflicted with one or more joint disease or illness include the period of relief from symptoms. Upon genetic editing of a cell within a joint, pro-inflammatory signaling is silenced through the targeted gene for the life of that cell and any mitotic progeny. By contrast, biologic treatments require periodic dosing, which may magnify the impact of the host of potentially severe side effects. Among various genetic approaches, the present disclosure is also superior due to,

among other reasons, a resistance to leakiness by virtue of modifying a protein receptor, rather than ablating expression of a ligand, which may result in compensatory effects (e.g., buildup of other factors due to lack of negative feedback). [0274] In some embodiments, the present disclosure includes a method for the treatment or prevention of a joint disease or condition in a subject in need thereof, the method comprising administering, to a joint of the subject, a pharmaceutical composition comprising a therapeutically effective amount of a Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) gene-editing system, the system comprising (i) a CRISPR Associated (Cas) protein; and (ii) at least one guide RNA targeting an IL1R1 gene, IL1RAP gene, TGFBR1 gene, TGFBR2 gene, IL6R gene, IL6ST gene, TNFRSF1A gene, TNFRSF1B gene, TNFRSF3 gene, TNFRSF4 gene, or TNFRSF11A gene or a combination thereof. In some embodiments, the joint disease or condition is osteoarthritis. In some embodiments, the joint disease or condition is psoriatic arthritis. In some embodiments, the joint disease or condition is gout.

[0275] In some embodiments, the present disclosure includes a method for the treatment or prevention of an arthritis. Non-limiting examples of arthritis that can be treated using the compositions and methods described herein include post-traumatic arthritis, osteoarthritis (a degenerative condition that affects the joints, most commonly the hips, knees, and hands), rheumatoid arthritis (an autoimmune disorder that causes inflammation in the joints and surrounding tissue), psoriatic arthritis (a type of arthritis that occurs in people with psoriasis, a skin condition characterized by scaly red patches), gout (a type of arthritis caused by the buildup of uric acid crystals in the joints), lupus (a chronic autoimmune disorder that can cause inflammation and damage to the joints, as well as other organs), ankylosing spondylitis (a type of arthritis that primarily affects the spine, causing inflammation and stiffness), reactive arthritis (a type of arthritis that occurs as a reaction to an infection in the body), septic arthritis (a type of arthritis caused by an infection in the joint), juvenile idiopathic arthritis (a form of arthritis that affects children under the age of 16), and fibromyalgia (a chronic pain disorder that can cause widespread pain and stiffness, including in the joints).

[0276] In some embodiments, the present disclosure includes a method for the treatment or prevention of pseudogout, Crystal arthropathies (caused by the formation of crystals in the joints, such as gout and pseudogout), or CPPD disease (calcium pyrophosphate deposition disease) also called chondroclacnosis.

[0277] In some embodiments, the present disclosure includes a method for the treatment or prevention of rheumatoid arthritis, psoriasis, asthma, inflammatory bowel disease, multiple sclerosis, Alzheimer's disease, Type 2 diabetes, cardiovascular disease, or cancer. In some embodiments, these disorders are treated by administering a CRISPR composition, as described herein, targeting an IL1 receptor, e.g., IL1R1 or IL1RAP.

#### B. Osteoarthritis

[0278] In one aspect, the present disclosure encompasses treatments for osteoarthritis (OA). In some embodiments, OA treatment comprises a therapeutically effective amount of a Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) gene-editing system, the system comprising: (i) a CRISPR Associated (Cas) protein; and (ii) at

least one guide RNA targeting IL1R1. In some embodiments, the OA treatment comprises a CRISPR gene-editing system targeting hIL1R1. In some embodiments, the OA treatment comprises a CRISPR gene-editing system targeting cIL1R1. In some embodiments, the OA treatment comprises a CRISPR gene-editing system R targeting eIL1R1. In some embodiments, the OA treatment comprises a CRISPR gene-editing system targeting fIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprising one or more sgRNAs targeting an exon of IL1R1 is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0279] In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 1 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 2 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 3 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 4 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 5 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 6 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 7 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 8 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 9 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 10 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 11 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 12 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 13 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 14 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 15 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 16 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 17 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 18 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 19 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 20 of hIL1R1. In some



























more sgRNAs targeting exon 10 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 11 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprising one or more sgRNAs targeting an exon of cTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0331] In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 1 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 2 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 3 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 4 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 5 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 6 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 7 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 8 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 9 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 10 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprising one or more sgRNAs targeting an exon of eTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0332] In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 1 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 2 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 3 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 4 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 5 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 6 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 7 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting

exon 8 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 9 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 10 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprises one or more sgRNAs targeting exon 11 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of OA comprising one or more sgRNAs targeting an exon of fTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

### C. Psoriatic Arthritis

[0333] In one aspect, the present disclosure encompasses treatments for psoriatic arthritis (PsA). In some embodiments, the psoriatic arthritis treatment comprises a therapeutically effective amount of a Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) gene-editing system, the system comprising: (i) a CRISPR Associated (Cas) protein; and (ii) at least one guide RNA targeting IL1R1. In some embodiments, the psoriatic arthritis treatment comprises a CRISPR gene-editing system targeting hIL1R1. In some embodiments, the psoriatic arthritis treatment comprises a CRISPR gene-editing system targeting cIL1R1. In some embodiments, the psoriatic arthritis treatment comprises a CRISPR gene-editing system R targeting eIL1R1. In some embodiments, the psoriatic arthritis treatment comprises a CRISPR gene-editing system targeting fIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of PsA comprising one or more sgRNAs targeting an exon of IL1R1 is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0334] In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 1 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 3 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 4 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 5 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 6 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 7 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 8 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 9 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one





























comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0385] In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 1 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 2 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 3 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 4 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 5 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 6 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 7 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 8 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 9 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 10 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 11 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of PsA comprising one or more sgRNAs targeting an exon of cTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0386] In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 1 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 2 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 3 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 4 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 5 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 6 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 7 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 8 of eTNFRSF11A. In some embodiments, the CRISPR gene-

editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 9 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 10 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of PsA comprising one or more sgRNAs targeting an exon of eTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240. [0387] In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 1 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 2 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 3 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 4 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 5 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 6 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 7 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 8 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 9 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 10 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of psoriatic arthritis comprises one or more sgRNAs targeting exon 11 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of PsA comprising one or more sgRNAs targeting an exon of fTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

#### D. Gout

[0388] In one aspect, the present disclosure encompasses treatments for gout and other crystallopathies affecting the joint, e.g., octacalcium phosphate and calcium pyrophosphate dihydrate in horses. In some embodiments, the gout treatment comprises a therapeutically effective amount of a Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) gene-editing system, the system comprising: (i) a CRISPR Associated (Cas) protein; and (ii) at least one guide RNA targeting IL1R1. In some embodiments, the gout treatment comprises a CRISPR gene-editing system targeting hIL1R1. In some embodiments, the gout treatment comprises a CRISPR gene-editing system targeting cIL1R1. In some embodiments, the gout treatment comprises a

CRISPR gene-editing system R targeting eIL1R1. In some embodiments, the gout treatment comprises a CRISPR gene-editing system targeting fIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprising one or more sgRNAs targeting an exon of IL1R1 is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

hIL1R1 is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

**[0390]** In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 1 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 2 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 3 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 4 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 5 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 6 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 7 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 8 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 9 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 10 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 11 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 12 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 13 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 14 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 15 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 16 of cIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprising one or more sgRNAs targeting an exon of cIL1R1 is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0391] In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 1 of eIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 2 of eIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 3 of eIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 4 of eIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 5 of eIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of gout



























CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 8 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 9 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 10 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprises one or more sgRNAs targeting exon 11 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of gout comprising one or more sgRNAs targeting an exon of fTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

#### V. Back or Spine Conditions or Disorders

##### A. Introduction

[0443] Back or spine conditions or disorders, including low back pain, cervical pain, sacral pain, thoracic pain, and pain or inflammation associated with discogenic disorders e.g., degenerative disc disease (DDD) or internal disc disruption (IDD), are a major cause of morbidity and disability worldwide for which few long-term options for amelioration currently exist. Andersson GB. Epidemiological features of chronic low-back pain. *Lancet.* 1999; 354:581-585. Presently available treatments include surgical or less invasive options that often fail to offer long-term palliation. Ju, et al. *Global Spine Journal* (2020): 2192568220963058. All vertebrate species are affected by back or spine conditions or disorders, including working animals, domestic pets, and their owners. All suffer from the associated discomfort, pain, and disability, depending on the degree of disease progression.

[0444] Back or spine conditions or disorders, such as low back pain, are complex diseases characterized by a multitude of inputs contributing to a progressive course of disability. Among these contributors are morphological irregularities (e.g., disc disruptions), inflammation, changes in the localized cellular environment (e.g., vascularization and/or innervation) and degenerative changes. Peng, Bao-Gan. *World Journal of Orthopedics* 4.2 (2013): 42. Each contributing factor is driven by differential expression of various gene products, including at least pro-inflammatory cytokines, growth factors, pain signaling molecules, and other effector biomolecules. There is a pressing need for new methods and compositions to treat this spectrum of disease and its associated disability.

[0445] The present disclosure provides compositions and methods for back or spine conditions or disorders. Particularly, said conditions are treated by reducing pro-inflammatory signaling mediated by inflammatory cytokines, such as, IL1 $\alpha$ , IL1 $\beta$ , TNF- $\alpha$ , IL6, IL8, IL18, IL33, matrix metalloproteinases (MMPs), or TGFB1, or TGFB2, binding to their cognate receptor(s). In some embodiments, such conditions or disorders include disorders of the intervertebral discs (IVDs). In some embodiments, the condition or disorder is DDD. In some embodiments, the condition or disorder is IDD. In some embodiments, the condition or disorder is low back pain. Some embodiments are further useful as it relates

to encapsulation in an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0446] Among the advantages of the present disclosure over treatments currently available for mammals afflicted with back or spine conditions or disorders include the period of relief from symptoms. Upon local administration (e.g., intradiscal injection) and subsequent genetic editing of a cell (e.g., a chondrocyte, a tenocyte, an osteocyte, a monocyte, a macrophage or the cells of the nucleus pulposus or annulus fibrosus), pro-inflammatory signaling is silenced through the targeted gene for the life of that cell. By contrast, biologic treatments require periodic dosing, which may magnify the impact of any side effects, which can be severe. Among various genetic approaches, the present disclosure is also superior due to the resistance to leakiness built in by virtue of modifying a protein receptor, rather than ablating its expression altogether.

##### B. Low Back Pain

[0447] In one aspect, the present disclosure encompasses treatments for low back pain. In some embodiments, the low back pain treatment comprises a therapeutically effective amount of a Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) gene-editing system, the system comprising: (i) a CRISPR Associated (Cas) protein; and (ii) at least one guide RNA targeting IL1R1. In some embodiments, the low back pain treatment comprises a CRISPR gene-editing system targeting hIL1R1. In some embodiments, the low back pain treatment comprises a CRISPR gene-editing system targeting cIL1R1. In some embodiments, the low back pain treatment comprises a CRISPR gene-editing system targeting eIL1R1. In some embodiments, the low back pain treatment comprises a CRISPR gene-editing system targeting fIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprising one or more sgRNAs targeting an exon of IL1R1 is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

[0448] In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 1 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 2 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 3 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 4 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 5 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 6 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 7 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 8 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more





























**[0500]** In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 1 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 2 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 3 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 4 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 5 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 6 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 7 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 8 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 9 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 10 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprising one or more sgRNAs targeting an exon of eTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

**[0501]** In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 1 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 2 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 3 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 4 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 5 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 6 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 7 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 8 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 9 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 10 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain comprises one or more sgRNAs targeting exon 11 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of low back pain

back pain comprising one or more sgRNAs targeting an exon of fTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

### C. DDD

**[0502]** In one aspect, the present disclosure encompasses treatments for degenerative disc disorder (DDD). In some embodiments, the DDD treatment comprises a therapeutically effective amount of a Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) gene-editing system, the system comprising: (i) a CRISPR Associated (Cas) protein; and (ii) at least one guide RNA targeting IL1R1. In some embodiments, the DDD treatment comprises a CRISPR gene-editing system targeting hIL1R1. In some embodiments, the DDD treatment comprises a CRISPR gene-editing system targeting cIL1R1. In some embodiments, the DDD treatment comprises a CRISP gene-editing system R targeting eIL1R1. In some embodiments, the DDD treatment comprises a CRISPR gene-editing system targeting fIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprising one or more sgRNAs targeting an exon of IL1R1 is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

**[0503]** In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 1 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 2 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 3 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 4 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 5 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 6 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 7 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 8 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 9 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 10 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 11 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 12 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 13 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 14 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more



























cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 4 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 5 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 6 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 7 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 8 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 9 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 10 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 11 of cTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprising one or more sgRNAs targeting an exon of cTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

**[0555]** In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 1 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 2 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 3 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 4 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 5 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 6 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 7 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 8 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 9 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 10 of eTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprising one or more sgRNAs targeting an exon of eTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

**[0556]** In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 1 of fTNFRSF11A. In some

embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 2 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 3 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 4 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 5 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 6 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 7 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 8 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 9 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprises one or more sgRNAs targeting exon 10 of fTNFRSF11A. In some embodiments, the CRISPR gene-editing system for the treatment of DDD comprising one or more sgRNAs targeting an exon of fTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

#### D. IDD

**[0557]** In one aspect, the present disclosure encompasses treatments for internal disc disruption (IDD). In some embodiments, the IDD treatment comprises a therapeutically effective amount of a Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) gene-editing system, the system comprising: (i) a CRISPR Associated (Cas) protein; and (ii) at least one guide RNA targeting IL1R1. In some embodiments, the IDD treatment comprises a CRISPR gene-editing system targeting hIL1R1. In some embodiments, the IDD treatment comprises a CRISPR gene-editing system targeting cIL1RT. In some embodiments, the IDD treatment comprises a CRISP gene-editing system R targeting eIL1R1. In some embodiments, the IDD treatment comprises a CRISPR gene-editing system targeting fIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of IDD comprising one or more sgRNAs targeting an exon of IL1R1 is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

**[0558]** In some embodiments, the CRISPR gene-editing system for the treatment of IDD comprises one or more sgRNAs targeting exon 1 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of IDD comprises one or more sgRNAs targeting exon 2 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of IDD comprises one or more sgRNAs targeting exon 3 of hIL1R1. In some embodiments, the CRISPR gene-editing system for the treatment of IDD





























comprising one or more sgRNAs targeting an exon of fTNFRSF11A is delivered via one or more lipid nanoparticles (LNPs), the LNPs comprising an LNP system described herein, e.g., and without limitation, any one of LNP systems LNP001 to LNP240.

## VI. Delivery

### A. Lipid Nanoparticles (LNP)

**[0612]** In some embodiments, a CRISPR gene-editing system is delivered by a nanoparticle. Without wishing to be bound by any particular theory, in certain embodiments, nucleic acids, when present in the nanoparticle, are resistant in aqueous solution to degradation with a nuclease. In other embodiments, proteins are protected from protease degradation. In some embodiments, proteins and nucleic acids encapsulated by nanoparticles are capable of penetrating the cellular plasma membrane.

**[0613]** Lipid nanoparticles comprising nucleic acids and their method of preparation is disclosed in at least WO2017/019935, WO2017/049074, WO2017/201346, WO2017/218704, WO2018/006052, WO2018/013525, WO2018/089540, WO2018/119115, WO2018/126084, WO2018/157009, WO2018/170336, WO2018/222890, WO2019/046809, WO2019/089828, WO2020/061284, WO2020/061317, WO2020/081938, WO2020/097511, WO2020/097520, WO2020/097540, WO2020/097548, WO2020/214946, WO2020/219941, WO2020/232276, WO2020/227615, WO2020/061295, WO2021/007278, WO2021/016430, WO2021/021988, EP Patent No. EP 2 972 360, US20200155691, US20200237671, U.S. Pat. Nos. 8,058,069, 8,492,359, 8,822,668, 9,364,435, 9,404,127, 9,504,651, 9,593,077, 9,738,593, 9,868,691, 9,868,692, 9,950,068, 10,138,213, 10,166,298, 10,221,127, 10,238,754, 10,266,485, 10,383,952, 10,730,924, 10,766,852, 11,079,379, 11,141,378 and 11,246,933, which are incorporated herein by reference in their entirety for all purposes.

### Lipid Nanoparticle Compositions

**[0614]** In some embodiments, the largest dimension of a nanoparticle composition is 1 micrometer or shorter (e.g., 1 micrometer, 900 nm, 800 nm, 700 nm, 600 nm, 500 nm, 400 nm, 300 nm, 200 nm, 175 nm, 150 nm, 125 nm, 100 nm, 75 nm, 50 nm, or shorter), e.g., when measured by dynamic light scattering (DLS), transmission electron microscopy, scanning electron microscopy, or another method. Nanoparticle compositions include, for example, lipid nanoparticles (LNPs), liposomes, lipid vesicles, and lipoplexes. In some embodiments, nanoparticle compositions are vesicles including one or more lipid bilayers. In certain embodiments, a nanoparticle composition includes two or more concentric bilayers separated by aqueous compartments. Lipid bilayers may be functionalized and/or crosslinked to one another. Lipid bilayers may include one or more ligands, proteins, or channels. In various embodiments, lipid nanoparticles described herein have a mean diameter of from about 30 nm to about 150 nm, from about 40 nm to about 150 nm, from about 50 nm to about 150 nm, from about 60 nm to about 130 nm, from about 70 nm to about 110 nm,

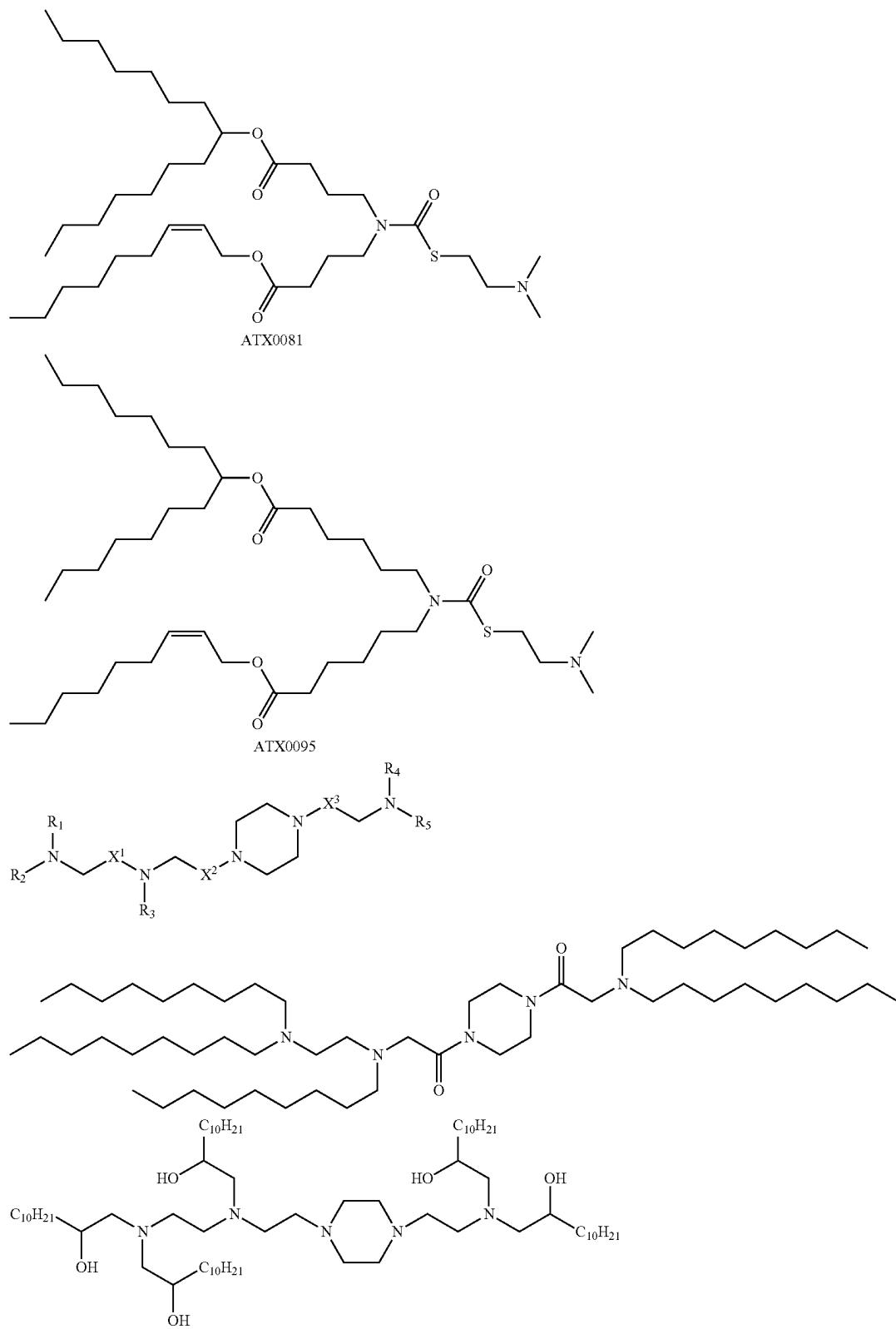
from about 70 nm to about 100 nm, from about 80 nm to about 100 nm, from about 90 nm to about 100 nm, from about 70 nm to about 90 nm, from about 80 nm to about 80 nm, or about 30 nm, 35 nm, 40 nm, 45 nm, 50 nm, 55 nm, 60 nm, 65 nm, 70 nm, 75 nm, 80 nm, 85 nm, 90 nm, 95 nm, 100 nm, 105 nm, 110 nm, 115 nm, 120 nm, 125 nm, 130 nm, 135 nm, 140 nm, 145 nm, or 150 nm, and are substantially non-toxic.

**[0615]** In certain embodiments, the lipid nanoparticles described herein comprise one or more components, including a lipid component, and (optionally) a structural component. The lipid component comprises lipids selected from ionizable and/or cationic lipids (i.e., lipids that may have a positive or partial positive charge at physiological pH), neutral lipids (e.g., phospholipids, or sphingolipids), and polymer-conjugated lipids (e.g., PEGylated lipids). In some embodiments, the lipid component comprises a single ionizable lipid. In other embodiments, the lipid component comprises 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, or 20 ionizable lipids. In some embodiments, the lipid component comprises a single neutral lipid. In other embodiments, the lipid component comprises 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, or 20 neutral lipids. In some embodiments, the lipid component comprises a single polymer-conjugated lipid. In other embodiments, the lipid component comprises 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, or 20 polymer-conjugated lipids. In some embodiments, the structural component comprises a single structural lipid. In other embodiments, the structural component comprises 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, or 20 structural lipids. In some embodiments, the lipid component comprises at least one cationic lipid, at least one neutral lipid, and at least one polymer-conjugated lipid. The present disclosure contemplates that the lipid component may comprise any combination of the foregoing constituents.

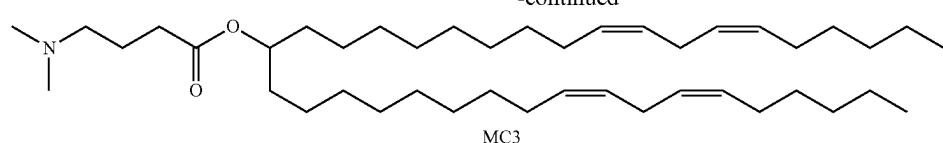
### Ionizable/Cationic Lipids

**[0616]** In some embodiments, the lipid component comprises an ionizable lipid. In some embodiments, the ionizable lipid is anionic. In other embodiments, the ionizable lipid is a cationic lipid. In some embodiments, the lipid component comprises cationic lipids including, but not limited to, a cationic lipid selected from the group consisting of 3-(didodecylamino)-N1,N1,4-tridodecyl-1-piperazineethanamine (KL10), N1-[2-(didodecylamino)ethyl]-N1,N4,N4-tridodecyl-1,4-piperazinediethanamine (KL22), 14,25-ditridodecyl-15,18,21,24-tetraaza-octatriacontane (KL25), 1,2-dilinoleyoxy-N,N-dimethylaminopropane (DLin-DMA), 2,2-dilinoleyl-4-dimethylaminomethyl-[1,3]-dioxolane (DLin-K-DMA), heptatriaconta-6,9,28,31-tetraen-19-yl 4-(dimethylamino)butanoate (DLin-MC3-DMA), 2,2-dilinoleyl-4-(2-dimethylaminoethyl)-[1,3]-dioxolane (DLin-KC2-DMA), 1,2-dioleyloxy-N,N-dimethylaminopropane (DODMA), 2-({8-[{3.beta.}-cholest-5-en-3-yloxy]octyl}oxy)-N,N-dimethyl-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA), (2R)-2-({8-[{3.beta.}-cholest-5-en-3-yloxy]octyl}oxy)-N,N-dimethyl-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA (2R)), (2S)-2-({8-[{3.beta.}-cholest-5-en-3-yloxy]octyl}oxy)-N,N-dimethyl-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA (2S)), a lipid including a cyclic amine group, and mixtures thereof.

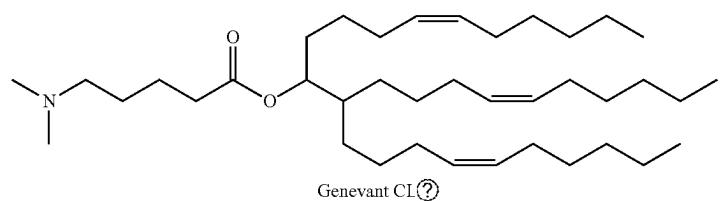
[0617] Non-exhaustive and non-limiting examples of cat-  
ionic lipids include:



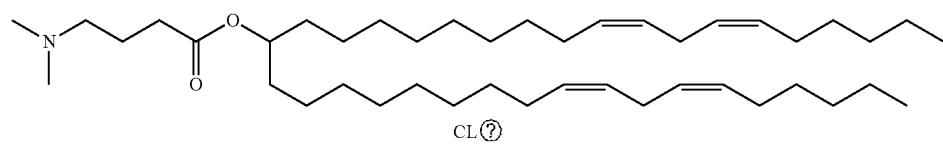
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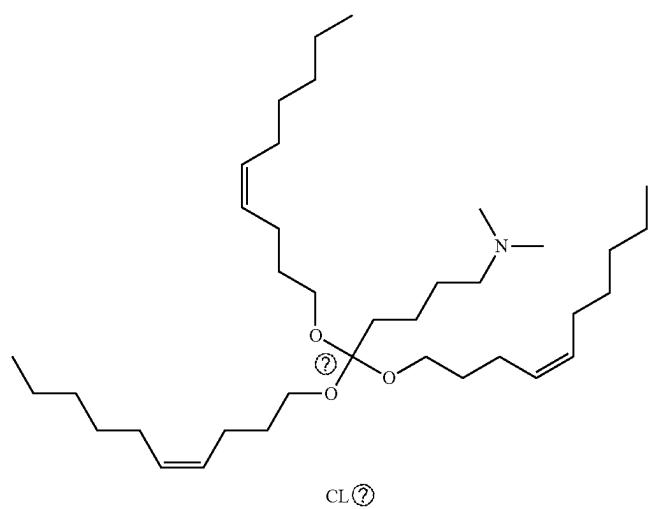
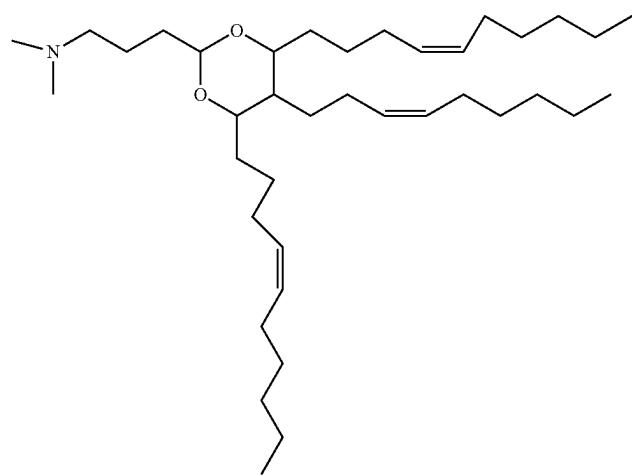
MC3



Genevant CL⑦

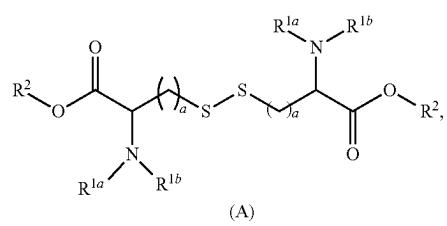
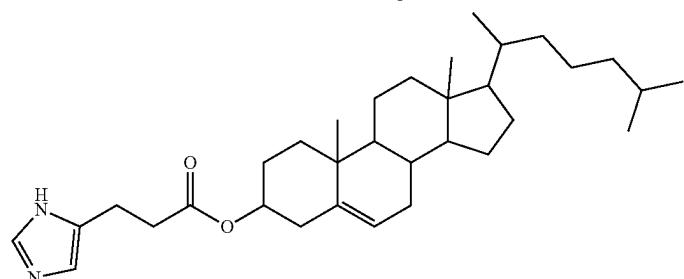
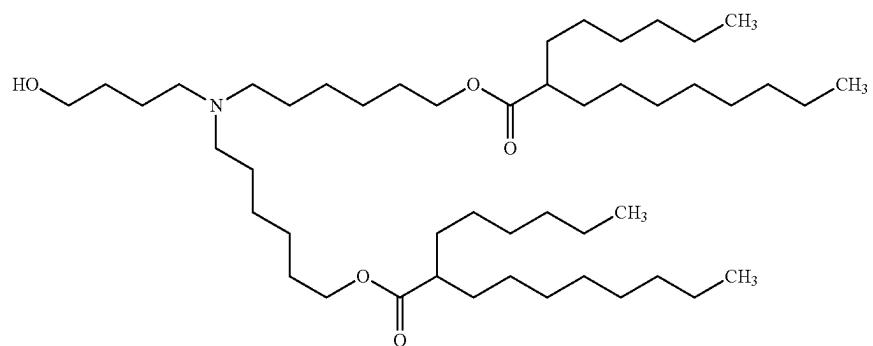
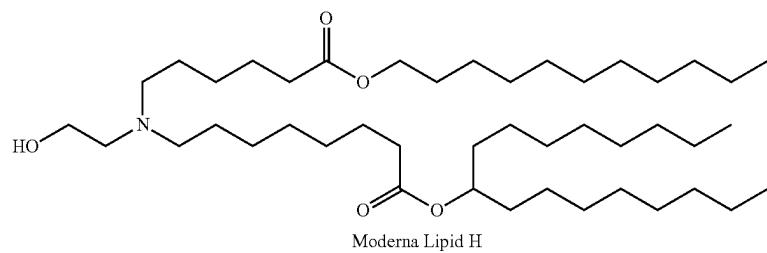
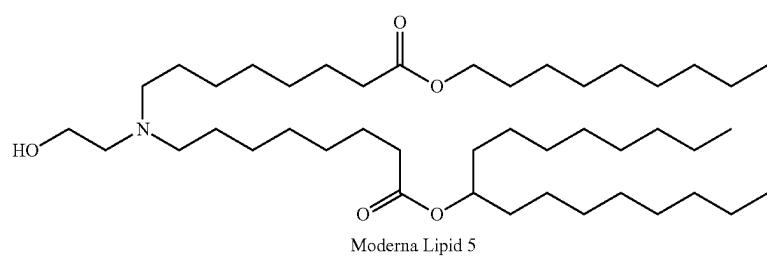
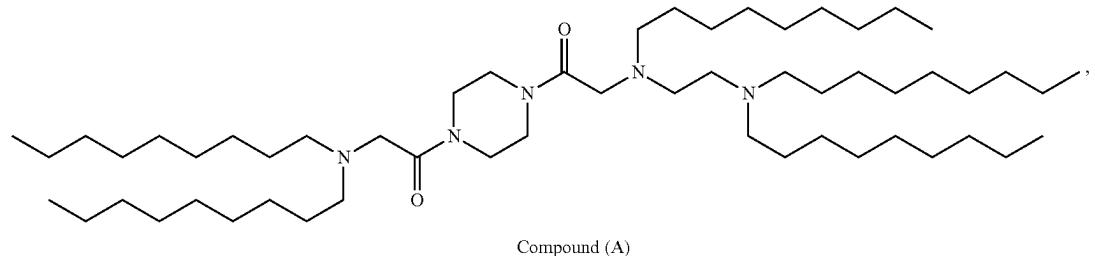


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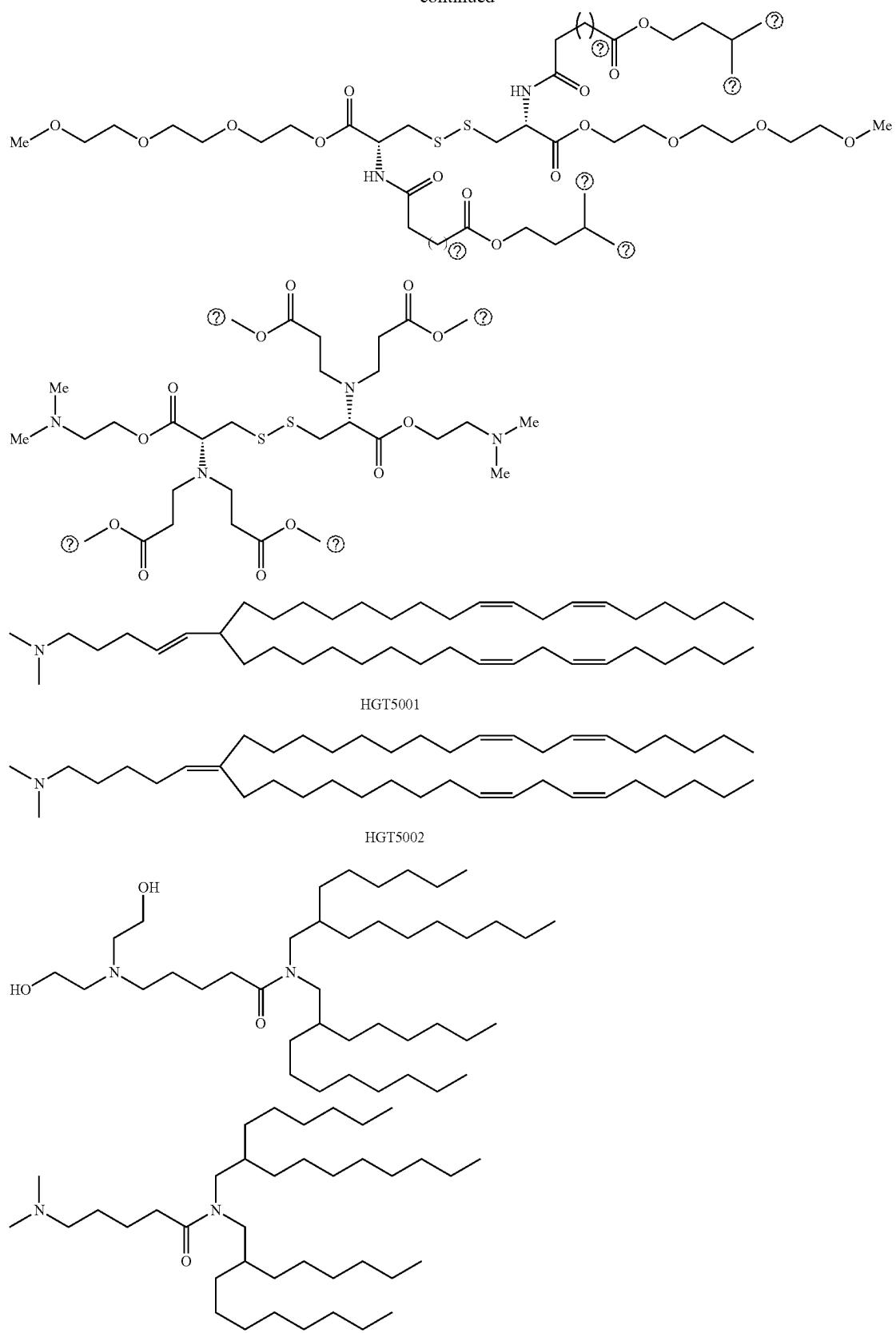


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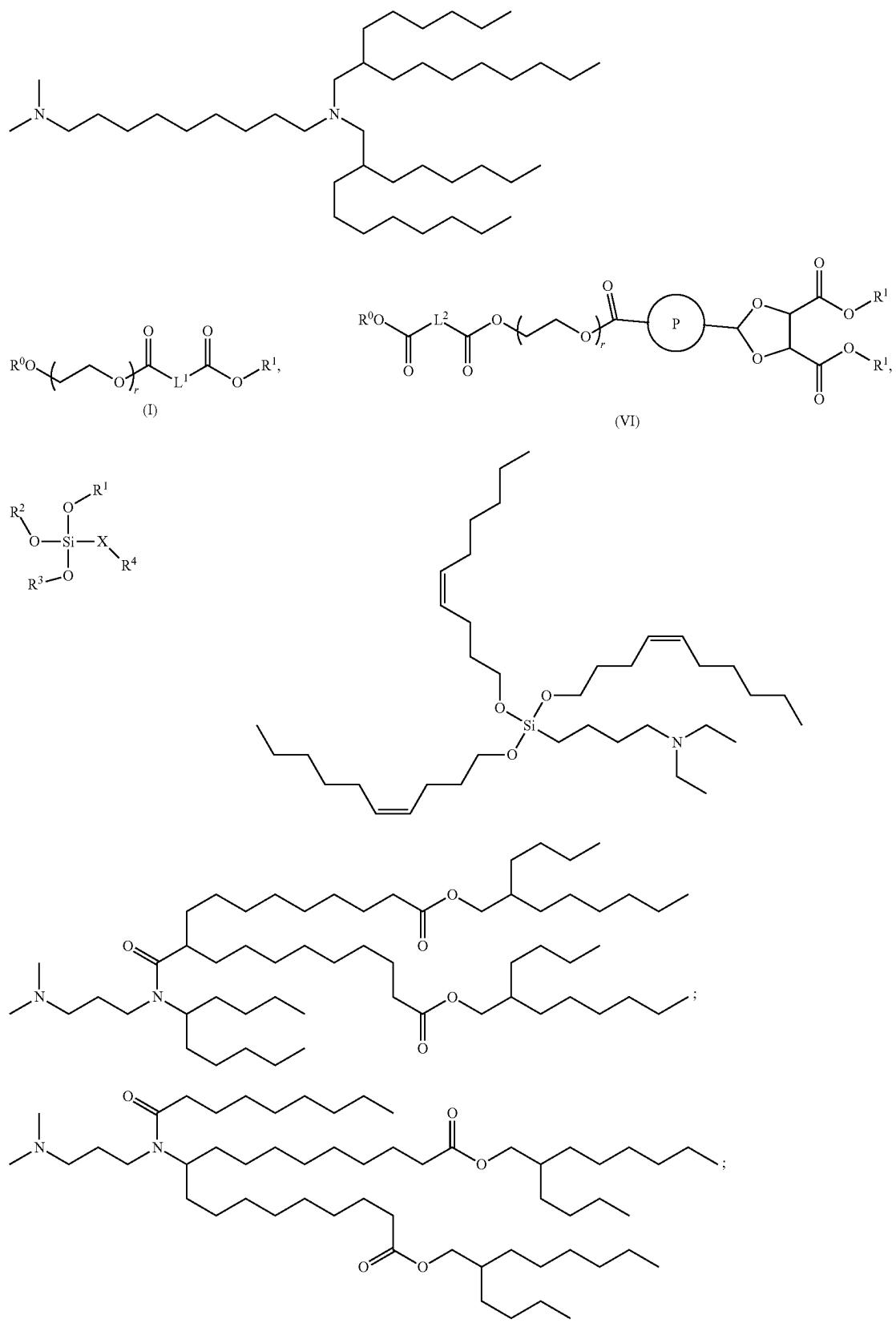
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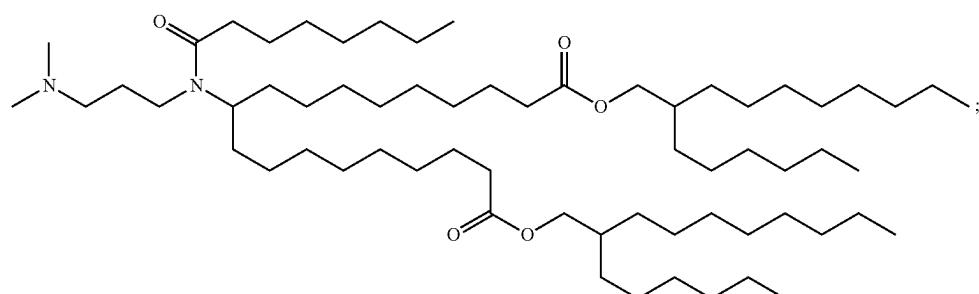
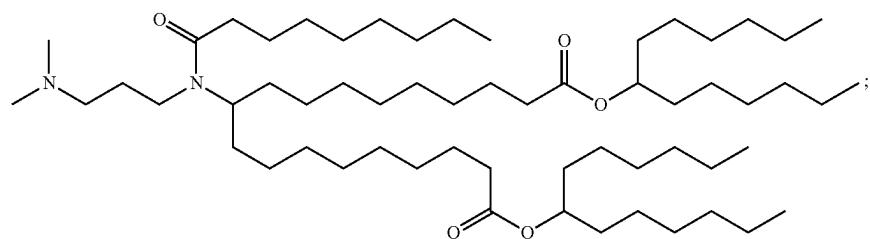
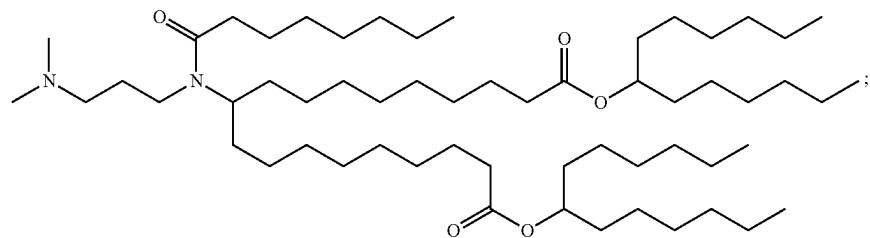
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### Neutral Lipids/Phospholipids

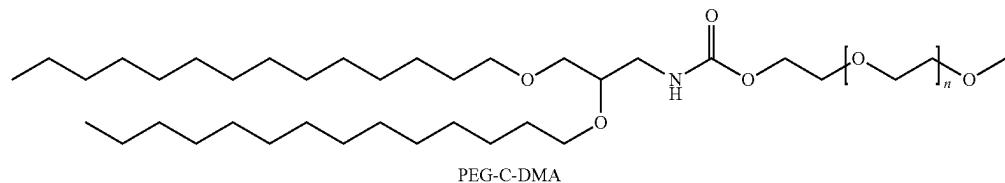
**[0618]** In some embodiments, the lipid component further comprises neutral lipids including, but not limited to, a phospholipid selected from the group consisting of 1,2-dilinoleoyl-sn-glycero-3-phosphocholine (DLPC), 1,2-dimyristoyl-sn-glycero-phosphocholine (DMPC), 1,2-dioleoyl-sn-glycero-3-phosphocholine (DOPC), 1,2-dipalmitoyl-sn-glycero-3-phosphocholine (DPPC), 1,2-distearoyl-sn-glycero-3-phosphocholine (DSPC), 1,2-diundecanoyl-sn-glycero-phosphocholine (DUPC), 1-palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine (POPC), 1,2-di-O-octadecenyl-sn-glycero-3-phosphocholine (18:0 Diether PC), 1-oleoyl-2-cholesterylhemisuccinoyl-sn-glycero-3-phosphocholine (OChemSPC), 1-hexadecyl-sn-glycero-3-phosphocholine (C16 Lyso PC), 1,2-dilinolenoyl-sn-glycero-3-phosphocholine, 1,2-diarachidonoyl-sn-glycero-3-phosphocholine, 1,2-didocosahexaenoyl-sn-glycero-3-phosphocholine, 1,2-dioleoyl-sn-glycero-3-phosphoethanolamine (DOPE), 1,2-diphytanoyl-sn-glycero-3-phosphoethanolamine (ME 16.0 PE), 1,2-distearoyl-sn-

glycero-3-phosphoethanolamine, 1,2-dilinoleoyl-sn-glycero-3-phosphoethanolamine, 1,2-dilinolenoyl-sn-glycero-3-phosphoethanolamine, 1,2-diarachidonoyl-sn-glycero-3-phosphoethanolamine, 1,2-didocosahexaenoyl-sn-glycero-3-phosphoethanolamine, 1,2-dioleoyl-sn-glycero-3-phospho-rac-(1-glycerol) sodium salt (DOPG), sphingomyelin (SM), and mixtures thereof.

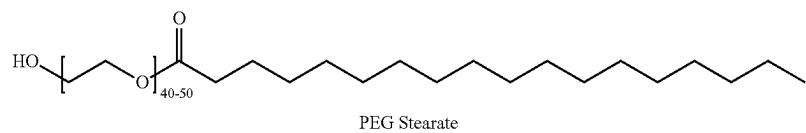
### Polymer-Conjugated Lipids

**[0619]** In some embodiments, the lipid component further comprises polymer-conjugated lipids, including, but not limited to, a PEGylated lipid selected from the group consisting of PEG-modified phosphatidylethanolamines, PEG-modified phosphatidic acids, PEG-modified ceramides, PEG-modified dialkylamines, PEG-modified diacylglycerols, PEG-modified dialkylglycerols, and mixtures thereof. For example, a PEG lipid may be PEG-c-DOMG, PEG-DMG, PEG<sub>2000</sub>-c-DMG, PEG-DLPE, PEG-DMPE, PEG-DPPC, PEG-DMA or a PEG-DSPE lipid.

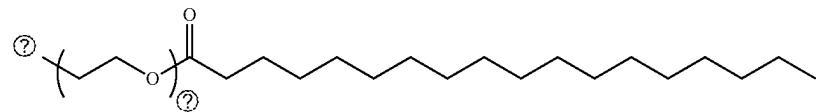
[0620] Non-exhaustive and non-limiting examples of PEG lipids include:



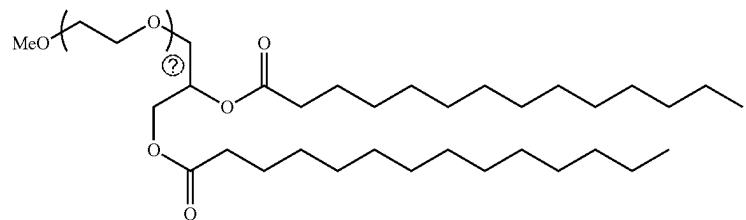
(Compound (I))



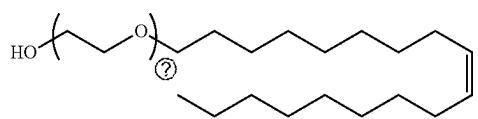
(PEG-1)



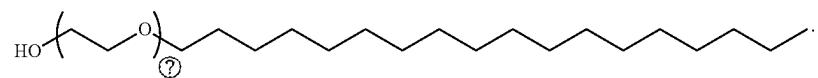
(PEG-2)



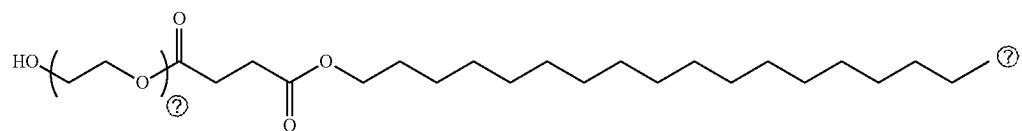
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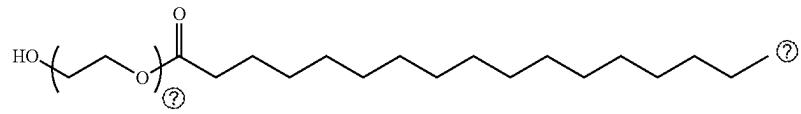
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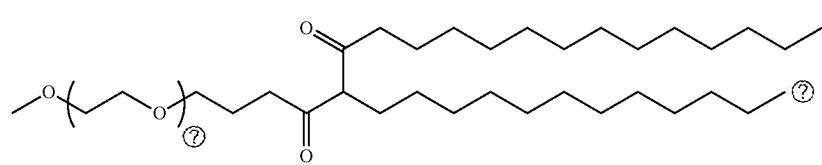
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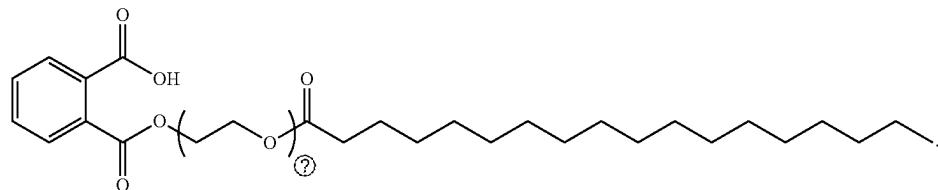


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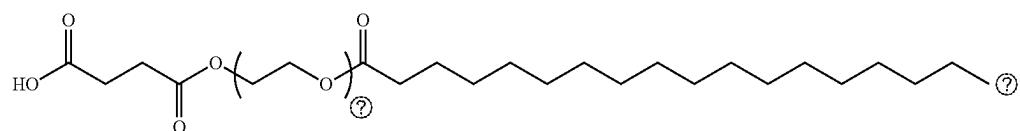


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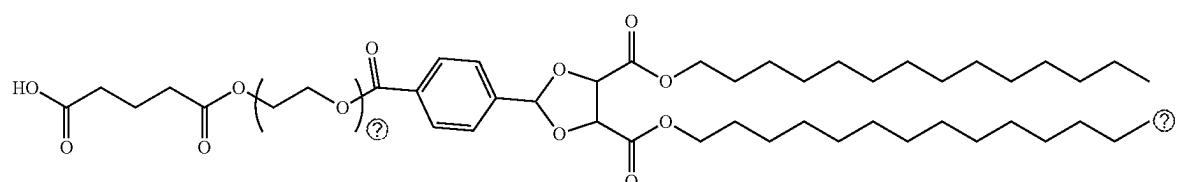
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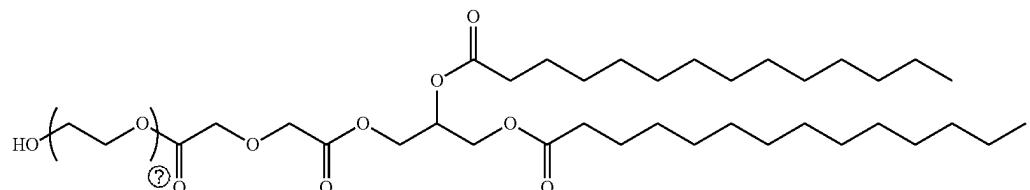
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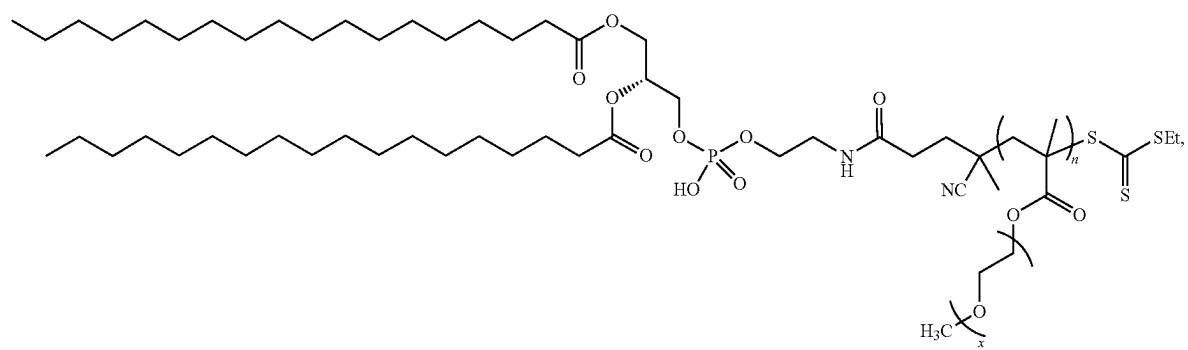
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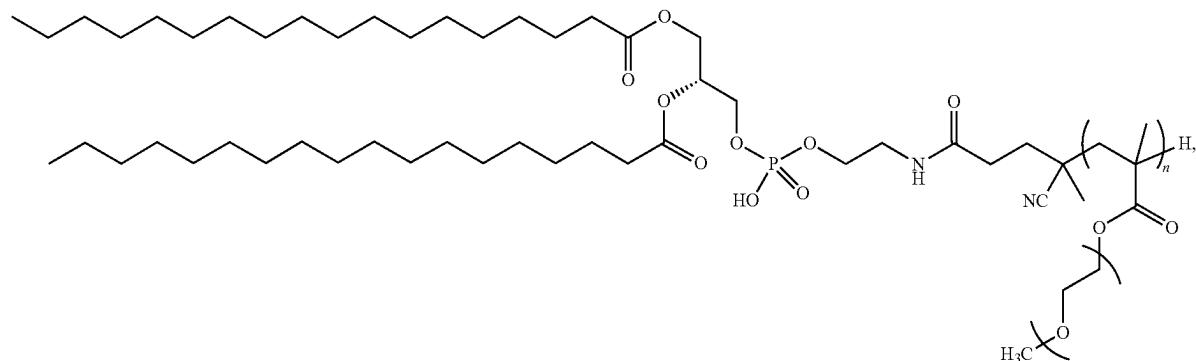
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(IIa)



(IIb)

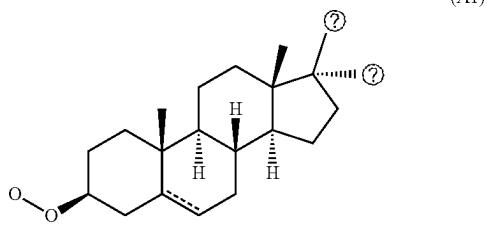
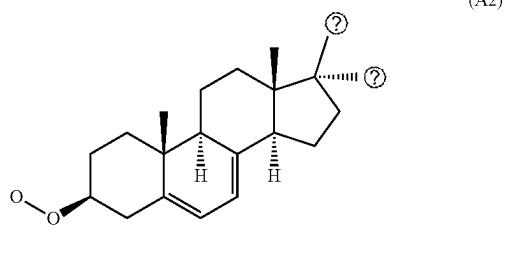
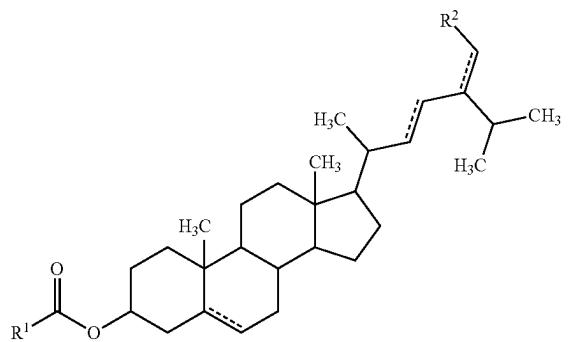
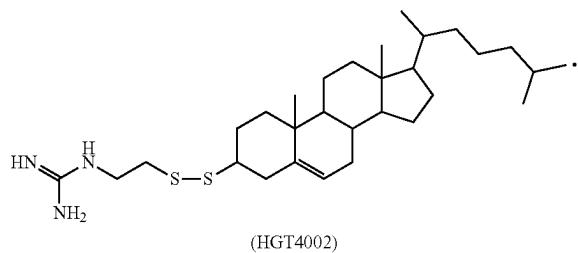


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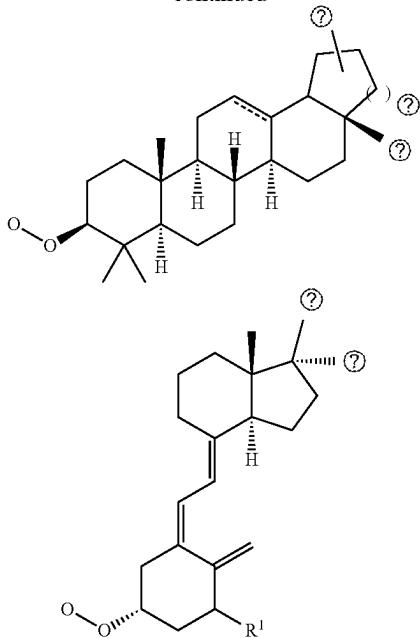
### Structural Lipids/Sterols

[0621] In some embodiments, the LNP further comprises a structural component. See generally Patel, S., et al. (2020). Nature Communications, 11(1), 1-13. In some embodiments, the structural component comprises a sterol including, but not limited to, a sterol selected from the group consisting of cholesterol, fecosterol, stigmasterol, stigmastanol, sitosterol,  $\beta$ -sitosterol, lupeol, betulin, ursolic acid, oleanolic acid, campesterol, fucosterol, brassicasterol, ergosterol, 9,11-dehydroergosterol, tomatidine, tomatine,  $\alpha$ -tocopherol, and mixtures thereof. In other embodiments, the structural lipid includes cholesterol and a corticosteroid (e.g., prednisolone, dexamethasone, prednisone, and hydrocortisone), or a combination thereof.

[0622] Non-exhaustive and non-limiting examples of structural lipids include:



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## Formulations

[0623] Nanoparticle compositions may include a lipid component and one or more additional components, such as a therapeutic and/or prophylactic. A nanoparticle composition may be designed for one or more specific applications or targets. The elements of a nanoparticle composition may be selected based on a particular application or target, and/or based on the efficacy, toxicity, expense, ease of use, availability, or other feature of one or more elements. Similarly, the particular formulation of a nanoparticle composition may be selected for a particular application or target according to, for example, the efficacy and toxicity of particular combinations of elements.

**[0624]** The lipid component of a nanoparticle composition may include, for example, a cationic lipid, a phospholipid (such as an unsaturated lipid, e.g., DOPE or DSPC), a PEG lipid, and a structural lipid. The elements of the lipid component may be provided in specific fractions.

[0625] In some embodiments, the lipid component of a nanoparticle composition includes an ionizable lipid, a phospholipid, a PEG lipid, and a structural lipid. In certain embodiments, the lipid component of the nanoparticle composition includes about 30 mol % to about 60 mol % ionizable lipid, about 0 mol % to about 30 mol % phospholipid, about 0 mol % to about 10 mol % of PEG lipid, and about 17.5 mol % to about 50 mol % structural lipid, provided that the total mol % does not exceed 100%. In some embodiments, the lipid component of the nanoparticle composition includes about 35 mol % to about 55 mol % compound of ionizable lipid, about 5 mol % to about 25 mol % phospholipid, about 0 mol % to about 10 mol % of PEG lipid, and about 30 mol % to about 40 mol % structural lipid. In a particular embodiment, the lipid component includes about 50 mol % said compound, about 10 mol % phospholipid, about 38.5 mol % structural lipid, and about 1.5 mol

% of PEG lipid. In another embodiment, the lipid component includes about 40 mol % said compound, about 20 mol % phospholipid, about 38.5 mol % structural lipid, and about 1.5 mol % of PEG lipid. In some embodiments, the phospholipid may be DOPE or DSPC. In other embodiments, the PEG lipid may be PEG-DMG and/or the structural lipid may be cholesterol.

[0626] In some embodiments, the ionizable lipids comprise between about 20 and about 60 mol % of the lipid component. In other embodiments, the ionizable lipids comprise between about 35 and about 55 mol % of the lipid component. In various embodiments, the ionizable lipids comprise about 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 35, 40, 45, 50, 55, or 60 mol % of the lipid component.

[0627] In some embodiments, the neutral lipids comprise between about 0 and about 30 mol % of the lipid component. In other embodiments, the neutral lipids comprise between about 5 and about 25 mol % of the lipid component. In various embodiments, the neutral lipids comprise about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 25, or 30 mol % of the lipid component.

[0628] In some embodiments, the polymer-conjugated lipids comprise between about 0 and about 15 mol % of the lipid component. In other embodiments, the polymer-conjugated lipids comprise between about 0.5 and about 10 mol % of the lipid component. In various embodiments, the polymer-conjugated lipids comprise about 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 5.5, 6, 6.5, 7, 7.5, 8, 8.5, 9, 9.5, 10, or 15 mol % of the lipid component.

[0629] In some embodiments, the structural component comprises about 17.5 mol % to about 50 mol % of the lipid component. In other embodiments, the structural component comprises about 30 to about 40 mol % of the lipid component. In various embodiments, the structural component comprises about 17.5, 20, 22.5, 25, 27.5, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, or 50 mol % of the lipid component.

[0630] The structural component may alternatively be expressed as a ratio relative to the lipid component. In some embodiments, the structural component is in a ratio of about 1:1 with the lipid component (sterol:lipids). In other embodiments, the structural component is in a ratio of about 1:5 with the lipid component (sterol:lipids). In various embodiments, the structural component is in a ratio of about 1:1, 1:2, 1:3, 1:4, 1:5, 1:6, 1:7, 1:8, 1:9, 1:10, 1:15, 1:20, or 1:25 with the lipid component (sterol:lipids).

[0631] Nanoparticle compositions may be designed for one or more specific applications or targets. For example, a nanoparticle composition may be designed to deliver a therapeutic and/or prophylactic such as an RNA to a particular cell, tissue, organ, or system or group thereof in a mammal's body. Physicochemical properties of nanoparticle compositions may be altered in order to increase selectivity for particular bodily targets. For instance, particle sizes may be adjusted based on the fenestration sizes of different organs. The therapeutic and/or prophylactic included in a nanoparticle composition may also be selected based on the desired delivery target or tar-gets. For example, a therapeutic and/or prophylactic may be selected for a particular indication, condition, disease, or disorder and/or for delivery to a particular cell, tissue, organ, or system or group thereof (e.g., localized or specific delivery). In certain embodiments, a nanoparticle composition may include an mRNA encoding a polypeptide of interest capable of being translated within

a cell to produce the polypeptide of interest. Such a composition may be designed to be specifically delivered to a particular organ. In some embodiments, a composition may be de-signed to be specifically delivered to a mammalian joint.

[0632] The amount of a therapeutic and/or prophylactic in a nanoparticle composition may depend on the size, composition, desired target and/or application, or other properties of the nanoparticle composition as well as on the properties of the therapeutic and/or prophylactic. For example, the amount of an RNA useful in a nanoparticle composition may depend on the size, sequence, and other characteristics of the RNA. The relative amounts of a therapeutic and/or prophylactic and other elements (e.g., lipids) in a nanoparticle composition may also vary. In some embodiments, the wt/wt ratio of the lipid component to a therapeutic and/or prophylactic in a nanoparticle composition may be from about 5:1 to about 60:1, such as 5:1, 6:1, 7:1, 8:1, 9:1, 10:1, 11:1, 12:1, 13:1, 14:1, 15:1, 16:1, 17:1, 18:1, 19:1, 20:1, 25:1, 30:1, 35:1, 40:1, 45:1, 50:1, and 60:1. For example, the wt/wt ratio of the lipid component to a therapeutic and/or prophylactic may be from about 10:1 to about 40:1. In certain embodiments, the wt/wt ratio is about 20:1. The amount of a therapeutic and/or prophylactic in a nanoparticle composition may, for example, be measured using absorption spectroscopy (e.g., ultraviolet-visible spectroscopy).

[0633] In some embodiments, the therapeutic and/or prophylactic comprises a nucleic acid component. In some embodiments, the nucleic acid component comprises RNA including, but not limited to, RNA selected from the group consisting of messenger RNA (mRNA), CRISPR RNA (crRNA), tracrRNA, single-guide RNA (sgRNA), short interfering RNA (siRNA), antisense oligonucleotides (ASO), and mixtures thereof. In other embodiments, the nucleic acid component comprises DNA including, but not limited to, DNA selected from the group consisting of linear DNA, plasmid DNA, antisense oligonucleotide, and mixtures thereof.

[0634] In some embodiments, a nanoparticle composition includes one or more RNAs, and the one or more RNAs, lipids, and amounts thereof may be selected to provide a specific N:P ratio. The N:P ratio of the composition refers to the molar ratio of nitrogen atoms in one or more lipids to the number of phosphate groups in an RNA. In general, a lower N:P ratio is preferred. The one or more RNA, lipids, and amounts thereof may be selected to provide an N:P ratio from about 2:1 to about 30:1, such as 2:1, 3:1, 4:1, 5:1, 6:1, 7:1, 8:1, 9:1, 10:1, 12:1, 14:1, 16:1, 18:1, 20:1, 22:1, 24:1, 26:1, 28:1, or 30:1. In certain embodiments, the N:P ratio may be from about 2:1 to about 8:1. In other embodiments, the N:P ratio is from about 5:1 to about 8:1. For example, the N:P ratio may be about 5.0:1, about 5.5:1, about 5.67:1, about 6.0:1, about 6.5:1, or about 7.0:1. For example, the N:P ratio may be about 5.67:1.

[0635] In some embodiments, the nucleic acid component is comprised of a modified nucleic acid. For example, an RNA may be a modified RNA. That is, an RNA may include one or more nucleobases, nucleosides, nucleotides, or linkers that are non-naturally occurring. A "modified" species may also be referred to herein as an "altered" species. Species may be modified or altered chemically, structurally,

or functionally. For example, a modified nucleobase species may include one or more substitutions that are not naturally occurring.

[0636] In certain embodiments, the present disclosure comprises methods for treating back or spine conditions or disorders. In other embodiments, the present disclosure comprises methods for treating discogenic disorders. In some embodiments, the present disclosure comprises methods for treating localized nociception, inflammation, or morphological changes associated with back or spine conditions or disorders in a subject in need thereof, the method comprising administering a therapeutically effective amount of a CRISPR-Cas composition encapsulated within or associated with a lipid nanoparticle (LNP), wherein the composition comprises one or more non-naturally occurring poly-nucleotides encoding a Cas9 protein and at least one sgRNA. In some embodiments, LNPs are administered intradiscally. In other embodiments, LNPs are administered epidurally. In some embodiments, LNPs are administered peridiscally. In some embodiments, LNPs are administered perivertebrally.

#### Physical Properties

[0637] The characteristics of a nanoparticle composition may depend on the components thereof. For example, a nanoparticle composition including cholesterol as a structural lipid may have different characteristics than a nanoparticle composition that includes a different structural lipid. Similarly, the characteristics of a nanoparticle composition may depend on the absolute or relative amounts of its components. For instance, a nanoparticle composition including a higher molar fraction of a phospholipid may have different characteristics than a nanoparticle composition including a lower molar fraction of a phospholipid. Characteristics may also vary depending on the method and conditions of preparation of the nanoparticle composition.

[0638] Nanoparticle compositions may be characterized by a variety of methods. For example, microscopy (e.g., transmission electron microscopy or scanning electron microscopy) may be used to examine the morphology and size distribution of a nanoparticle composition. Dynamic light scattering or potentiometry (e.g., potentiometric titrations) may be used to measure zeta potentials. Dynamic light scattering may also be utilized to determine particle sizes. Instruments such as the Zetasizer Nano ZS (Malvern Instruments Ltd, Malvern, Worcestershire, UK) may also be used to measure multiple characteristics of a nanoparticle composition, such as particle size, polydispersity index, and zeta potential.

[0639] The mean size of a nanoparticle composition may be between 10 nm and 1 micrometer, e.g., measured by dynamic light scattering (DLS). For example, the mean size may be from about 40 nm to about 150 nm, such as about 40 nm, 45 nm, 50 nm, 55 nm, 60 nm, 65 nm, 70 nm, 75 nm, 80 nm, 85 nm, 90 nm, 95 nm, 100 nm, 105 nm, 110 nm, 115 nm, 120 nm, 125 nm, 130 nm, 135 nm, 140 nm, 145 nm, or 150 nm. In some embodiments, the mean size of a nanoparticle composition may be from about 50 nm to about 100 nm, from about 50 nm to about 90 nm, from about 50 nm to about 80 nm, from about 50 nm to about 70 nm, from about 50 nm to about 60 nm, from about 60 nm to about 100 nm, from about 60 nm to about 90 nm, from about 60 nm to about 80 nm, from about 60 nm to about 70 nm, from about 70 nm to about 100 nm, from about 70 nm to about 90 nm, from about 70 nm to about 80 nm, from about 80 nm to

about 100 nm, from about 80 nm to about 90 nm, or from about 90 nm to about 100 nm. In certain embodiments, the mean size of a nanoparticle composition may be from about 70 nm to about 100 nm. In a particular embodiment, the mean size may be about 80 nm. In other embodiments, the mean size may be about 100 nm.

[0640] A nanoparticle composition may be relatively homogenous. A polydispersity index may be used to indicate the homogeneity of a nanoparticle composition, e.g., the particle size distribution of the nanoparticle compositions. A small (e.g., less than 0.3) polydispersity index generally indicates a narrow particle size distribution. A nanoparticle composition may have a polydispersity index from about 0 to about 0.25, such as 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.10, 0.11, 0.12, 0.13, 0.14, 0.15, 0.16, 0.17, 0.18, 0.19, 0.20, 0.21, 0.22, 0.23, 0.24, or 0.25. In some embodiments, the polydispersity index of a nanoparticle composition may be from about 0.10 to about 0.20.

[0641] The zeta potential of a nanoparticle composition may be used to indicate the electrokinetic potential of the composition. For example, the zeta potential may describe the surface charge of a nanoparticle composition. Nanoparticle compositions with relatively low charges, positive or negative, are generally desirable, as more highly charged species may interact undesirably with cells, tissues, and other elements in the body. In some embodiments, the zeta potential of a nanoparticle composition may be from about -10 mV to about +20 mV, from about -10 mV to about +15 mV, from about -10 mV to about +10 mV, from about -10 mV to about +5 mV, from about -10 mV to about 0 mV, from about -10 mV to about -5 mV, from about -5 mV to about +20 mV, from about -5 mV to about +15 mV, from about -5 mV to about +10 mV, from about -5 mV to about +5 mV, from about -5 mV to about 0 mV, from about 0 mV to about +20 mV, from about 0 mV to about +15 mV, from about 0 mV to about +10 mV, from about 0 mV to about +5 mV, from about +5 mV to about +20 mV, from about +5 mV to about +15 mV, or from about +5 mV to about +10 mV.

[0642] The efficiency of encapsulation of a therapeutic and/or prophylactic describes the amount of therapeutic and/or prophylactic that is encapsulated or otherwise associated with a nanoparticle composition after preparation, relative to the initial amount provided. The encapsulation efficiency is desirably high (e.g., close to 100%). The encapsulation efficiency may be measured, for example, by comparing the amount of therapeutic and/or prophylactic in a solution containing the nanoparticle composition before and after breaking up the nanoparticle composition with one or more organic solvents or detergents. Fluorescence may be used to measure the amount of free therapeutic and/or prophylactic (e.g., RNA) in a solution. For the nanoparticle compositions described herein, the encapsulation efficiency of a therapeutic and/or prophylactic may be at least 50%, for example 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%. In some embodiments, the encapsulation efficiency may be at least 80%. In certain embodiments, the encapsulation efficiency may be at least 90%.

[0643] A nanoparticle composition may optionally comprise one or more coatings. For example, a nanoparticle composition may be formulated in a capsule, film, or tablet having a coating. A capsule, film, or tablet including a composition described herein may have any useful size, tensile strength, hardness, or density.

**[0644]** Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. Although methods and materials similar or equivalent to those described herein may be used in the practice or testing of the present disclosure, suitable methods and materials are described below. All publications, patent applications, patents, and other references mentioned herein are incorporated by reference in their entirety. In addition, the materials, methods, and examples are illustrative only and not intended to be limiting.

**[0645]** In some embodiments, the CRISPR gene-editing system comprises one or more RNA-containing compositions. In some embodiments, the CRISPR gene-editing system further comprises one or more nanoparticles. In some embodiments, said one or more RNA-containing compositions comprises a guide RNA. In some embodiments, said one or more RNA-containing compositions comprises an mRNA. In some embodiments, said one or more RNA-containing compositions comprises an RNP (e.g., Cas9 and a guide RNA). In some embodiments, said one or more nanoparticles are lipid nanoparticles (LNP).

**[0646]** In some embodiments, the CRISPR gene-editing system comprises one or more LNPs collectively encapsulating (i) the RNA-guided nuclease or the nucleic acid encoding the RNA-guided nuclease and (ii) the at least one guide RNA or the nucleic acid encoding the at least one guide RNA. In some embodiments, the one or more LNPs comprises a first plurality of LNP encapsulating the RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and a second plurality of LNP encapsulating the at least one guide RNA or a nucleic acid encoding at least one guide RNA.

**[0647]** In some embodiments, the one or more LNP comprises a component selected from the group consisting of 3-(didodecylamino)-N1,N1,4-tridodecyl-1-piperazineethanamine (KL10), N1-[2-(didodecylamino)ethyl]-N1,N4,N4-tridodecyl-1,4-piperazinediethanamine (KL22), 14,25-ditridodecyl-15,18,21,24-tetraaza-octatriacontane (KL25), 1,2-dilinoleyoxy-N,N-dimethylaminopropane (DLin-DMA), 2,2-dilinoleyl-4-dimethylaminomethyl-[1,3]-dioxolane (DLin-K-DMA), heptatriaconta-6,9,28,31-tetraen-19-yl 4-(dimethylamino)butanoate (DLin-MC3-DMA), 2,2-dilinoleyl-4-(2-dimethylaminoethyl)-[1,3]-dioxolane (DLin-KC2-DMA), 1,2-dioleyloxy-N,N-dimethylaminopropane (DODMA), 2-[{8-[{(3.beta.)-cholest-5-en-3-yloxy]octyl}oxy]-N,N-dimethyl-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA), (2R)-2-{[8-[(3.beta.)-cholest-5-en-3-yloxy]octyl}oxy]-N,N-dimethyl-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA (2R)), (2S)-2-{[8-[(3.beta.)-cholest-5-en-3-yloxy]octyl}oxy]-N,N-dimethyl-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA (2S)), a lipid including a cyclic amine group, and a mixture thereof.

**[0648]** In some embodiments, the one or more LNP comprises a component selected from the group consisting of 1,2-dilinoleyl-sn-glycero-3-phosphocholine (DLPC), 1,2-dimyristoyl-sn-glycero-phosphocholine (DMPC), 1,2-dioleoyl-sn-glycero-3-phosphocholine (DOPC), 1,2-dipalmitoyl-sn-glycero-3-phosphocholine (DPPC), 1,2-distearoyl-sn-glycero-3-phosphocholine (DSPC), 1,2-diundecanoyl-sn-glycero-phosphocholine (DUPC), 1-palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine (POPC), 1,2-di-O-

octadecenyl-sn-glycero-3-phosphocholine (18:0 Diether PC), 1-oleoyl-2-cholesterylhemicuccinoyl-sn-glycero-3-phosphocholine (OChemSPC), 1-hexadecyl-sn-glycero-3-phosphocholine (C16 Lyso PC), 1,2-dilinolenoyl-sn-glycero-3-phosphocholine, 1,2-diarachidonoyl-sn-glycero-3-phosphocholine, 1,2-didocosahexaenoyl-sn-glycero-3-phosphocholine, 1,2-dioleoyl-sn-glycero-3-phosphoethanolamine (DOPE), 1,2-diphytanoyl-sn-glycero-3-phosphoethanolamine (ME 16.0 PE), 1,2-distearoyl-sn-glycero-3-phosphoethanolamine, 1,2-dilinoleoyl-sn-glycero-3-phosphoethanolamine, 1,2-dilinolencyl-sn-glycero-3-phosphoethanolamine, 1,2-diarachidonoyl-sn-glycero-3-phosphoethanolamine, 1,2-didocosahexaenoyl-sn-glycero-3-phosphoethanolamine, 1,2-dioleoyl-sn-glycero-3-phospho-rac-(1-glycerol) sodium salt (DOPG), sphingomyelin (SM), and a mixture thereof.

**[0649]** In some embodiments, the one or more LNP comprises a component selected from the group consisting of PEG-modified phosphatidylethanolamines, PEG-modified phosphatidic acids, PEG-modified ceramides, PEG-modified dialkylamines, PEG-modified diacylglycerols, PEG-modified dialkylglycerols, and mixtures thereof. For example, a PEG lipid may be PEG-c-DOMG, PEG-DMG, PEG-DLPE, PEG-DMPE, PEG-DPPC, PEG-DMA, a PEG-DSPE lipid, and a mixture thereof.

**[0650]** In some embodiments, the one or more LNP comprises a component selected from the group consisting of a cholesterol, fecosterol, stigmasterol, stigmastanol, sitosterol,  $\beta$ -sitosterol, lupeol, betulin, ursolic acid, oleanolic acid, campesterol, fucosterol, brassicasterol, ergosterol, 9, 11-dehydroergosterol, tomatidine, tomatine,  $\alpha$ -tocopherol, and a mixture thereof.

**[0651]** In some embodiments, use of the CRISPR gene-editing system further comprising one or more LNPs to target a gene selected from IL1R1, IL1RAP, TGFBR1, TGFBR2, IL6R, IL6ST, TNFRSF1A, TNFRSF1B, TNFRSF3, TNFRSF4, TNFRSF11A, and combinations thereof is therapeutic.

**[0652]** In some aspect, use of the CRISPR gene-editing system further comprising one or more LNPs to target TGFBR1 and/or TGFBR2 is therapeutic. In some embodiments, use of the system treats one or more joint disease or illness or one or more back or spine conditions or disorders. In some embodiments, use of the system treats one or more musculoskeletal diseases, conditions, and illnesses, including, but not limited to, Loeys-Dietz Syndrome, osteoarthritis, Marfan syndrome, aortic aneurysm (e.g., familial thoracic 3 aortic aneurysm), craniofacial abnormalities, and combinations thereof. In other embodiments, use of the system treats neoplastic diseases, conditions, and illnesses, including, but not limited to, pancreatic cancer, multiple self-healing squamous epithelioma (Ferguson-Smith disease), gastrointestinal stromal tumors (GIST), hereditary nonpolyposis colorectal cancer (Lynch Syndrome), metastatic colorectal carcinoma, bone neoplasms, anaplastic carcinoma, spindle-cell carcinoma, lung neoplasms, brain neoplasms, and combinations thereof.

**[0653]** In some embodiments, the CRISPR gene-editing system further comprising one or more LNPs to target IL1R1 and/or IL1RAP is therapeutic. In some embodiments, use of the system treats one or more joint disease or illness or one or more back or spine conditions or disorders. In some embodiments, use of the system treats one or more musculoskeletal diseases, conditions, and illnesses, includ-

ing, but not limited to, rheumatoid arthritis, gout, osteoarthritis, osteoporosis, intervertebral disc disease (IVDD), psoriatic arthritis, arthritis, polymyositis, proliferative synovitis, bone neoplasms, sarcoid myopathy, cortex bone disorders, idiopathic scoliosis, tendinopathy, myofibrillar myopathy, enthesis-related arthritis, ankylosing spondylitis, degenerative polyarthritis, arthropathy, osteitis deformans, prolapsed lumbar disc, polymyositis ossificans, idiopathic polymyositis, Luft Disease, adult-onset Still's Disease, osteoarthritis deformans, Bachet's Disease, and combinations thereof. In other embodiments, use of the system treats one or more neoplastic diseases, conditions, and illnesses, including, but not limited to, osteosarcoma, multiple myeloma, lymphoma (e.g., B-cell or cutaneous T-cell lymphoma), leukemia, thyroid carcinoma, glioma, renal cell carcinoma, chondrosarcoma, glioblastoma, melanoma, neuroblastoma, polycystic ovary syndrome, Kaposi sarcoma, squamous cell carcinoma, leiomyosarcoma, malignant peripheral nerve sheath tumor, Ewing's sarcoma, esophageal neoplasms, colon cancer, lung cancer, breast cancer, pancreatic cancer, stomach cancer, epithelial ovarian cancer, cholelithiasis, liver cancer, skin cancer, prostate cancer, cervical cancer, ovarian cancer, bladder cancer, oral cavity cancer, and combinations thereof. In other embodiments, use of the system treats one or more inflammatory diseases, conditions, and illnesses, including, but not limited to, autoinflammatory disease (AID), cryopyrin-associated periodic syndrome (CAPS), systemic lupus erythematosus (SLE), inflammatory bowel disease (IBD) and combinations thereof. In some embodiments, use of the system treats one or more cardiac diseases, conditions, and illnesses, including, but not limited to, myocardial infarction, heart failure (e.g., refractory or acute decompensated heart failure), arrhythmias, pericarditis (e.g., refractory idiopathic pericarditis), myocarditis, sepsis-induced cardiomyopathy, atherosclerosis, coronary artery disease and combinations thereof. In some embodiments, use of the system treats one or more neurological diseases, conditions, and illnesses, including, but not limited to, acute thrombotic stroke, epilepsy, multiple sclerosis, Alzheimer's Disease and combinations thereof. In some embodiments, use of the system treats one or more ophthalmic diseases, conditions, and illnesses, including, but not limited to, uveitis, scleritis, Sjogren syndrome, dry eye and combinations thereof. In some embodiments, use of the system treats one or more diseases, conditions, and illnesses, including, but not limited to, chronic kidney disease, Type 2 diabetes, gastroesophageal reflux disease (GERD), non-HP-associated peptic ulcer disease, and pulmonary fibrosis.

## B. Virus-Like Particles

**[0654]** In one aspect, the present disclosure encompasses means for delivering a CRISPR gene-editing system to a mammalian cell via a virus-like particle (VLP). In some embodiments, a CRISPR gene-editing system is delivered by a VLP. Without wishing to be bound by any particular theory, in certain embodiments, nucleic acids, when present in the particle, are resistant in aqueous solution to degradation with a nuclease. In other embodiments, proteins are protected from protease degradation while present in the particle. In some embodiments, proteins and nucleic acids encapsulated by VLPs are capable of penetrating the cellular plasma membrane.

**[0655]** In some embodiments, the CRISPR gene-editing system comprises one or more RNA-containing compositions. In some embodiments, the CRISPR gene-editing system further comprises one or more VLPs. In some embodiments, said one or more RNA-containing compositions comprises a guide RNA. In some embodiments, said one or more RNA-containing compositions comprises an mRNA. In some embodiments, said one or more RNA-containing compositions comprises an RNP (e.g., Cas9 and a guide RNA).

**[0656]** In some embodiments, the CRISPR gene-editing system comprises one or more virus-like particles collectively encapsulating (i) the RNA-guided nuclease or the nucleic acid encoding the RNA-guided nuclease and (ii) the at least one guide RNA or the nucleic acid encoding the at least one guide RNA. In some embodiments, the one or more virus-like particles comprises a first plurality of virus-like particles encapsulating the RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and a second plurality of virus-like particles encapsulating the at least one guide RNA or a nucleic acid encoding at least one guide RNA.

**[0657]** In some embodiments, use of the CRISPR gene-editing system further comprising one or more LNPs to target a gene selected from IL1R1, IL1RAP, TGFBR1, TGFBR2, IL6R, IL6ST, TNFRSF1A, TNFRSF1B, TNFRSF3, TNFRSF4, TNFRSF11A, and combinations thereof is therapeutic.

**[0658]** In one aspect, use of the CRISPR gene-editing system further comprising one or more VLPs to target TGFBR1 and/or TGFBR2 is therapeutic. In some embodiments, use of the system treats one or more joint disease or illness or one or more back or spine conditions or disorders. In some embodiments, use of the system treats one or more musculoskeletal diseases, conditions, and illnesses, including, but not limited to, Loey-Dietz Syndrome, osteoarthritis, Marfan syndrome, aortic aneurysm (e.g., familial thoracic 3 aortic aneurysm), craniofacial abnormalities, and combinations thereof. In other embodiments, use of the system treats neoplastic diseases, conditions, and illnesses, including, but not limited to, pancreatic cancer, multiple self-healing squamous epithelioma (Ferguson-Smith disease), gastrointestinal stromal tumors (GIST), hereditary nonpolyposis colorectal cancer (Lynch Syndrome), metastatic colorectal carcinoma, bone neoplasms, anaplastic carcinoma, spindle-cell carcinoma, lung neoplasms, brain neoplasms, and combinations thereof.

**[0659]** In some embodiments, the CRISPR gene-editing system further comprising one or more VLPs to target IL1R1 and/or IL1RAP is therapeutic. In some embodiments, use of the system treats one or more joint disease or illness or one or more back or spine conditions or disorders. In some embodiments, use of the system treats one or more musculoskeletal diseases, conditions, and illnesses, including, but not limited to, rheumatoid arthritis, gout, osteoarthritis, osteoporosis, intervertebral disc disease (IVDD), psoriatic arthritis, arthritis, polymyositis, proliferative synovitis, bone neoplasms, sarcoid myopathy, cortex bone disorders, idiopathic scoliosis, tendinopathy, myofibrillar myopathy, enthesis-related arthritis, ankylosing spondylitis, degenerative polyarthritis, arthropathy, osteitis deformans, prolapsed lumbar disc, polymyositis ossificans, idiopathic polymyositis, Luft Disease, adult-onset Still's Disease, osteoarthritis deformans, Bachet's Disease, and combinations thereof.

tions thereof. In other embodiments, use of the system treats one or more neoplastic diseases, conditions, and illnesses, including, but not limited to, osteosarcoma, multiple myeloma, lymphoma (e.g., B-cell or cutaneous T-cell lymphoma), leukemia, thyroid carcinoma, glioma, renal cell carcinoma, chondrosarcoma, glioblastoma, melanoma, neuroblastoma, polycystic ovary syndrome, Kaposi sarcoma, squamous cell carcinoma, leiomyosarcoma, malignant peripheral nerve sheath tumor, Ewing's sarcoma, esophageal neoplasms, colon cancer, lung cancer, breast cancer, pancreatic cancer, stomach cancer, epithelial ovarian cancer, cholelithiasis, liver cancer, skin cancer, prostate cancer, cervical cancer, ovarian cancer, bladder cancer, oral cavity cancer, and combinations thereof. In other embodiments, use of the system treats one or more inflammatory diseases, conditions, and illnesses, including, but not limited to, autoinflammatory disease (AID), cryopyrin-associated periodic syndrome (CAPS), systemic lupus erythematosus (SLE), inflammatory bowel disease (IBD) and combinations thereof. In some embodiments, use of the system treats one or more cardiac diseases, conditions, and illnesses, including, but not limited to, myocardial infarction, heart failure (e.g., refractory or acute decompensated heart failure), arrhythmias, pericarditis (e.g., refractory idiopathic pericarditis), myocarditis, sepsis-induced cardiomyopathy, atherosclerosis, coronary artery disease and combinations thereof. In some embodiments, use of the system treats one or more neurological diseases, conditions, and illnesses, including, but not limited to, acute thrombotic stroke, epilepsy, multiple sclerosis, Alzheimer's Disease and combinations thereof. In some embodiments, use of the system treats one or more ophthalmic diseases, conditions, and illnesses, including, but not limited to, uveitis, scleritis, Sjogren asthenia, dry eye and combinations thereof. In some embodiments, use of the system treats one or more diseases, conditions, and illnesses, including, but not limited to, chronic kidney disease, Type 2 diabetes, gastroesophageal reflux disease (GERD), non-HP-associated peptic ulcer disease, and pulmonary fibrosis.

#### C. Miscellaneous Modes of Delivery

##### 1. Liposomes

**[0660]** In some embodiments, nucleic acids encoding a CRISPR gene-editing system targeting a gene selected from IL1R1, IL1R2, IL1RAP, IL1RL1, TGFBR1, TGFBR2, IL6R, IL6ST, TNFRSF1A, TNFRSF1B, TNFRSF3, TNFRSF4, TNFRSF11A, and combinations thereof (e.g., Cas9 or gRNA) are entrapped in liposomes bearing positive charges on their surface (e.g., lipofectins), which can be tagged with antibodies against cell surface antigens of the target cells. These delivery vehicles can also be used to deliver Cas9 protein/gRNA complexes.

**[0661]** In some embodiments, the CRISPR gene-editing system comprises one or more RNA-containing compositions. In some embodiments, the CRISPR gene-editing system further comprises one or more liposomes. In some embodiments, said one or more RNA-containing compositions comprises a guide RNA. In some embodiments, said one or more RNA-containing compositions comprises an mRNA. In some embodiments, said one or more RNA-containing compositions comprises an RNP (e.g., Cas9 and a guide RNA).

**[0662]** In some embodiments, wherein the composition comprises one or more liposomes collectively encapsulating (i) the RNA-guided nuclease or the nucleic acid encoding the RNA-guided nuclease and (ii) the at least one guide RNA or the nucleic acid encoding the at least one guide RNA. In some embodiments, the one or more liposomes comprises a first plurality of liposomes encapsulating the RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and a second plurality of liposomes encapsulating the at least one guide RNA or a nucleic acid encoding at least one guide RNA.

**[0663]** In some embodiments, use of the CRISPR gene-editing system further comprising one or more LNPs to target a gene selected from IL1R1, IL1RAP, TGFBR1, TGFBR2, IL6R, IL6ST, TNFRSF1A, TNFRSF1B, TNFRSF3, TNFRSF4, TNFRSF11A, and combinations thereof is therapeutic.

**[0664]** In one aspect, use of the CRISPR gene-editing system further comprising one or more liposomes to target TGFBR1 and/or TGFBR2 is therapeutic. In some embodiments, use of the system treats one or more joint disease or illness or one or more back or spine conditions or disorders. In some embodiments, use of the system treats one or more musculoskeletal diseases, conditions, and illnesses, including, but not limited to, Loeys-Dietz Syndrome, osteoarthritis, Marfan syndrome, aortic aneurysm (e.g., familial thoracic 3 aortic aneurysm), craniofacial abnormalities, and combinations thereof. In other embodiments, use of the system treats neoplastic diseases, conditions, and illnesses, including, but not limited to, pancreatic cancer, multiple self-healing squamous epithelioma (Ferguson-Smith disease), gastrointestinal stromal tumors (GIST), hereditary nonpolyposis colorectal cancer (Lynch Syndrome), metastatic colorectal carcinoma, bone neoplasms, anaplastic carcinoma, spindle-cell carcinoma, lung neoplasms, brain neoplasms, and combinations thereof.

**[0665]** In some embodiments, the CRISPR gene-editing system further comprising one or more liposomes to target IL1R1 and/or IL1RAP is therapeutic. In some embodiments, use of the system treats one or more joint disease or illness or one or more back or spine conditions or disorders. In some embodiments, use of the system treats one or more musculoskeletal diseases, conditions, and illnesses, including, but not limited to, rheumatoid arthritis, gout, osteoarthritis, osteoporosis, intervertebral disc disease (IVDD), psoriatic arthritis, arthritis, polymyositis, proliferative synovitis, bone neoplasms, sarcoid myopathy, cortex bone disorders, idiopathic scoliosis, tendinopathy, myofibrillar myopathy, enthesitis-related arthritis, ankylosing spondylitis, degenerative polyarthritis, arthropathy, osteitis deformans, prolapsed lumbar disc, polymyositis ossificans, idiopathic polymyositis, Luft Disease, adult-onset Still's Disease, osteoarthritis deformans, Bachet's Disease, and combinations thereof. In other embodiments, use of the system treats one or more neoplastic diseases, conditions, and illnesses, including, but not limited to, osteosarcoma, multiple myeloma, lymphoma (e.g., B-cell or cutaneous T-cell lymphoma), leukemia, thyroid carcinoma, glioma, renal cell carcinoma, chondrosarcoma, glioblastoma, melanoma, neuroblastoma, polycystic ovary syndrome, Kaposi sarcoma, squamous cell carcinoma, leiomyosarcoma, malignant peripheral nerve sheath tumor, Ewing's sarcoma, esophageal neoplasms, colon cancer, lung cancer, breast cancer, pancreatic cancer, stomach cancer, epithelial ovarian cancer,

colorectal cancer, liver cancer, skin cancer, prostate cancer, cervical cancer, ovarian cancer, bladder cancer, oral cavity cancer, and combinations thereof. In other embodiments, use of the system treats one or more inflammatory diseases, conditions, and illnesses, including, but not limited to, autoinflammatory disease (AID), cryopyrin-associated periodic syndrome (CAPS), systemic lupus erythematosus (SLE), inflammatory bowel disease (IBD) and combinations thereof. In some embodiments, use of the system treats one or more cardiac diseases, conditions, and illnesses, including, but not limited to, myocardial infarction, heart failure (e.g., refractory or acute decompensated heart failure), arrhythmias, pericarditis (e.g., refractory idiopathic pericarditis), myocarditis, sepsis-induced cardiomyopathy, atherosclerosis, coronary artery disease and combinations thereof. In some embodiments, use of the system treats one or more neurological diseases, conditions, and illnesses, including, but not limited to, acute thrombotic stroke, epilepsy, multiple sclerosis, Alzheimer's Disease and combinations thereof. In some embodiments, use of the system treats one or more ophthalmic diseases, conditions, and illnesses, including, but not limited to, uveitis, scleritis, Sjogren asthenia, dry eye and combinations thereof. In some embodiments, use of the system treats one or more diseases, conditions, and illnesses, including, but not limited to, chronic kidney disease, Type 2 diabetes, gastroesophageal reflux disease (GERD), non-HP-associated peptic ulcer disease, and pulmonary fibrosis.

## 2. Lipid Nanocrystals (LNC)

**[0666]** In one aspect, the present disclosure encompasses means for delivering a CRISPR gene-editing system to a mammalian cell via a lipid nanocrystal (LNC). In some embodiments, a CRISPR gene-editing system is delivered by a LNC. Without wishing to be bound by any particular theory, in certain embodiments, nucleic acids, when present in the nanocrystal, are resistant in aqueous solution to degradation with a nuclease. In other embodiments, proteins are protected from protease degradation while present in the nanocrystal. In some embodiments, proteins and nucleic acids encapsulated by nanocrystal are capable of penetrating the cellular plasma membrane.

**[0667]** In some embodiments, the CRISPR gene-editing system comprises one or more RNA-containing compositions. In some embodiments, the CRISPR gene-editing system further comprises one or more nanocrystals. In some embodiments, said one or more RNA-containing compositions comprises a guide RNA. In some embodiments, said one or more RNA-containing compositions comprises an mRNA. In some embodiments, said one or more RNA-containing compositions comprises an RNP (e.g., Cas9 and a guide RNA). In some embodiments, said one or more nanocrystals are lipid nanocrystals (LNC).

**[0668]** In some embodiments, the CRISPR gene-editing system comprises one or more LNCs collectively encapsulating (i) the RNA-guided nuclease or the nucleic acid encoding the RNA-guided nuclease and (ii) the at least one guide RNA or the nucleic acid encoding the at least one guide RNA. In some embodiments, the one or more LNCs comprises a first plurality of LNC encapsulating the RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and a second plurality of LNC encapsulating the at least one guide RNA or a nucleic acid encoding at least one guide RNA.

**[0669]** In some embodiments, use of the CRISPR gene-editing system further comprising one or more LNP to target a gene selected from IL1R1, IL1RAP, TGFBR1, TGFBR2, IL6R, IL6ST, TNFRSF1A, TNFRSF1B, TNFRSF3, TNFRSF4, TNFRSF11A, and combinations thereof is therapeutic.

**[0670]** In one aspect, use of the CRISPR gene-editing system further comprising one or more LNCs to target TGFBR1 and/or TGFBR2 is therapeutic. In some embodiments, use of the system treats one or more joint disease or illness or one or more back or spine conditions or disorders. In some embodiments, use of the system treats one or more musculoskeletal diseases, conditions, and illnesses, including, but not limited to, Loeys-Dietz Syndrome, osteoarthritis, Marfan syndrome, aortic aneurysm (e.g., familial thoracic 3 aortic aneurysm), craniofacial abnormalities, and combinations thereof. In other embodiments, use of the system treats neoplastic diseases, conditions, and illnesses, including, but not limited to, pancreatic cancer, multiple self-healing squamous epithelioma (Ferguson-Smith disease), gastrointestinal stromal tumors (GIST), hereditary nonpolyposis colorectal cancer (Lynch Syndrome), metastatic colorectal carcinoma, bone neoplasms, anaplastic carcinoma, spindle-cell carcinoma, lung neoplasms, brain neoplasms, and combinations thereof.

**[0671]** In some embodiments, the CRISPR gene-editing system further comprising one or more LNCs to target IL1R1 and/or IL1RAP is therapeutic. In some embodiments, use of the system treats one or more joint disease or illness or one or more back or spine conditions or disorders. In some embodiments, use of the system treats one or more musculoskeletal diseases, conditions, and illnesses, including, but not limited to, rheumatoid arthritis, gout, osteoarthritis, osteoporosis, intervertebral disc disease (IVDD), psoriatic arthritis, arthritis, polymyositis, proliferative synovitis, bone neoplasms, sarcoid myopathy, cortex bone disorders, idiopathic scoliosis, tendinopathy, myofibrillar myopathy, enthesis-related arthritis, ankylosing spondylitis, degenerative polyarthritis, arthropathy, osteitis deformans, prolapsed lumbar disc, polymyositis ossificans, idiopathic polymyositis, Luft Disease, adult-onset Still's Disease, osteoarthritis deformans, Bachet's Disease, and combinations thereof. In other embodiments, use of the system treats one or more neoplastic diseases, conditions, and illnesses, including, but not limited to, osteosarcoma, multiple myeloma, lymphoma (e.g., B-cell or cutaneous T-cell lymphoma), leukemia, thyroid carcinoma, glioma, renal cell carcinoma, chondrosarcoma, glioblastoma, melanoma, neuroblastoma, polycystic ovary syndrome, Kaposi sarcoma, squamous cell carcinoma, leiomyosarcoma, malignant peripheral nerve sheath tumor, Ewing's sarcoma, esophageal neoplasms, colon cancer, lung cancer, breast cancer, pancreatic cancer, stomach cancer, epithelial ovarian cancer, cholelithiasis, liver cancer, skin cancer, prostate cancer, cervical cancer, ovarian cancer, bladder cancer, oral cavity cancer, and combinations thereof. In other embodiments, use of the system treats one or more inflammatory diseases, conditions, and illnesses, including, but not limited to, autoinflammatory disease (AID), cryopyrin-associated periodic syndrome (CAPS), systemic lupus erythematosus (SLE), inflammatory bowel disease (IBD) and combinations thereof. In some embodiments, use of the system treats one or more cardiac diseases, conditions, and illnesses, including, but not limited to, myocardial infarction, heart failure

(e.g., refractory or acute decompensated heart failure), arrhythmias, pericarditis (e.g., refractory idiopathic pericarditis), myocarditis, sepsis-induced cardiomyopathy, atherosclerosis, coronary artery disease and combinations thereof. In some embodiments, use of the system treats one or more neurological diseases, conditions, and illnesses, including, but not limited to, acute thrombotic stroke, epilepsy, multiple sclerosis, Alzheimer's Disease and combinations thereof. In some embodiments, use of the system treats one or more ophthalmic diseases, conditions, and illnesses, including, but not limited to, uveitis, scleritis, Sjogren asthenia, dry eye and combinations thereof. In some embodiments, use of the system treats one or more diseases, conditions, and illnesses, including, but not limited to, chronic kidney disease, Type 2 diabetes, gastroesophageal reflux disease (GERD), non-HP-associated peptic ulcer disease, and pulmonary fibrosis.

## VII. Pharmaceutical Compositions

**[0672]** In one aspect, the present disclosure encompasses pharmaceutical compositions comprising a CRISPR gene-editing system for treatment of a mammal in need thereof. In some embodiments, the CRISPR gene-editing system targets a gene selected from IL1R1, IL1R2, IL1RAP, IL1RL1, TGFBR1, TGFBR2, IL6R, IL6ST, TNFRSF1A, TNFRSF1B, TNFRSF3, TNFRSF4, TNFRSF11A, and combinations thereof. In some embodiments, the mammal is selected from a human, a dog, a horse, and a cat.

### A. IL1A

**[0673]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL1A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL1A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 769-786. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some

DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL1A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

### B. IL1B

**[0674]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL1B gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL1B gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 787-805. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some

embodiments, the pharmaceutical composition targeting an IL1B gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

### C. IL1R1

**[0675]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL1R1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL1R1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 806-839. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL1R1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

myalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

### D. IL1RAP

**[0676]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL1RAP gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL1RAP gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 840-887. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL1RAP gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### E. IL4

[0677] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL4 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL4 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 888-911. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL4 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### F. IL6R

[0678] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL6R gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL6R gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 929-963. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL6R gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### G. IL6ST

[0679] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL6ST gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a

nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL6ST gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 964-990. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL6ST gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### H. TNFRSF1A

**[0680]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TNFRSF1A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TNFRSF1A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2575-2622. In some

embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TNFRSF1A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### I. TNFRSF1B

**[0681]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TNFRSF1B gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TNFRSF1B gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2623-2670. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In

some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TNFRSF1B gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### J. TNFRSF3

**[0682]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TNFRSF3 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TNFRSF3 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1664-1684. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TNFRSF3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TNFRSF3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### K. TNFRSF4

**[0683]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TNFRSF4 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TNFRSF4 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1685-1711. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments,

steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TNFRSF4 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondyloarthritis, plantar fasciitis, degenerative polyarthritits, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### L. TNFRSF11A

**[0684]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TNFRSF11A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TNFRSF11A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1711-1759. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000.

PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TNFRSF11A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondyloarthritis, plantar fasciitis, degenerative polyarthritits, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### M. ADAM17

**[0685]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an ADAM17 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an ADAM17 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1-48. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-

PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an ADAM17 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### N. ADAMTS1

**[0686]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an ADAMTS1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an ADAMTS1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 49-96. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition tar-

geting an ADAMTS1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### O. ADAMTS5

**[0687]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an ADAMTS1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an ADAMTS1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 97-144. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an ADAMTS1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic

arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### P. ADM

**[0688]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an ADM gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an ADM gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 145-192. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an ADM gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### Q. ATP1A1

**[0689]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an ATP1A1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an ATP1A1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 193-240. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an ATP1A1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

philic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### R. BDNF

**[0690]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an BDNF gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an BDNF gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 241-281. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an BDNF gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### S. CALCA

**[0691]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CALCA

gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CALCA gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 282-301. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CALCA gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### T. CALCB

**[0692]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CALCB gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CALCB gene, and an LNP system

selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 302-318. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CALCB gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondyloarthritis, plantar fasciitis, degenerative polyarthritidis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### U. CALCRL

**[0693]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CALCRL gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CALCRL gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 319-340. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from

about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CALCRL gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondyloarthritis, plantar fasciitis, degenerative polyarthritidis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### V. CCL2

**[0694]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CCL2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CCL2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 341-357. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cho-

lesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CCL2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### W. CCL3

**[0695]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CCL3 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CCL3 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 358-374. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component

comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CCL3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### X. CCL5

**[0696]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CCL5 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CCL5 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 375-391. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component

comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CCL5 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### Y. CCL7

**[0697]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CCL7 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CCL7 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 392-408. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some

embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CCL7 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### Z. CCL20

**[0698]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CCL20 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CCL20 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 409-425. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CCL20 gene is formulated for treating one or more disorders

selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### AA. CCN2

**[0699]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CCN2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CCN2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 426-473. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CCN2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar

der (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### BB. CCR7

**[0700]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CCR7 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CCR7 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 474-517. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CCR7 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar

fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### CC. CRCP

**[0701]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CRCP gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CRCP gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 518-534. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CRCP gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### DD. CXCL1

**[0702]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CXCL1

gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CXCL1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 535-551. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CXCL1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### EE. CXCL2

**[0703]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CXCL2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CXCL2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments,

the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 552-568. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CXCL2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### FF. CXCL3

**[0704]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CXCL3 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CXCL3 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 569-585. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38,

about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the pharmaceutical composition targeting an CXCL3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### GG. CXCL5

**[0705]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CXCL5 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CXCL5 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 586-602. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl

modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CXCL5 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### HH. CXCL6

**[0706]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CXCL6 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CXCL6 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 603-619. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diam-

PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CXCL6 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### II. CXCL8

**[0707]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CXCL8 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CXCL8 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 620-636. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diam-

eter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CXCL8 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### JJ. CXCR1

**[0708]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CXCR1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CXCR1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 637-655. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CXCR1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis,

post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### KK. CXCR2

**[0709]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an CXCR2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an CXCR2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 656-672. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an CXCR2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain,

lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### LL. FGF2

**[0710]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an FGF2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an FGF2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 673-720. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an FGF2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### MM. FGFR1

**[0711]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an FGFR1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an FGFR1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 721-768. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an FGFR1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### NN. IL10

**[0712]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL10

gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL10 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 991-1007. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL10 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### OO. IL10RA

**[0713]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL10RA gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL10RA gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments,

the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1008-1055. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL10RA gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### PP. IL10RB

**[0714]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL10RB gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL10RB gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1056-1082. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38,

about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL10RB gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

QQ. IL13

**[0715]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL13 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL13 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1083-1104. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component.

or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL13 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

RR. IL13RA1

**[0716]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL13RA1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL13RA1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1105-1130. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed

herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL13RA1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### SS. IL13RA2

**[0717]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL13RA2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL13RA2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1131-1147. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL13RA2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL13RA2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### TT. IL17A

**[0718]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL17A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL17A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1148-1173. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diam-

eter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL17A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### UU. IL17RA

**[0719]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL17RA gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL17RA gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1174-1221. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL17RA gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis,

post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### VV. IL18

**[0720]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL18 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL18 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1222-1238. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL18 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain,

lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### WW. IL18R1

**[0721]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL18R1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL18R1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1239-1262. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL18R1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### XX. IL18RAP

**[0722]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an IL18RAP gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an IL18RAP gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1263-1310. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an IL18RAP gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### YY. MMP1

**[0723]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MMP1

gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MMP1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1311-1343. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

## ZZ. MMP2

**[0724]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MMP2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MMP2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments,

the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1344-1391. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

## AAA. MMP3

**[0725]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MMP3 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MMP3 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1392-1417. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38,

about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

### BBB. MMP7

**[0726]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MMP7 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MMP7 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1418-1436. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some

or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP7 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

### CCC. MMP8

**[0727]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MMP8 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MMP8 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1437-1474. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some

embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP8 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### DDD. MMP10

**[0728]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MMP10 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MMP10 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1475-1497. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP10 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

ments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP10 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### EEE. MMP12

**[0729]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MMP12 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MMP12 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1498-1541. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP12 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP12 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### FFF. MMP13

**[0730]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MMP13 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MMP13 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1542-1568. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MMP13 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness

(e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### GGG. MRGPRX2

**[0731]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an MRGPRX2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an MRGPRX2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1569-1585. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an MRGPRX2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal con-

dition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### HHH. NGF

**[0732]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an NGF gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an NGF gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1586-1628. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an NGF gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

tious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### III. NGFR

**[0733]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an NGFR gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an NGFR gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1629-1676. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an NGFR gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

## JJJ. NTF3

[0734] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an NTF3 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an NTF3 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1725-1746. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an NTF3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

## KKK. NTF4

[0735] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an NTF4 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a

nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an NTF4 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1677-1724. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an NTF4 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

## LLL. NTRK1

[0736] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an NTRK1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an NTRK1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1747-1794. In some embodiments, the

LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an NTRK1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### MMM. NTRK2

**[0737]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an NTRK2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an NTRK2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1795-1842. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In

some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an NTRK2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### NNN. RAMP1

**[0738]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an RAMP1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an RAMP1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1843-1859. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component

comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an RAMP1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### OOO. SCN1A

[0739] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN1A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN1A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1860-1907. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated

Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an SCN1A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### PPP. SCN2A

[0740] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN2A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN2A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 1908-1955. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of

DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an SCN2A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### QQQ. SCN3A

**[0741]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN3A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN3A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 1956-2003. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some

embodiments, the pharmaceutical composition targeting an SCN3A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### RRR. SCN4A

**[0742]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN4A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN4A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2004-2051. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an SCN4A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibro-

myalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### SSS. SCN5A

**[0743]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN5A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN5A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2052-2099. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an SCN5A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

(iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### TTT. SCN8A

**[0744]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN8A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN8A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2100-2147. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an SCN8A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

## UUU. SCN9A

[0745] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN9A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN9A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2148-2195. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an SCN9A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritits, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

## VVV. SCN10A

[0746] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN10A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a

nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN10A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2196-2243. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an SCN10A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritits, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

## WWW. SCN11A

[0747] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an SCN11A gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an SCN11A gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA

sequence selected from SEQ ID NOS: 2244-2291. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an SCN11A gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

### XXX. TAC1

**[0748]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TAC1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TAC1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 2292-2308. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38,

about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TACT gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

### YYY. TAC3

**[0749]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TAC3 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TAC3 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOS: 2309-2325. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more,

or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TAC3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### ZZZ. TACR1

**[0750]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TACR1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TACR1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2326-2373. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments,

embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TACR1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### AAAA. TACR2

**[0751]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TACR2 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA targeting an TACR2 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2374-2421. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments,

PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TACR2 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### BBBB. TACR3

**[0752]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TACR3 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TACR3 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2422-2469. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TACR3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

tical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TACR3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### CCCC. TIMP1

**[0753]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TIMP1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TIMP1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2470-2509. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TIMP1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### DDDD. TIMP3

**[0754]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TIMP3 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TIMP3 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2510-2557. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TIMP3 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc

disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

#### EEEE. TNF

**[0755]** In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an TNF gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an TNF gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2558-2574. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an TNF gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

FFFF. YAP1

[0756] In some embodiments, the disclosure provides one or more pharmaceutical compositions targeting an YAP1 gene, the composition comprising a plurality of LNPs, wherein the LNPs comprise an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease and at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting an YAP1 gene, and an LNP system selected from any one of LNP001 to LNP240. In some embodiments, the at least one guide RNA has a crRNA sequence selected from SEQ ID NOs: 2671-2718. In some embodiments, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments, the steroid component comprises cholesterol. In some embodiments, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000. In some embodiments, the plurality of LNPs have an average diameter between about 60 nm and about 120 nm. In some embodiments, the concentration of LNPs in the pharmaceutical composition is between 0.001 and 3 mg/ml. In some embodiments, the pharmaceutical composition targeting an YAP1 gene is formulated for treating one or more disorders selected from (i) a joint disease or illness (e.g., arthritis, post-traumatic arthritis, osteoarthritis, rheumatoid arthritis, psoriatic arthritis, lupus, ankylosing spondylitis, reactive arthritis, septic arthritis, juvenile idiopathic arthritis, fibromyalgia, crystal arthropathies, gout, pseudogout, or CPPD disease/chondrocalcinosis), (ii) a spinal condition or disorder (e.g. discogenic disease, low back pain, neck pain, lumbar pain, degenerative disc disease, annual ligament tear, herniated disc, spinal facet joint arthritis, intervertebral disc disease, spondylosis, painful scoliosis, or spinal stenosis), (iii) a musculoskeletal disorder (e.g. fibrosis, scarring, infectious intermittent joint effusion, spondylarthritis, plantar fasciitis, degenerative polyarthritis, hemophilic arthropathy, inflammatory myopathy with abundant macrophages, or polymyositis), and/or any combinations thereof.

### VIII. Administration Routes

[0757] The methods and compositions herein described encompass the use of pharmaceutical compositions comprising a CRISPR gene-editing system as an active ingredient.

[0758] Depending on the method/route of administration, pharmaceutical dosage forms come in several types. These include many kinds of liquid, solid, and semisolid dosage forms. Common pharmaceutical dosage forms include pill, tablet, or capsule, drink or syrup, and natural or herbal form such as plant or food of sorts, among many others. Notably, the route of administration (ROA) for drug delivery is dependent on the dosage form of the substance in question. A liquid pharmaceutical dosage form is the liquid form of a dose of a chemical compound used as a drug or medication intended for administration or consumption.

[0759] As described below, a composition of the present disclosure can be delivered to a subject subcutaneously (e.g., intra-articular or intradiscal injection), dermally (e.g., transdermally via patch), and/or via implant. Exemplary pharmaceutical dosage forms include, e.g., pills, osmotic delivery systems, elixirs, emulsions, hydrogels, suspensions, syrups, capsules, tablets, orally dissolving tablets (ODTs), gel capsules, thin films, adhesive topical patches, lollipops, lozenges, chewing gum, dry powder inhalers (DPIs), vaporizers, nebulizers, metered dose inhalers (MDIs), ointments, transdermal patches, intradermal implant.

[0760] As used herein, "dermal delivery" or "dermal administration" can refer to a route of administration wherein the pharmaceutical dosage form is taken to, or through, the dermis (i.e., layer of skin between the epidermis (with which it makes up the cutis) and subcutaneous tissues). "Subcutaneous delivery" can refer to a route of administration wherein the pharmaceutical dosage form is to or beneath the subcutaneous tissue layer.

[0761] Methods of formulating suitable pharmaceutical compositions are known in the art, see, e.g., Remington: The Science and Practice of Pharmacy, 21st ed., 2005; and the books in the series Drugs and the Pharmaceutical Sciences: a Series of Textbooks and Monographs (Dekker, N.Y.). For example, solutions or suspensions used for parenteral, intradermal, or subcutaneous application can include the following components: a sterile diluent such as water for injection, saline solution, fixed oils, polyethylene glycols, glycerin, propylene glycol or other synthetic solvents; antibacterial agents such as benzyl alcohol or methyl parabens; antioxidants such as ascorbic acid or sodium bisulfate; chelating agents such as ethylenediaminetetraacetic acid; buffers such as acetates, citrates or phosphates and agents for the adjustment of tonicity such as sodium chloride or dextrose. pH can be adjusted with acids or bases, such as hydrochloric acid or sodium hydroxide. The parenteral preparation can be enclosed in ampoules, disposable syringes or multiple dose vials made of glass or plastic.

[0762] Pharmaceutical compositions suitable for injectable use can include sterile aqueous solutions (where water soluble) or dispersions and sterile powders for the extemporaneous preparation of sterile injectable solutions or dispersion. For intravenous administration, suitable carriers include physiological saline, bacteriostatic water, Cremophor EL™ (BASF, Parsippany, N.J.) or phosphate buffered saline (PBS). In all cases, the composition must be sterile and should be fluid to the extent that easy syringability exists. It should be stable under the conditions of manufacture and storage and must be preserved against the contaminating action of microorganisms such as bacteria and fungi. The carrier can be a solvent or dispersion medium containing, for example, water, ethanol, polyol (for example, glycerol, propylene glycol, and liquid polyethylene glycol), and

the like), and suitable mixtures thereof. The proper fluidity can be maintained, for example, by the use of a coating such as lecithin, by the maintenance of the required particle size in the case of dispersion and by the use of surfactants. Prevention of the action of microorganisms can be achieved by various antibacterial and antifungal agents, for example, parabens, chlorobutanol, phenol, ascorbic acid, thimerosal, and the like. In many cases, it will be preferable to include isotonic agents, for example, sugars, polyalcohols such as mannitol, sorbitol, sodium chloride in the composition. Prolonged absorption of the injectable compositions can be brought about by including in the composition an agent that delays absorption, for example, aluminum monostearate and gelatin.

[0763] Sterile injectable solutions can be prepared by incorporating the active compound in the required amount in an appropriate solvent with one or a combination of ingredients enumerated above, as required, followed by filtered sterilization. Generally, dispersions are prepared by incorporating the active compound into a sterile vehicle, which contains a basic dispersion medium and the required other ingredients from those enumerated above. In the case of sterile powders for the preparation of sterile injectable solutions, the preferred methods of preparation are vacuum drying and freeze-drying, which yield a powder of the active ingredient plus any additional desired ingredient from a previously sterile-filtered solution thereof.

[0764] Therapeutic compounds that are or include nucleic acids can be administered by any method suitable for administration of nucleic acid agents, such as a DNA vaccine. These methods include gene guns, bio injectors, and skin patches as well as needle-free methods such as the micro-particle DNA vaccine technology disclosed in U.S. Pat. No. 6,194,389, and the mammalian transdermal needle-free vaccination with powder-form vaccine as disclosed in U.S. Pat. No. 6,168,587. Additionally, intranasal delivery is possible, as described in, *inter alia*, Hamajima et al., *Clin. Immunol. Immunopathol.*, 88(2), 205-10 (1998). Liposomes (e.g., as described in U.S. Pat. No. 6,472,375) and microencapsulation can also be used. Biodegradable targetable microparticle delivery systems can also be used (e.g., as described in U.S. Pat. No. 6,471,996).

[0765] Therapeutic compounds can be prepared with carriers that will protect the therapeutic compounds against rapid elimination from the body, such as a controlled release formulation, including implants and microencapsulated delivery systems. Biodegradable, biocompatible polymers can be used, such as collagen, ethylene vinyl acetate, poly-anhydrides (e.g., poly[1,3-bis(carboxyphenoxy)propane-co-sebacic-acid](PCPP-SA) matrix, fatty acid dimer-sebacic acid (FAD-SA) copolymer, poly(lactide-co-glycolide)), polyglycolic acid, collagen, polyorthoesters, polyethylene glycol-coated liposomes, hyaluronic acid and polylactic acid. Such formulations can be prepared using standard techniques, or obtained commercially, e.g., from Alza Corporation and Nova Pharmaceuticals, Inc. Liposomal suspensions (including liposomes targeted to selected cells with monoclonal antibodies to cellular antigens) can also be used as pharmaceutically acceptable carriers. These can be prepared according to methods known to those skilled in the art, for example, as described in U.S. Pat. No. 4,522,811. Semisolid, gelling, soft-gel, or other formulations (including controlled release) can be used, e.g., when administration to a surgical site is desired. Methods of making such formu-

lations are known in the art and can include the use of biodegradable, biocompatible polymers. See, e.g., Sawyer et al., *Yale J Biol Med.* 2006 December; 79(3-4): 141-152.

[0766] The pharmaceutical compositions described herein may be included in a container, kit, pack, or dispenser together with instructions for administration.

#### A. Systemic Administration

[0767] In some embodiments, a pharmaceutical composition comprising a CRISPR gene-editing system is administered systemically to a mammal in need thereof. In some embodiments, the composition is formulated for intravenous injection. In some embodiments, the composition is formulated for oral administration. In some embodiments, the composition is formulated for parenteral administration.

#### B. Local Administration

[0768] In some embodiments, a pharmaceutical composition comprising a CRISPR gene-editing system is administered locally to a mammal in need thereof. In some embodiments, the local administration is an intra-articular injection. In some embodiments, the composition is formulated for intradiscal injection. In some embodiments, the composition is formulated for epidural injection. In some embodiments, the composition is formulated for peridiscal injection. In some embodiments, the composition is formulated for pervertebral injection. In some embodiments, composition is formulated for administration to the facet joints of the spine. [0769] In some embodiments, a pharmaceutical composition comprising a CRISPR gene-editing system is administered locally to a mammal in need thereof during a surgical procedure. In some embodiments, a pharmaceutical composition comprising a CRISPR gene-editing system is administered locally to a mammal in need thereof 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 20, or 30 days after a surgical procedure.

#### EXAMPLES

[0770] The embodiments encompassed herein are now described with reference to the following examples. These examples are provided for the purpose of illustration only and the disclosure encompassed herein should in no way be construed as being limited to these examples, but rather should be construed to encompass any and all variations which become evident as a result of the teachings provided herein.

##### Example 1: Design of Crispr/Cas9 Guide RNAs Targeting cIL1A and cIL1B

[0771] Publicly accessible genomes (human, hg38; dog, CanFam3.1), collapsed gene models (merged Ensembl/Havana), tissue-specific exon expression ([gtexportal.org](http://gtexportal.org)) and various gRNA models were then used to select two to five individual crRNA sequences per gene, targeting canine and human interleukin-1 alpha (IL1A) and interleukin-1 beta (IL1B). The following gRNA design rules were applied:

- [0772] 1. The gRNA target region was limited to the first 5-50% of the coding sequence (CDS).
- [0773] 2. Single gRNAs were ranked according to maximal on-target editing using Azimuth 2.0 model (10.1038/nbt.3437) and minimal off-target editing using Cutting Frequency Determination (CFD) (10.1038/nbt.3437) and the specificity score from Hsu et al. (10.1038/nbt.2647).

[0774] 3. Highly ranked sgRNA with high frameshift frequencies (>75%) and uniform DNA repair outcomes (>0.48) as predicted by inDelphi (10.1038/s41586-018-0686-x) were selected for in vitro synthesis.

[0775] Using these selection criteria, crRNA guide sequences targeting different exons of the respective target genes were selected for further investigation. Specifically, as shown in FIG. 83A, sg235 (SEQ ID NO:2719) and sg236 (SEQ ID NO:2720) targeting exons 3 and 4 of the hIL1A gene were selected. Likewise, as shown in FIG. 83B, sg237 (SEQ ID NO:2721), sg238 (SEQ ID NO: 2722), sg248 (SEQ ID NO: 2723), sg249 (SEQ ID NO: 2724), and sg250 (SEQ ID NO: 2725) targeting exons 3, 4, and 5 of the hIL1B gene were selected. As shown in FIG. 83C, sg239 (SEQ ID NO: 2726), sg240 (SEQ ID NO: 2727), sg251 (SEQ ID NO:2728), and sg252 (SEQ ID NO:2729) targeting exons 3, 4, and 5 of the cIL1A gene were selected. Likewise, as shown in FIG. 83D, sg241 (SEQ ID NO: 2730) and sg242 (SEQ ID NO: 2731) targeting exons 3 and 4 of the cIL1B gene were selected.

[0776] sgRNAs, fusing the selected crRNA guide sequences to a scaffold sequence were then synthesised (Synthego) with scaffold modifications designed to increase their stability and decrease their cellular immunogenicity. Primers for genotyping were designed to be at least 200 bp from the target site and generate PCR amplicons <1.5 kb and synthesized (Merck).

[0777] The following quantities were used for single electroporation-based transfection using the 4D-nucleofector (Lonza, Catalog AAF-1002B and AAF-1002X) and nucleocuvette strips. 80 pmol synthesised sgRNA were pre-complexed with 4 µg Cas9 nuclease at room temperature for at least 10 min. 300-400K dissociated cells were washed with PBS before resuspending them in 20 µl supplemented P3 nucleofection solution and adding the Cas9 RNP complex. These cells were then transferred into a nucleocuvette well and electroporated using the pulse code ER-100. Directly after electroporation, the nucleocuvette was placed into the 37° C./5% CO<sub>2</sub> incubator for 10 min for the cells to recover from the electrical voltage. Afterwards, 80 µl growth medium was added to the nucleocuvette well and cells transferred into 6-well dishes with prewarmed growth medium.

[0778] Between two- and eleven-days post-electroporation, genomic DNA was extracted from 50-200K cells using DNeasy Blood & Tissue kit (Qiagen, Catalog 69506). Single gRNA target (and off-target) regions were amplified by PCR.

[0779] PCR products were size-verified by gel electrophoresis, purified using QIAquick PCR purification kit (Qiagen, Catalog 28106) and submitted for Sanger sequencing at Source BioScience. Sanger traces (ab1) were deconvoluted using ICE version 1.2 (found online at the URL [github.com/synthego-open/ice](https://github.com/synthego-open/ice)) to infer CRISPR edits. In addition, machine-learning predictions of gene editing using the selected probes was generated using inDelphi. In addition, the predicted off-target sites were analysed through direct sequencing to verify whether gRNA facilitates off-target editing.

[0780] Results of the empirical experiments and machine-learning prediction of gene editing using the selected guide sequences are shown in FIG. 83 and support the overall design method for various gene targets in any well-annotated genome.

#### Example 2: Functional Impact of IL1A/IL1B Editing

[0781] The sgRNAs with the highest knockout (KO) scores from Example 8 (i.e., the highest frameshift frequency) were used to generate double IL-1 $\alpha$ /IL-1 $\beta$  knock out (KO) cells. Specifically, human chondrocytes were edited to achieve >99% IL-1 $\alpha$  KO using crRNA sequence CAGAGACAGAUGAUCAAUGG and 67% IL-1 $\beta$  KO using crRNA sequence GUGCAGUUCAGUGAUCGUAC. Canine chondrocytes were edited to achieve 97% IL-1 $\alpha$  KO using crRNA sequence GACAUCCCAGCUUACCUUCA and 99% IL-1 $\beta$  KO using crRNA sequence ACUCUU-GUUACAGAGCUGGU.

[0782] Canine chondrocytes (Catalog Cn402K-05), human chondrocytes (Catalog 402-05a) and human fibroblast-like synovial cells (Catalog 408-05a) were purchased as frozen stocks ( $5 \times 10^5$  cells) from Cell Applications, Inc., San Diego, CA. Chondrocytes were cultured in growth medium consisting of DMEM/Ham's F12 (Gibco, Catalog 21331-020) supplemented with 20% (v/v) untreated FBS (Gibco, Catalog 10270-106) and 1 $\times$ GlutaMAX (Gibco, Catalog 35050-038). Synovial cells were cultured in growth medium consisting of DMEM (Gibco, Catalog 11960-044), 10% non-treated FBS (Gibco, Catalog 10270-106) and 1 $\times$ GlutaMAX (Gibco, Catalog 35050-038). Cells were confirmed as being negative for *Mycoplasma* spp. and subjected to STR profiling prior to use. For electroporation and subculture, cells were dissociated using 0.25% trypsin (Gibco, Catalog 25200056). Trypsin was quenched with 9 volumes of growth medium and cells were spun at 1,000 g to remove the supernatant.

[0783] Interleukin-1 (IL-1) release was induced by challenging sub-confluent monolayers of cells (edited or wild-type non-edited) with lipopolysaccharide (LPS). In brief, non-edited (control) and double IL-1 $\alpha$ /IL-1 $\beta$  KO (edited) human or canine chondrocytes were seeded at density of approximately  $5 \times 10^4$  cells per well in 24-well plates. After 24-48 hours, the medium was replaced with fresh, serum-free medium containing either LPS (50 µg/ml) or PBS vehicle and the plates returned to the incubator. Plates were harvested after 6 and 24 hours for the determination of IL-1 release. Media were snap-frozen in liquid nitrogen and stored at -20° C. until they were assayed.

[0784] The concentration of IL-1A and IL-1B in the culture medium was measured with species-specific commercial assays, following the manufacturer's instructions. Prior to measurement, frozen media were thawed and then centrifuged (1,500 g for 2 mins) in order to remove cellular debris. Aliquots of medium were measured in duplicate and the concentration of IL-1 determined from a standard curve of recombinant human or canine IL-1A or IL-1B, as appropriate. The results of IL-1 alpha release in canine cells are shown in FIGS. 84A (6 hours) and 84B (24 hours). The results of IL-1 beta release in canine cells are shown in FIGS. 84C (6 hours) and 84D (24 hours). The results of IL-1 alpha release in human cells are shown in FIGS. 85A (6 hours) and 85B (24 hours). The results of IL-1 beta release in human cells are shown in FIGS. 85C (6 hours) and 85D (24 hours).

[0785] Taken together, these results demonstrated the functional impact on editing either human or canine cells.

**Example 3: Impact of Cas9 Mutant with Enhanced Specificity**

**[0786]** The analysis of gene editing specificity reported in Example 8 was repeated using an enhanced Specificity CRISPR associated protein 9. The eSpCas9 includes three specificity enhancing mutations: K848A, K1003A, and R1060A, as described in Slaymaker et al., *Science*, 351:84-88 (2016). The eSpCas9 was expressed in *E. coli* and purified to homogeneity. The construct has a molecular weight of 161 kDa and contains N-terminal Flag-tags and a C-terminal hexa-His-tag. The sequence of the eSpCas9 is:

```
(SEQ ID NO: 2733)
MDYKDHDGDYKDHDIDYKDDDKMAPKKRKVGIHGVPAADKKYSIGLD
IGTNSVGWAVITDEYKVPSKKPKVLGNLTDRHISKKNLIGALLFDGETA
EATRLKRTARRYTRRKNRICYLQEIFSNEAKVDDSSFHRLEESPLVE
EDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLA
LAHMIKFRGHFLIEGDLNPDNSVDKLFIQLVQTYNQLFEENPINASGV
DAKAILSARLSKSRRLENLIAQLPGEKKNGLFGNLIALSLGLTPNFKSN
FDLAEDAKLQLSKDTYDDDLDNLLAQIGDQYADLFLAAKNLSDAILLSD
ILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKYKEIFFD
QSKNGYAGYIDGGASQEEFYKFIFKPILEKMDGTEELLVQLNREDLLRKQ
RTFDNGSIPHQIHLGELHAILRQQEDFYPFLKDNRREKIEKILTFRIPYY
VGPLARGNSRFAMTRKSEETITPWNFEEVVDKGASAQSFIERMNTNFDK
NLPNEVKLPKHSSLLEYEYFTVYNELTKVKYVTEGMRKPAAFLSGEQKKAI
DLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYHDLL
KIIKDKDFLDNEENEDEDIVLTTLTFEDREMIEERLKTYAHLFDDKV
MKQLKRRRTGWRGLSRKLINGIRDQSGKTILDPLKSDGFANRNPQMQL
IHDDSLTFKEDIQKAQVSGQGDSLHEHIANLAGSPAIIKGILQTVKVVD
ELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERMKRIEEGIKELGSQ
ILKEHPVENTQLQNEKLYLYLQNQGRDMYVQDQELDINRLSDYDVDHIVP
QSFLADDSDIDNKVLTRSKRNKGKSDNVPSSEEVVKMKNYWRQLLNALKI
TQRKFDNLTKAERGGLSELDKAGFIKRQLVETRQTKHVAQILDSRMNT
KYDENDKLIREVKVITLKS KLVSDFRKDFQFYKVREINNYHHAHDAYLN
AVVGTALIKKYPALSEFVYGDYKVYDVRKMIAKSEQEIGKATAKYFFY
SNIMNNFKTEITLANGEIRKAPLIETNGETGEIVWDKGRDFATVRKVLS
MPQVNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKYGGFDSP
TVAYSVLVVAKEVKGSKKLKSVKELLGITIMERSSFEKNPIDFLEAKG
YKEVKKDLIIKLPKYSLFELENGRKMLASAGELQKGNELALPSKYVNF
LYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIIEQISEFSKRVILAD
ANLDKVLSAYNKHRDKPIREQAENI IHLFTLTLNGLAPAAFKYFDTTIDR
KRYTSTKEVLDATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKK
KKAALAEHHHHHH.
```

**[0787]** Briefly, the same sgRNAs used in Example 1, and shown in FIG. 83, were complexed with the eSpCas9. Single guide RNAs (sgRNAs), fusing the selected crRNA guide sequences to a scaffold sequence were then synthesised (Synthego) with scaffold modifications designed to increase their stability and decrease their cellular immunogenicity. Primers for genotyping were designed to be at least 200 bp from the target site and generate PCR amplicons <1.5 kb and synthesized (Merck).

**[0788]** The following quantities were used for single electroporation-based transfection using the 4D-nucleofector (Lonza, Catalog AAF-1002B and AAF-1002X) and nucleocuvette strips. 80 pmol synthesised sgRNA were pre-complexed with eSpCas9 nuclease at room temperature for at least 10 min. 300-400K dissociated cells were washed with PBS before resuspending them in 20 µl supplemented P3 nucleofection solution and adding the Cas9 RNP complex. These cells were then transferred into a nucleocuvette well and electroporated using the pulse code ER-100. Directly after electroporation, the nucleocuvette was placed into the 37° C./5% CO<sub>2</sub> incubator for 10 min for the cells to recover from the electrical voltage. Afterwards, 80 µl growth medium was added to the nucleocuvette well and cells transferred into 6-well dishes with prewarmed growth medium.

**[0789]** Between two- and eleven-days post-electroporation, genomic DNA was extracted from 50-200K cells using DNeasy Blood & Tissue kit (Qiagen, Catalog 69506). Single gRNA target (and off-target) regions were amplified by PCR.

**[0790]** PCR products were size-verified by gel electrophoresis, purified using QIAquick PCR purification kit (Qiagen, Catalog 28106) and submitted for Sanger sequencing at Source BioScience. Sanger traces (ab1) were deconvoluted using ICE version 1.2 (found online at the URL [github.com/synthego-open/ice](http://github.com/synthego-open/ice)) to infer CRISPR edits. In addition, machine-learning predictions of gene editing using the selected probes was generated using inDelphi. In addition, the predicted off-target sites were analysed through direct sequencing to verify whether gRNA facilitates off-target editing.

**[0791]** As compared to the Cas9 editing reported in Example 1, use of eSpCas9 reduces off-target editing without losing on-target activity. For example, the off-target editing by sgRNA #242 (targeting cIL1B) of three loci, having 2, 3, and 3 mismatches, respectively, were evaluated by amplifying and then sequencing the loci reported in Table 3. As shown in Table 3, the first off-target loci experienced no editing in the experiment described in Example 8, and was not tested here. The second off-target loci experienced almost complete off-target editing (98-99%) in the experiment described in Example 8, but experienced no editing when eSpCas9 was used. The third off-target loci experienced some editing (0-25%) in the experiment described in Example 8, but again experienced no editing when eSpCas9 was used. Further, as shown in Table 2, the “enhanced on-target score,” corresponding to editing using eSpCas9 as described in this example, for each sgRNA tested was as high, if not higher, than the “on-target score,” corresponding to the editing described in Example 1.

TABLE 2

#	Target Exon #	Strand	On-target metrics		Precision score**	Frameshift %***	Enhanced on-target score****	Off-target metrics	
			On-target score*	Precision score**				Off-target score^	
sg235	3	-	69.5	0.57	93.7	72.4	67		
sg236	4	-	53.9	0.65	93.9	66.7	76.9		
Human IL1B (IL1B-201; GRCh38)									
sg237	3	+	60.9	0.57	80	64.7	66.7		
sg238	4	-	58.3	0.6	86.3	65.7	49.3		
sg248	5	+	61.7	0.65	89.9	69.6	86.5		
sg249	5	+	68.2	0.52	75.3	65.9	93.8		
sg250	5	-	64.4	0.48	83.9	65.2	91		
Canine IL1A (IL1A-201; CanFam3.1)									
sg239	3	+	49.5	0.55	78.3	58.1	57.7		
sg240	4	+	43.8	0.61	87.2	59.0	41.8		
sg251	5	+	72.1	0.49	77.6	67.7	85.1		
sg252	5	-	68.4	0.48	90.4	68.8	66.6		

TABLE 3

Sequence	PAM	Score	Gene	Chrom- osome	Strand	Posi- tion	Mis- matches	CRISPR Edits with Cas9	CRISPR Edits with enhanced specificity Cas9
ACTCTTGTACAG AGCTGGT	GGG	100	ENSCAFG00 000007249	chr17	1	37022194	0		
ACTTTTGTTCAG AGCTGGT	CAG	6.16161972		chr33	-1	20234937	2	0%	
CCTCATGCTACAG AGCTGGT	GGG	2.76564774		chr1	-1	47541563	3	98-99%	0%
GTGCTTGTACAG AGCTGGT	GGG	2.32143742		chr26	-1	32323843	3	0-25%	0%

#### Example 4: Selection of gRNAs Targeting IL1A and IL1B in Humans and Canines

**[0792]** Given that the ultimate goal of CRISPR target design is fabrication of nucleotide sequences that will hybridize to genomic DNA sequences resulting in the most robust knockout of a targeted gene as part of the CRISPR/Cas system, the process begins with assessment of splicing at the target loci. Human IL1A exhibits almost exclusively canonical splicing (i.e., no major variants) across various tissue types with the mature mRNA including exons 2-7, making each of these a potential CRISPR gRNA target (FIG. 86). Additional functional analysis (see Michlits, et al. (2020). *Nature Methods*, 17(7), 708-716) of the hIL-1a gene demonstrated that all functional domains cluster within Exons 5-7. In order to avoid a truncation that retains residual post-editing functionality, hIL-1a CRISPR targets were limited to those upstream of the functional domain cluster (exons 2-4). In so doing, a resultant frameshift or premature stop codon (i.e., missense mutation) at the editing site will impact all functional domains.

**[0793]** Similar analysis of hIL1B found a stronger overall expression pattern and more variation in splicing as compared to hIL1A (FIG. 87). However, as no tissue exhibited a variant omitting exons 2-7, each of these remained viable CRISPR targets. Application of the same functional analysis

tools for hIL-1b found that functional domains cluster in exons 5-7, leaving exons 2-4 as viable CRISPR targets.

**[0794]** Having established the human gene targets, emphasis then shifted to gRNA targeting domain design. Generated CRISPR targeting domains were first tested in silico through at least, four separate algorithms, yielding scores assessing cutting activity (On-Target score; see Doench et al. (2016). *Nature Biotechnology*, 34(2), 184-191), reproducibility of the particular mutation via double-strand break repair mechanisms (Precision score; inDelphi), likelihood of creating a frameshift mutation (Frameshift score; inDelphi), and specificity of gRNA binding (Off-Target score; CRISPOR) (FIG. 88). Cutoffs were set for On-Target score at >0.30 and for Off-Target scores of 0 for 0 or 1 mismatch (first two columns).

**[0795]** The same design process was then repeated for the orthologous canine gene targets. Splicing and functional analyses of canine IL-1a (cIL-1a) shows 6 exons (exons 2-7) incorporated in the mature mRNA, of which exons 6 and 7 contain functional domains (FIG. 89A). This leaves exons 2-5 as viable CRISPR targets. The mature mRNA of cIL1B contains exons 2-7, with the core function domains clustered in exons 5-7 (FIG. 89B). As such, exons 2-4 are potentially viable CRISPR targets. Generated candidate CRISPR targeting domains were then analyzed in silico; those exceeding the minimum cutoff scores are shown in FIG. 90.

**Example 5: Characterizing the Ablation of IL1A and IL1B in Primary Human and Canine Cells**

[0796] Algorithm-validated gRNA targeting domains were then tested in primary cells. Briefly, plasmid DNA encoding a sgRNA with the selected targeting domain was introduced into the primary cells with an encoded Cas9 plasmid via electroporation. Pooled cell populations then underwent DNA extraction and sequencing to assess editing efficiency.

[0797] The results, shown in FIG. 91, demonstrate a wide range of editing efficiencies in both human (FIG. 91A) and canine (FIGS. 91B-91C) cells. Indeed, for each gene target, at least one targeting domain demonstrates effective editing (between 89% and 99%) with reproducibility between different cell types (in canine).

[0798] However, in addition to this robust editing of the gene target, sgRNA 242, which targets cIL1B, also exhibited high levels of off-target editing, as anticipated by the in silico analysis (FIG. 92). To rectify these off-target effects, the experiments were repeated with an engineered, enhanced-specificity Cas9 (eSpCas9). This engineered Cas9 completely abrogated the previously-observed off-target effects while still maintain maximal editing efficiency at the target site (FIG. 93). These data demonstrate that the lead candidate sgRNA targeting domain for each gene target can safely (i.e., without off-target effects) and reliably generate genetic knockouts through creation of a primary missense mutation within the targeted locus in human (FIG. 94A) and canine (FIG. 94B) chondrocytes.

[0799] Taken together, these data presented a strong profile for ablating gene expression of IL1A and IL1B in primary mammalian cells.

**Example 6: Coadministration of IL1A and IL1B-Targeted sgRNAs in Primary Mammalian Cells**

[0800] Having observed robust and reproducible efficacy for each individual gRNA targeting domain, the lead candidates were next assessed in the context of co-administration in order to generate double knockouts. To do this, each sgRNA was administered to canine synoviocytes as described above either simultaneously or sequentially (in either order). The results show that sgRNA 242 is highly effective at knocking out cIL1B under all conditions (FIG. 95). Conversely, sgRNA 240, targeting cIL1A, demonstrated optimal efficacy when it edits first with both simultaneous and secondary administration reducing efficacy by roughly 30%. However, such significant reduction likely remains beyond the threshold needed to impact functionality at the organismal level. As such, these results show that more than one sgRNA may be used within a single cell, preferably to silence multiple genes within a single pathway, thereby maximizing functional efficacy.

**Example 7: Applicability to Additional Species**

[0801] Interleukin 1 is a highly conserved gene in terms of both sequence and function among mammals (see Dinarello, C. A. (1991). *Blood* 77 (8): 1627-1652). As a result, the gRNA targeting domains that have been generated and characterized for specific species may result in efficient editing of the IL-1 locus of additional species. An alignment to discover conserved IL1A (FIG. 96A) and IL1B (FIG. 96B) gene target positions among humans, horses, dogs, and

mice finds relatively few mismatched base pairs across all species at particular target positions. For example, sgRNA 239, targeting cIL1A, is predicted to also edit mouse IL1A. Given the reported flexibility within the CRISPR/Cas system to tolerate imperfect sequence alignments under certain conditions (see generally Zischewski, J., et al. (2017). *Biotechnology Advances*, 35(1), 95-104), cross-species reactivity for particular targeting domains is wholly anticipated for any number of conserved genes, including, but not limited to, any of those genes listed in Table 4.

TABLE 4

List of Potential pro-inflammatory gene targets.

6-phosphogluconate dehydrogenase (6PGD)
Alcohol dehydrogenase (ADH)
Aldehyde dehydrogenase (ALDH2)
AP-1
B-cell lymphoma-extra large (Bcl-XL)
BCL2 apoptosis regulator (Bcl-2)
Bcl-2-associated X protein (BAX)
Catalase (CAT)
c-Jun N-terminal kinase (JNK)
Coenzyme Q10
CYP2E1
Cytochrome c (Cyt c)
F1Fo-ATP synthase
Ferritin heavy chain (FHC)
Glucose-6-phosphate dehydrogenase (G6PD)
Glutamylcysteine synthetase (GCS)
Glutathione (GSH) synthase
Glutathione Peroxidase 1 (GPX1)
Glutathione Peroxidase 2 (GPX2)
Glutathione Peroxidase 3 (GPX3)
Glutathione Peroxidase 4 (GPX4)
Glutathione Peroxidase 5 (GPX5)
Glutathione Peroxidase 6 (GPX6)
Glutathione Peroxidase 7 (GPX7)
Glutathione Peroxidase 8 (GPX8)
Glutathione reductase (GR)
Glycerol 3-phosphate dehydrogenase
Growth arrest and DNA damage (GADD 45)
Hypoxia-inducible factor 1-alpha (HIF-1 $\alpha$ )
Mitogen-activated protein kinase (MAPK)
NADH-ubiquinone oxidoreductase
NADPH oxidase 4 (NOX4)
NADPH oxidase 5 (NOX5)
Nuclear factor kappa-light-chain-enhancer of activated B cells (NF- $\kappa$ B)
Nuclear factor $\kappa$ B (NF- $\kappa$ B) essential modulator (NEMO)
p46Shc (SHC isoform)
p52Shc (SHC isoform)
p53 upregulated modulator of apoptosis (PUMA)
p66Shc (SHC isoform)
Phosphoinositide 3-kinase (PI3-K)
Proline oxidase (PIG6, POX)
Quinone oxidoreductase (PIG3, NQO1)
Sestrin 1 (SESN1)
Sestrin 2 (SESN2)
SHC adaptor protein 1 (SHC1)
Superoxide dismutase 1 (SOD1)
Superoxide dismutase 2 (SOD2)
Superoxide dismutase 3 (SOD3)
TNF alpha induced protein 3 (TNFAIP3)
Tumor protein 53 (p53)
Tumor protein p53 inducible nuclear protein 1 (TP53INP1)
Ubiquinol-cytochrome c oxidoreductase
2B4
ABCA1
ACPs
ADAR-1
ADSS
AIG1
AIM2
APOBEC3
ARRB2

TABLE 4-continued

List of Potential pro-inflammatory gene targets.

B2M
BCAS3
BMP4
C10orf32
C21orf33
CASP1
CCL5
CD160
Cd53
CDKN2A
CHEK1
CNNM2
CNTNAP2
CSMD1
CTLA-4
CTSB
C-type lectin receptors CLRs
CXCL10
CYP17A1
DDX60
DYNC1I1
FOXO3a
GPC6
GRN
HCK
HECW1
HLA
IFI30
IFI44L
IFI6
IFITM
IFITM1/3
IFITM2
IFITM3
IL-18
IL-1 $\alpha$
IL-1 $\beta$
interferon- $\gamma$
interleukin-12 (IL-12)
IRF
IRF-1
IRF3
IRF7
LAG-3
LIPC
MDA5/IFIH1
MPAK

TABLE 4-continued

List of Potential pro-inflammatory gene targets.

MYH9
MYO16
MYO5A
NAIP
NF- $\kappa$ B
NLRP4
NLRP3
NOD2
nucleotide oligomerization and binding domain NOD-like receptors
OAS1
OAS2
OASL
parkin gene (PARK2)
PD-1
PLEKHG1
PRKCA
PTBP1
PYCARD
Pyrin-HIN (PYHIN) domain containing receptors (e.g. AIM2)
Any retinoic acid inducible gene-I (RIG-I)-like receptors (RLRs)___
RFC3
RGS1
RIG-I/DDX58
SAMHD1
SF3A1/SF3B1
SFXN2
SLAMF7
SLC41A1
SLC8A1
SLCO3A1
STAT1
Tetherin
TLR5
TLR7
TLR9
Any Toll-like receptors (TLRs)
TREM2
TREX1
TRIM5
TTLL7
TYROBP

## Example 8: LNP Systems

[0802] In some embodiments, the following LNP systems are used to formulate pharmaceutical compositions described herein:

TABLE 5

LNP Systems (all values are a molar ratio relative to a 100 total)

LNP System#	Ionizable Lipid Component (molar ratio or molar ratio range)	Neutral/Helper Lipid Component (molar ratio or molar ratio range)	Steroid System Component (molar ratio or molar ratio range)	PEGylated Lipid Component (molar ratio or molar ratio range)
LNP001	Dlin-KC2-DMA (42-50)	DSPC (8-10)	(36-46)	(balance to 100)
LNP002	Dlin-KC2-DMA (42-50)	DSPC (10-12)	(36-46)	(balance to 100)
LNP003	Dlin-KC2-DMA (42-50)	DSPC (10 $\pm$ 0.5)	(36-46)	(balance to 100)
LNP004	Dlin-KC2-DMA (42-50)	DOPE (8-10)	(36-46)	(balance to 100)
LNP005	Dlin-KC2-DMA (42-50)	DOPE (10-12)	(36-46)	(balance to 100)
LNP006	Dlin-KC2-DMA (42-50)	DOPE (10 $\pm$ 0.5)	(36-46)	(balance to 100)
LNP007	Dlin-KC2-DMA (42-50)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP008	Dlin-KC2-DMA (42-50)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP009	Dlin-KC2-DMA (42-50)	DOTMA (10 $\pm$ 0.5)	(36-46)	(balance to 100)
LNP010	Dlin-KC2-DMA (42-50)	DPPC (8-10)	(36-46)	(balance to 100)
LNP011	Dlin-KC2-DMA (42-50)	DPPC (10-12)	(36-46)	(balance to 100)
LNP012	Dlin-KC2-DMA (42-50)	DPPC (10 $\pm$ 0.5)	(36-46)	(balance to 100)
LNP013	Dlin-KC2-DMA (40-48)	DSPC (8-10)	(36-46)	(balance to 100)
LNP014	Dlin-KC2-DMA (40-48)	DSPC (10-12)	(36-46)	(balance to 100)
LNP015	Dlin-KC2-DMA (40-48)	DSPC (10 $\pm$ 0.5)	(36-46)	(balance to 100)

TABLE 5-continued

LNP Systems (all values are a molar ratio relative to a 100 total)				
LNP System#	Ionizable Lipid Component (molar ratio or molar ratio range)	Neutral/Helper Lipid Component (molar ratio or molar ratio range)	Steroid System Component (molar ratio or molar ratio range)	PEGylated Lipid Component (molar ratio or molar ratio range)
LNP016	Dlin-KC2-DMA (40-48)	DOPE (8-10)	(36-46)	(balance to 100)
LNP017	Dlin-KC2-DMA (40-48)	DOPE (10-12)	(36-46)	(balance to 100)
LNP018	Dlin-KC2-DMA (40-48)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP019	Dlin-KC2-DMA (40-48)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP020	Dlin-KC2-DMA (40-48)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP021	Dlin-KC2-DMA (40-48)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP022	Dlin-KC2-DMA (40-48)	DPPC (8-10)	(36-46)	(balance to 100)
LNP023	Dlin-KC2-DMA (40-48)	DPPC (10-12)	(36-46)	(balance to 100)
LNP024	Dlin-KC2-DMA (40-48)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP025	Dlin-KC2-DMA (43-45)	DSPC (8-10)	(36-46)	(balance to 100)
LNP026	Dlin-KC2-DMA (43-45)	DSPC (10-12)	(36-46)	(balance to 100)
LNP027	Dlin-KC2-DMA (43-45)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP028	Dlin-KC2-DMA (43-45)	DOPE (8-10)	(36-46)	(balance to 100)
LNP029	Dlin-KC2-DMA (43-45)	DOPE (10-12)	(36-46)	(balance to 100)
LNP030	Dlin-KC2-DMA (43-45)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP031	Dlin-KC2-DMA (43-45)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP032	Dlin-KC2-DMA (43-45)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP033	Dlin-KC2-DMA (43-45)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP034	Dlin-KC2-DMA (43-45)	DPPC (8-10)	(36-46)	(balance to 100)
LNP035	Dlin-KC2-DMA (43-45)	DPPC (10-12)	(36-46)	(balance to 100)
LNP036	Dlin-KC2-DMA (43-45)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP037	Dlin-KC2-DMA (44 ± 0.5)	DSPC (8-10)	(36-46)	(balance to 100)
LNP038	Dlin-KC2-DMA (44 ± 0.5)	DSPC (10-12)	(36-46)	(balance to 100)
LNP039	Dlin-KC2-DMA (44 ± 0.5)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP040	Dlin-KC2-DMA (44 ± 0.5)	DOPE (8-10)	(36-46)	(balance to 100)
LNP041	Dlin-KC2-DMA (44 ± 0.5)	DOPE (10-12)	(36-46)	(balance to 100)
LNP042	Dlin-KC2-DMA (44 ± 0.5)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP043	Dlin-KC2-DMA (44 ± 0.5)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP044	Dlin-KC2-DMA (44 ± 0.5)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP045	Dlin-KC2-DMA (44 ± 0.5)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP046	Dlin-KC2-DMA (44 ± 0.5)	DPPC (8-10)	(36-46)	(balance to 100)
LNP047	Dlin-KC2-DMA (44 ± 0.5)	DPPC (10-12)	(36-46)	(balance to 100)
LNP048	Dlin-KC2-DMA (44 ± 0.5)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP049	Dlin-MC3-DMA (44-50)	DSPC (8-10)	(36-46)	(balance to 100)
LNP050	Dlin-MC3-DMA (44-50)	DSPC (10-12)	(36-46)	(balance to 100)
LNP051	Dlin-MC3-DMA (44-50)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP052	Dlin-MC3-DMA (44-50)	DOPE (8-10)	(36-46)	(balance to 100)
LNP053	Dlin-MC3-DMA (44-50)	DOPE (10-12)	(36-46)	(balance to 100)
LNP054	Dlin-MC3-DMA (44-50)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP055	Dlin-MC3-DMA (44-50)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP056	Dlin-MC3-DMA (44-50)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP057	Dlin-MC3-DMA (44-50)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP058	Dlin-MC3-DMA (44-50)	DPPC (8-10)	(36-46)	(balance to 100)
LNP059	Dlin-MC3-DMA (44-50)	DPPC (10-12)	(36-46)	(balance to 100)
LNP060	Dlin-MC3-DMA (44-50)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP061	Dlin-MC3-DMA (50-54)	DSPC (8-10)	(36-46)	(balance to 100)
LNP062	Dlin-MC3-DMA (50-54)	DSPC (10-12)	(36-46)	(balance to 100)
LNP063	Dlin-MC3-DMA (50-54)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP064	Dlin-MC3-DMA (50-54)	DOPE (8-10)	(36-46)	(balance to 100)
LNP065	Dlin-MC3-DMA (50-54)	DOPE (10-12)	(36-46)	(balance to 100)
LNP066	Dlin-MC3-DMA (50-54)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP067	Dlin-MC3-DMA (50-54)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP068	Dlin-MC3-DMA (50-54)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP069	Dlin-MC3-DMA (50-54)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP070	Dlin-MC3-DMA (50-54)	DPPC (8-10)	(36-46)	(balance to 100)
LNP071	Dlin-MC3-DMA (50-54)	DPPC (10-12)	(36-46)	(balance to 100)
LNP072	Dlin-MC3-DMA (50-54)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)

TABLE 5-continued

LNP Systems (all values are a molar ratio relative to a 100 total)				
LNP System#	Ionizable Lipid Component (molar ratio or molar ratio range)	Neutral/Helper Lipid Component (molar ratio or molar ratio range)	Steroid System Component (molar ratio or molar ratio range)	PEGylated Lipid Component (molar ratio or molar ratio range)
LNP073	Dlin-MC3-DMA (49-51)	DSPC (8-10)	(36-46)	(balance to 100)
LNP074	Dlin-MC3-DMA (49-51)	DSPC (10-12)	(36-46)	(balance to 100)
LNP075	Dlin-MC3-DMA (49-51)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP076	Dlin-MC3-DMA (49-51)	DOPE (8-10)	(36-46)	(balance to 100)
LNP077	Dlin-MC3-DMA (49-51)	DOPE (10-12)	(36-46)	(balance to 100)
LNP078	Dlin-MC3-DMA (49-51)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP079	Dlin-MC3-DMA (49-51)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP080	Dlin-MC3-DMA (49-51)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP081	Dlin-MC3-DMA (49-51)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP082	Dlin-MC3-DMA (49-51)	DPPC (8-10)	(36-46)	(balance to 100)
LNP083	Dlin-MC3-DMA (49-51)	DPPC (10-12)	(36-46)	(balance to 100)
LNP084	Dlin-MC3-DMA (49-51)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP085	Dlin-MC3-DMA (50 ± 0.5)	DSPC (8-10)	(36-46)	(balance to 100)
LNP086	Dlin-MC3-DMA (50 ± 0.5)	DSPC (10-12)	(36-46)	(balance to 100)
LNP087	Dlin-MC3-DMA (50 ± 0.5)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP088	Dlin-MC3-DMA (50 ± 0.5)	DOPE (8-10)	(36-46)	(balance to 100)
LNP089	Dlin-MC3-DMA (50 ± 0.5)	DOPE (10-12)	(36-46)	(balance to 100)
LNP090	Dlin-MC3-DMA (50 ± 0.5)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP091	Dlin-MC3-DMA (50 ± 0.5)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP092	Dlin-MC3-DMA (50 ± 0.5)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP093	Dlin-MC3-DMA (50 ± 0.5)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP094	Dlin-MC3-DMA (50 ± 0.5)	DPPC (8-10)	(36-46)	(balance to 100)
LNP095	Dlin-MC3-DMA (50 ± 0.5)	DPPC (10-12)	(36-46)	(balance to 100)
LNP096	Dlin-MC3-DMA (50 ± 0.5)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP097	SM-102 (44-50)	DSPC (8-10)	(36-46)	(balance to 100)
LNP098	SM-102 (44-50)	DSPC (10-12)	(36-46)	(balance to 100)
LNP099	SM-102 (44-50)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP100	SM-102 (44-50)	DOPE (8-10)	(36-46)	(balance to 100)
LNP101	SM-102 (44-50)	DOPE (10-12)	(36-46)	(balance to 100)
LNP102	SM-102 (44-50)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP103	SM-102 (44-50)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP104	SM-102 (44-50)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP105	SM-102 (44-50)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP106	SM-102 (44-50)	DPPC (8-10)	(36-46)	(balance to 100)
LNP107	SM-102 (44-50)	DPPC (10-12)	(36-46)	(balance to 100)
LNP108	SM-102 (44-50)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP109	SM-102 (50-54)	DSPC (8-10)	(36-46)	(balance to 100)
LNP110	SM-102 (50-54)	DSPC (10-12)	(36-46)	(balance to 100)
LNP111	SM-102 (50-54)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP112	SM-102 (50-54)	DOPE (8-10)	(36-46)	(balance to 100)
LNP113	SM-102 (50-54)	DOPE (10-12)	(36-46)	(balance to 100)
LNP114	SM-102 (50-54)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP115	SM-102 (50-54)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP116	SM-102 (50-54)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP117	SM-102 (50-54)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP118	SM-102 (50-54)	DPPC (8-10)	(36-46)	(balance to 100)
LNP119	SM-102 (50-54)	DPPC (10-12)	(36-46)	(balance to 100)
LNP120	SM-102 (50-54)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP121	SM-102 (49-51)	DSPC (8-10)	(36-46)	(balance to 100)
LNP122	SM-102 (49-51)	DSPC (10-12)	(36-46)	(balance to 100)
LNP123	SM-102 (49-51)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP124	SM-102 (49-51)	DOPE (8-10)	(36-46)	(balance to 100)
LNP125	SM-102 (49-51)	DOPE (10-12)	(36-46)	(balance to 100)
LNP126	SM-102 (49-51)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP127	SM-102 (49-51)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP128	SM-102 (49-51)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP129	SM-102 (49-51)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)

TABLE 5-continued

LNP Systems (all values are a molar ratio relative to a 100 total)				
LNP System#	Ionizable Lipid Component (molar ratio or molar ratio range)	Neutral/Helper Lipid Component (molar ratio or molar ratio range)	Steroid System Component (molar ratio or molar ratio range)	PEGylated Lipid Component (molar ratio or molar ratio range)
LNP130	SM-102 (49-51)	DPPC (8-10)	(36-46)	(balance to 100)
LNP131	SM-102 (49-51)	DPPC (10-12)	(36-46)	(balance to 100)
LNP132	SM-102 (49-51)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP133	SM-102 (50 ± 0.5)	DSPC (8-10)	(36-46)	(balance to 100)
LNP134	SM-102 (50 ± 0.5)	DSPC (10-12)	(36-46)	(balance to 100)
LNP135	SM-102 (50 ± 0.5)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP136	SM-102 (50 ± 0.5)	DOPE (8-10)	(36-46)	(balance to 100)
LNP137	SM-102 (50 ± 0.5)	DOPE (10-12)	(36-46)	(balance to 100)
LNP138	SM-102 (50 ± 0.5)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP139	SM-102 (50 ± 0.5)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP140	SM-102 (50 ± 0.5)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP141	SM-102 (50 ± 0.5)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP142	SM-102 (50 ± 0.5)	DPPC (8-10)	(36-46)	(balance to 100)
LNP143	SM-102 (50 ± 0.5)	DPPC (10-12)	(36-46)	(balance to 100)
LNP144	SM-102 (50 ± 0.5)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP145	ALC-0315 (44-50)	DSPC (8-10)	(36-46)	(balance to 100)
LNP146	ALC-0315 (44-50)	DSPC (10-12)	(36-46)	(balance to 100)
LNP147	ALC-0315 (44-50)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP148	ALC-0315 (44-50)	DOPE (8-10)	(36-46)	(balance to 100)
LNP149	ALC-0315 (44-50)	DOPE (10-12)	(36-46)	(balance to 100)
LNP150	ALC-0315 (44-50)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP151	ALC-0315 (44-50)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP152	ALC-0315 (44-50)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP153	ALC-0315 (44-50)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP154	ALC-0315 (44-50)	DPPC (8-10)	(36-46)	(balance to 100)
LNP155	ALC-0315 (44-50)	DPPC (10-12)	(36-46)	(balance to 100)
LNP156	ALC-0315 (44-50)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP157	ALC-0315 (50-54)	DSPC (8-10)	(36-46)	(balance to 100)
LNP158	ALC-0315 (50-54)	DSPC (10-12)	(36-46)	(balance to 100)
LNP159	ALC-0315 (50-54)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP160	ALC-0315 (50-54)	DOPE (8-10)	(36-46)	(balance to 100)
LNP161	ALC-0315 (50-54)	DOPE (10-12)	(36-46)	(balance to 100)
LNP162	ALC-0315 (50-54)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP163	ALC-0315 (50-54)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP164	ALC-0315 (50-54)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP165	ALC-0315 (50-54)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP166	ALC-0315 (50-54)	DPPC (8-10)	(36-46)	(balance to 100)
LNP167	ALC-0315 (50-54)	DPPC (10-12)	(36-46)	(balance to 100)
LNP168	ALC-0315 (50-54)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP169	ALC-0315 (49-51)	DSPC (8-10)	(36-46)	(balance to 100)
LNP170	ALC-0315 (49-51)	DSPC (10-12)	(36-46)	(balance to 100)
LNP171	ALC-0315 (49-51)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP172	ALC-0315 (49-51)	DOPE (8-10)	(36-46)	(balance to 100)
LNP173	ALC-0315 (49-51)	DOPE (10-12)	(36-46)	(balance to 100)
LNP174	ALC-0315 (49-51)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP175	ALC-0315 (49-51)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP176	ALC-0315 (49-51)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP177	ALC-0315 (49-51)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP178	ALC-0315 (49-51)	DPPC (8-10)	(36-46)	(balance to 100)
LNP179	ALC-0315 (49-51)	DPPC (10-12)	(36-46)	(balance to 100)
LNP180	ALC-0315 (49-51)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP181	ALC-0315 (50 ± 0.5)	DSPC (8-10)	(36-46)	(balance to 100)
LNP182	ALC-0315 (50 ± 0.5)	DSPC (10-12)	(36-46)	(balance to 100)
LNP183	ALC-0315 (50 ± 0.5)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP184	ALC-0315 (50 ± 0.5)	DOPE (8-10)	(36-46)	(balance to 100)
LNP185	ALC-0315 (50 ± 0.5)	DOPE (10-12)	(36-46)	(balance to 100)
LNP186	ALC-0315 (50 ± 0.5)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP187	ALC-0315 (50 ± 0.5)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP188	ALC-0315 (50 ± 0.5)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP189	ALC-0315 (50 ± 0.5)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP190	ALC-0315 (50 ± 0.5)	DPPC (8-10)	(36-46)	(balance to 100)
LNP191	ALC-0315 (50 ± 0.5)	DPPC (10-12)	(36-46)	(balance to 100)
LNP192	ALC-0315 (50 ± 0.5)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP193	LP01 (42-45)	DSPC (8-10)	(36-46)	(balance to 100)
LNP194	LP01 (42-45)	DSPC (10-12)	(36-46)	(balance to 100)
LNP195	LP01 (42-45)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP196	LP01 (42-45)	DOPE (8-10)	(36-46)	(balance to 100)
LNP197	LP01 (42-45)	DOPE (10-12)	(36-46)	(balance to 100)
LNP198	LP01 (42-45)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)

TABLE 5-continued

LNP Systems (all values are a molar ratio relative to a 100 total)				
LNP System#	Ionizable Lipid Component (molar ratio or molar ratio range)	Neutral/Helper Lipid Component (molar ratio or molar ratio range)	Steroid System Component (molar ratio or molar ratio range)	PEGylated Lipid Component (molar ratio or molar ratio range)
LNP199	LP01 (42-45)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP200	LP01 (42-45)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP201	LP01 (42-45)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP202	LP01 (42-45)	DPPC (8-10)	(36-46)	(balance to 100)
LNP203	LP01 (42-45)	DPPC (10-12)	(36-46)	(balance to 100)
LNP204	LP01 (42-45)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP205	LP01 (45-50)	DSPC (8-10)	(36-46)	(balance to 100)
LNP206	LP01 (45-50)	DSPC (10-12)	(36-46)	(balance to 100)
LNP207	LP01 (45-50)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP208	LP01 (45-50)	DOPE (8-10)	(36-46)	(balance to 100)
LNP209	LP01 (45-50)	DOPE (10-12)	(36-46)	(balance to 100)
LNP210	LP01 (45-50)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP211	LP01 (45-50)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP212	LP01 (45-50)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP213	LP01 (45-50)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP214	LP01 (45-50)	DPPC (8-10)	(36-46)	(balance to 100)
LNP215	LP01 (45-50)	DPPC (10-12)	(36-46)	(balance to 100)
LNP216	LP01 (45-50)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP217	LP01 (44-46)	DSPC (8-10)	(36-46)	(balance to 100)
LNP218	LP01 (44-46)	DSPC (10-12)	(36-46)	(balance to 100)
LNP219	LP01 (44-46)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP220	LP01 (44-46)	DOPE (8-10)	(36-46)	(balance to 100)
LNP221	LP01 (44-46)	DOPE (10-12)	(36-46)	(balance to 100)
LNP222	LP01 (44-46)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP223	LP01 (44-46)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP224	LP01 (44-46)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP225	LP01 (44-46)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP226	LP01 (44-46)	DPPC (8-10)	(36-46)	(balance to 100)
LNP227	LP01 (44-46)	DPPC (10-12)	(36-46)	(balance to 100)
LNP228	LP01 (44-46)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP229	LP01 (45 ± 0.5)	DSPC (8-10)	(36-46)	(balance to 100)
LNP230	LP01 (45 ± 0.5)	DSPC (10-12)	(36-46)	(balance to 100)
LNP231	LP01 (45 ± 0.5)	DSPC (10 ± 0.5)	(36-46)	(balance to 100)
LNP232	LP01 (45 ± 0.5)	DOPE (8-10)	(36-46)	(balance to 100)
LNP233	LP01 (45 ± 0.5)	DOPE (10-12)	(36-46)	(balance to 100)
LNP234	LP01 (45 ± 0.5)	DOPE (10 ± 0.5)	(36-46)	(balance to 100)
LNP235	LP01 (45 ± 0.5)	DOTMA (8-10)	(36-46)	(balance to 100)
LNP236	LP01 (45 ± 0.5)	DOTMA (10-12)	(36-46)	(balance to 100)
LNP237	LP01 (45 ± 0.5)	DOTMA (10 ± 0.5)	(36-46)	(balance to 100)
LNP238	LP01 (45 ± 0.5)	DPPC (8-10)	(36-46)	(balance to 100)
LNP239	LP01 (45 ± 0.5)	DPPC (10-12)	(36-46)	(balance to 100)
LNP240	LP01 (45 ± 0.5)	DPPC (10 ± 0.5)	(36-46)	(balance to 100)

**[0803]** In some embodiments of any one LNP system LNP001 to LNP240, the LNP system comprises the steroid component in a molar ratio from about 36 to about 46, or from about 38 to about 42. In some embodiments of any one LNP system LNP001 to LNP240, the LNP system comprises the steroid component in a molar ratio of about 36±0.5, about 37±0.5, about 38±0.5, about 39±0.5, about 40±0.5, about 41±0.5, about 42±0.5, about 43±0.5, about 44±0.5, about 45±0.5, or about ±0.5. In some embodiments, the LNP system comprises the steroid component in a molar ratio of about 37, about 38, about 39, about 40, about 41, or about 42. In some embodiments of any one LNP system LNP001 to LNP240, the steroid component comprises cholesterol. In some embodiments of any one LNP system LNP001 to LNP240, the steroid component comprises cholesterol and one or more additional steroids, wherein cholesterol is about 60% (w/w) or more, about 65% (w/w) or more, about 70% (w/w) or more, about 75% (w/w) or more, or about 80% (w/w) or more of the steroid component. In some embodiments, the steroid component comprises dexamethasone. In

some embodiments, the steroid component comprises a modified cholesterol, e.g., a hydroxy- or alkyl modification. In some embodiments, the steroid component comprises any other steroid disclosed herein. In some embodiments, the LNP system comprises the PEGylated Lipid Component in a molar ratio from about 1.5 to about 2.5. In some embodiments, the PEGylated Lipid Component comprises a PEG2000 lipid. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG and/or DMG-C-PEG. In some embodiments, the PEGylated Lipid Component comprises one or more of DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, C14-PEG2000, C16-PEG2000, and/or C18-PEG2000.

#### Example 9: Screening of LNP Formulations

**[0804]** LNPs described herein typically include a Biodegradable Ionizable Lipid, a Helper phospholipid, Cholesterol (with the potentially addition of dexamethasone or other steroids), and a PEG2000 component. Molar Lipid

Ratio described herein are ratio of ionizable lipid:helper lipid:cholesterol [:Dexamethasone]:PEG.

**[0805]** Modifying molar lipid ratios: Without wishing to be bound by any particular theory, when altering the molar lipid ratio of ionizable lipids, the amount of cholesterol or helper lipid would either be supplemented or decreased. Typically, ionizable lipids are kept at a set ratio, while helper lipids, cholesterol and PEG content are varied. If you were to alter the amount of PEG, you would subsequently adjust the amount of primarily cholesterol and sometimes the helper lipid. If altering the helper lipid, the amount of cholesterol in combination with this is adjusted. Dexamethasone, or another similar steroid in the LNP structure directly replaces some of the cholesterol due to similar chemical structures/function in the LNP.

**[0806]** Typical ionizable lipids are: Dlin-KC2-DMA: molar ratios between 42-50, e.g., and without limitation, 44; Dlin-MC3-DMA: molar ratios between 44-50, e.g., and without limitation, 50; SM-102 (Lipid H): molar ratios between 44-50, e.g., and without limitation, 50; ALC-0315: molar ratios between 44-50, e.g., and without limitation, 50; LP01 (LP000001): molar ratios between 42-50, e.g., and without limitation, 45. Helper Lipids can have molar ratios between 9-11, e.g., and without limitation, 10. Helper lipids include DSPC, DOPE, DOTMA, and DPPC. PEG2000 lipids have typical molar ratios between about 1.5 and about 2.5. These include, without limitation, DMG-PEG, DMG-C-PEG, DMG-PEG2000, DMG-C-PEG2000, DSG-PEG2000, and C14,16,18-PEG2000. Cholesterol molar lipid ratios are typically between about 36 and about 46. Dexamethasone or similar steroids supplement a portion of the molar lipid ratio of cholesterol, for example, and without limitation, a ratio of cholesterol:dexamethasone (C:D) of 9:1. Molar lipid ratio ranges of Cholesterol can be, without limitation, between about 8 and about 10 and Dexamethasone, without limitation, between about 0.1 and about 2. Without wishing to be bound by any particular theory, modifications or replacements for cholesterol can be considered, for example, and without limitation, hydroxy- or alkyl modification (e.g., to improve mRNA delivery) or substitution with potentially therapeutic moieties such as anti-inflammatory steroids. In some embodiments, Molar N/P ratios can be between 1-8. Without wishing to be bound by any particular, it is believed that to reduce inflammatory response of LNPs *in vivo*, the ideal range is 1-5 for N/P, however, N/P ratios of 6-8 are tested.

**[0807]** In some embodiments, LNP formulations described herein include LP01:DSPC:Cholesterol:DMG-PEG2000, LP01:DSPC:Cholesterol:Dexamethasone:DMG-PEG2000, MC3:DSPC:Cholesterol:DMG-PEG2000, MC3:DSPC:Cholesterol:Dexamethasone:DMG-PEG2000, MC3:DSPC:Cholesterol:DMG-C-PEG2000, SM-102:DSPC:Cholesterol:Dexamethasone:DMG-C-PEG2000, SM-102:DSPC:Cholesterol:DMG-PEG2000, SM-102:DSPC:

Cholesterol:Dexamethasone:DMG-PEG2000, ALC-0315:DSPC:Cholesterol:DMG-PEG2000, ALC-0315:DSPC:Cholesterol:Dexamethasone:DMG-PEG2000. In some embodiments, molar ratios of formulations described herein include 50:10:38.5:1.5; 45:9:44:2.

**[0808]** Biophysical Assays: LNPs efficiency to encapsulate nucleic acids, meet sizing criteria and are homogeneity, stability after freeze-thaw cycles. Payloads encapsulated: GFP mRNA, Luciferase mRNA, and our CRISPR/cas9 therapeutic (sgRNA and cas9 mRNA).

TABLE 6

LNPs of various formulations created and screened.			
ID	LNP Composition	Ratio	RNA Payload
LNP 02	MC3:DSPC:Cholesterol:D MG-PEG2000	50:10:38.5:1.5	OCB02::cas9 mRNA
LNP 03	MC3:DSPC:Cholesterol:De xamethasone:DMG- PEG2000	50:10:34.65: 3.85:1.5	OCB02::cas9 mRNA
LNP 04	MC3:DSPC:Cholesterol:D MG-C-PEG2000	50:10:38.5:1.5	OCB02::cas9 mRNA
LNP 05	KC2:DPPC:Cholesterol:C1 6-C-PEG2000	47.4:10.1: 39.5:4	Luciferase mRNA
LNP 07	MC3:DPSC:Cholesterol:D MG-PEG2000	50:10:38.5:1.5	Luciferase mRNA

#### Addition Notes on LNP Components:

**[0809]** Ionizable lipid (titratable charge): Contains a tertiary amine, making this positively charged at an acidic pH, neutrally charged at a physiological pH. Become protonated after endosomal uptake into the cytosol. See below for specifications on different types of ionizable lipids.

**[0810]** Helper phospholipid: anionic endosomal phospholipids that interact with protonated ionizable lipids to form cone shaped ion pairs that enhance cell membrane fusion and disruption, endosomal escape, and cargo release into cell cytosol.

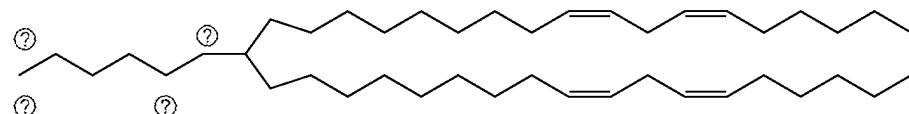
**[0811]** Cholesterol: maintains nanoparticle membrane integrity, assists encapsulation of nucleic acids, and enhances circulation by reducing surface bound proteins.

**[0812]** Dexamethasone or similar steroids: provide an anti-inflammatory component to the LNPs, which decrease immunogenicity and increase transfection rates, particularly *in vivo*.

**[0813]** PEGylated lipid (PEG2000): improves circulation half-life, reduces aggregation of LNPs, and reduces interactions with serum proteins such as opsonins, enhancing stability *in vivo*. Additionally assists particle stability, particle sizing, and biodistribution.

#### Ionizable Lipids:

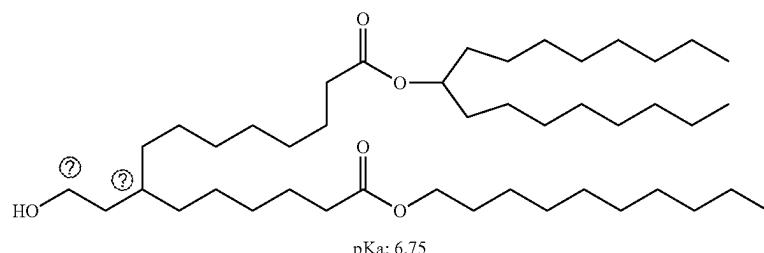
##### A. Dlin-MC3-DMA:



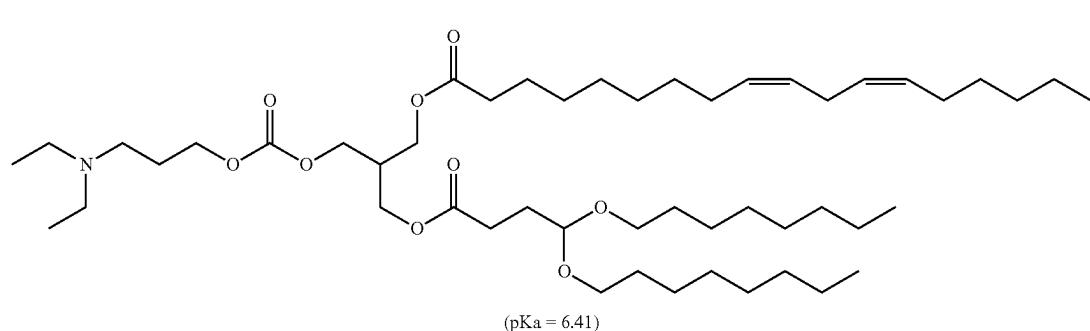
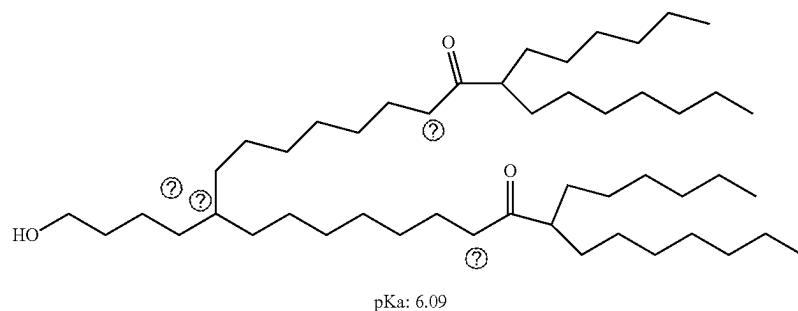
pKa: 6.44

-continued

B. SM-102:



C.Alc-0315:



(?) indicates text missing or illegible when filed

**[0814]** Cationic lipids: such as DODMA, DOTMA, and DOTAP have a quaternary amine group, which leaves them permanently positively charged. Use of this alone was in the past proven to have poor circulation and increased toxicity *in vivo*. Led to the development of ionizable lipids.

**[0815]** Ionizable lipids are characterised by replacing quaternary amine to be tertiary, which enables them to be pH dependent, i.e., is neutrally charged at physiological pH and becomes positively charged (protonated) at acidic pH.

**[0816]** This increases circulation half-life and reduces toxicity *in vivo*.

**[0817]** pKa of ionizable lipid drives performance *in vivo*. pKas between 6-6.7 have been shown to be optimal for delivery of RNA therapeutics. However, the relative pKa of combination of all components in LNP influences LNP transfection—tertiary amine, quaternary amino and hydroxyl group from ionizable lipid, helper lipids and cholesterol all alter relative pKa due to proximity between headgroups. This subsequently affects overall surface charge.

**[0818]** PH dependency enables efficient encapsulation of RNAs in acidic buffer as well as assists RNA release once uptaken by cells.

**[0819]** Further, these lipid pairs form an inverted hexagonal HII phase, which assists membrane disruption, endosomal escape, and subsequent payload release into cytosol of cells.

**[0820]** Albertson et al., 2022, Lipid packing theory showing relationship between amphipathic compounds and geometry once self-assembled. Proposed mechanism by which ionizable lipids mediate endosomal disruption (also known as molecular shape hypothesis). <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC9250827/>

**[0821]** MC3, ALC-0135, SM-102 all have tertiary amines (making them ionizable), no stereo centres, and ester linkers. ALC-0135 and SM-102 have been improved upon MC3 to incorporate additional ester bonds (to assist with biodegradability and reduce bioaccumulation).

**[0822]** Unsaturation of linear tails increases delivery efficiency and fluidity in ionizable lipids, i.e., this enables bilayer lipids to form a non-bilayer phase that will increase membrane disruption and payload release. Additional research explored branching the tails of ionizable lipids, which further increased potency when delivering mRNA therapeutics due to an increase in protonation of ionizable lipids at endosomal pH and cross-section of lipid tails. This modification also led to an increase in cone-shape structure,

which facilitates membrane disruption, endosomal escape, and payload release into the cytosol (Albertson et al., 2022).

[0823] LP01 incorporates increase in ester bonds as well as branching of tails, which has been shown to be an efficient and safe delivery system for in-vivo gene editing in animal models. LP01 has less liver bioaccumulation and fewer safety risks.

#### REFERENCES

[0824] Hald Albertsen, C, Kulkami J A., Witzigmann D, Lind M, Petersson K and Simonsen J B. The role of lipid components in lipid nanoparticles for vaccines and gene therapy. Advanced Drug Delivery Reviews 2022; 188: 114416.

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[0828] All publications, patents, and patent applications herein are incorporated by reference to the same extent as if each individual publication, patent, or patent application was specifically and individually indicated to be incorporated by reference. In the event of a conflict between a term herein and a term in an incorporated reference, the term herein controls.

[0829] While some embodiments have been shown and described herein, it will be obvious to those skilled in the art that such embodiments are provided by way of example only. It is not intended that the disclosure be limited by the specific examples provided within the specification. While the disclosure has been described with reference to the aforementioned specification, the descriptions and illustrations of the embodiments herein are not meant to be construed in a limiting sense. Numerous variations, changes, and substitutions will now occur to those skilled in the art without departing from the disclosure.

[0830] Furthermore, it shall be understood that all aspects of the disclosure are not limited to the specific depictions, configurations or relative proportions set forth herein which depend upon a variety of conditions and variables. It should be understood that various alternatives to the embodiments of the disclosure described herein can be employed in practicing the disclosure. It is therefore contemplated that the disclosure shall also cover any such alternatives, modifications, variations or equivalents. It is intended that the following claims define the scope of the disclosure and that methods and structures within the scope of these claims and their equivalents be covered thereby.

[0831] The examples set forth above are provided to give those of ordinary skill in the art a complete disclosure and description of how to make and use the embodiments of the compositions, systems and methods of the disclosure, and are not intended to limit the scope of what the inventors regard as their invention. Modifications of the above-described modes for carrying out the embodiments of the disclosure that are obvious to persons of skill in the art are intended to be within the scope of the following claims. All patents and publications mentioned in the specification are

indicative of the levels of skill of those skilled in the art to which the disclosure pertains.

[0832] All headings and section designations are used for clarity and reference purposes only and are not to be considered limiting in any way. For example, those of skill in the art will appreciate the usefulness of combining various aspects from different headings and sections as appropriate according to the spirit and scope of the disclosure described herein.

[0833] It is to be understood that the methods described herein are not limited to the particular methodology, protocols, subjects, and sequencing techniques described herein and as such can vary. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to limit the scope of the methods and compositions described herein, which will be limited only by the appended claims. While some embodiments of the present disclosure have been shown and described herein, it will be obvious to those skilled in the art that such embodiments are provided by way of example only. Numerous variations, changes, and substitutions will now occur to those skilled in the art without departing from the disclosure. It should be understood that various alternatives to the embodiments of the disclosure described herein can be employed in practicing the disclosure. It is intended that the following claims define the scope of the disclosure and that methods and structures within the scope of these claims and their equivalents be covered thereby.

[0834] Several aspects are described with reference to example applications for illustration. Unless otherwise indicated, any embodiment can be combined with any other embodiment. It should be understood that numerous specific details, relationships, and methods are set forth to provide a full understanding of the features described herein. A skilled artisan, however, will readily recognize that the features described herein can be practiced without one or more of the specific details or with other methods. The features described herein are not limited by the illustrated ordering of acts or events, as some acts can occur in different orders and/or concurrently with other acts or events. Furthermore, not all illustrated acts or events are required to implement a methodology in accordance with the features described herein.

[0835] While some embodiments have been shown and described herein, it will be obvious to those skilled in the art that such embodiments are provided by way of example only. It is not intended that the disclosure be limited by the specific examples provided within the specification. While the disclosure has been described with reference to the aforementioned specification, the descriptions and illustrations of the embodiments herein are not meant to be construed in a limiting sense. Numerous variations, changes, and substitutions will now occur to those skilled in the art without departing from the disclosure.

[0836] Furthermore, it shall be understood that all aspects of the disclosure are not limited to the specific depictions, configurations or relative proportions set forth herein which depend upon a variety of conditions and variables. It should be understood that various alternatives to the embodiments of the disclosure described herein can be employed in practicing the disclosure. It is therefore contemplated that the disclosure shall also cover any such alternatives, modifications, variations or equivalents. It is intended that the following claims define the scope of the disclosure and that methods and structures within the scope of these claims and their equivalents be covered thereby.

## SEQUENCE LISTING

The patent application contains a lengthy sequence listing. A copy of the sequence listing is available in electronic form from the USPTO web site (<https://seqdata.uspto.gov/?pageRequest=docDetail&DocID=US20250263681A1>). An electronic copy of the sequence listing will also be available from the USPTO upon request and payment of the fee set forth in 37 CFR 1.19(b)(3).

What is claimed is:

1. A pharmaceutical composition for treating a disorder, the composition comprising a plurality of lipid nanoparticles (LNPs) encapsulating:
  - (i) an RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease; and
  - (ii) at least one guide RNA or a nucleic acid encoding at least one guide RNA targeting a gene selected from FGF2, CCN2, NGF, NTF3, NTF4, BDNF, FGFR1, NGFR, NTRK1, NTRK2, ADAM17, ADAMTS1, ADAMTS5, MMP1, MMP2, MMP3, MMP7, MMP8, MMP10, MMP12, MMP13, TIMP1, TIMP3, CXCL1, CXCL2, CXCL3, CXCL5, CXCL6, CXCL8, CCL2, CCL3, CCL5, CCL7, CCL20, IL1A, IL1B, IL4, IL6, IL10, IL13, IL17A, IL18, TNF, CXCR1, CXCR2, CCR7, TNFRSF1A, TNFRSF1B, IL1R1, IL1RAP, IL4R, IL6R, IL6ST, IL10RA, IL10RB, IL13RA1, IL13RA2, IL17RA, IL18R1, IL18RAP, SCN1A, SCN2A, SCN3A, SCN4A, SCN5A, SCN8A, SCN9A, SCN10A, SCN11A, TAC1, TAC3, TACR1, TACR2, TACR3, ATP1A1, CALCA, CALCB, CALCRL, RAMP1, ADM, CRCP, YAP1, MRGPRX2, TGFB, TGFB1, and TGFB2.
2. The pharmaceutical composition of claim 1, wherein the at least one guide RNA comprises a crRNA sequence selected from any one of SEQ ID NOs: 1-2731.
3. The pharmaceutical composition of claim 1 or 2, wherein the disorder is a musculoskeletal disorder.
4. The pharmaceutical composition of claim 3, wherein the musculoskeletal disorder is selected from the group consisting of Rheumatoid Arthritis, Gout, Osteoarthritis, Osteoporosis, Intervertebral disc disease (IVDD), Psoriatic arthritis (PsA), Arthritis, Polymyositis, Proliferative synovitis, Malignant bone neoplasm, Sarcoid Myopathy, Cortex Bone Disorders, Idiopathic Scoliosis, Tendinopathy, Myofibrillar Myopathy, Enthesis-Related Arthritis, Ankylosing spondylitis, Degenerative polyarthritides, Arthropathy, Osteitis Deformans, Prolapsed Lumbar Disc, Polymyositis Ossificans, Idiopathic Polymyositis, Lunt Disease, Adult-onset Still's Disease, Osteoarthritis Deformans, and Bachet's Disease.
5. The pharmaceutical composition of claim 3, wherein the musculoskeletal disorder is selected from the group consisting of Rheumatoid arthritis, Idiopathic osteoporosis, Post-menopausal osteoporosis, Paget's disease, Osteoarthritis, Juvenile idiopathic arthritis (JIA), Still's disease, Ankylosing spondylitis, Polymyalgia rheumatica, Arthritis, Secondary malignant neoplasm of bone, Type II, Mucolipidoses, Sjogren's Syndrome, Psoriatic arthritis, Rheumatism, Castleman's disease, Degenerative Polyarthritides, Arthropathy, Bone neoplasm, Osteoporosis, Massive osteolysis, Bone fracture healing, Systemic sclerosis, Systemic Juvenile Idiopathic, Arthritis, and Synovitis.
6. The pharmaceutical composition of claim 3, wherein the musculoskeletal disorder is selected from the group consisting of Arthritis, Infectious Intermittent joint effusion, Ankylosing spondylitis, Arthritis, Osteoarthritis, Spondylarthritis, Plantar fasciitis, Degenerative polyarthritis, Hemophilic arthropathy, Inflammatory myopathy with abundant macrophages, Polymyositis, Tendinosis, Malignant Bone Neoplasm, Osteoporosis, Psoriatic arthritis, Rheumatism, Rheumatoid arthritis, Adult Still's disease, Juvenile arthritis, Early rheumatoid arthritis, Palindromic rheumatism, Gout, Infectious Arthritis, Myotonic dystrophy, Dermatomyositis, Hemophilic arthropathy, Osteopenia, Sjogren's syndrome, Juvenile Idiopathic Arthritis, Myasthenia gravis, Osteolysis, Inflammation, Degenerative polyarthritis, Osteitis deformans, Pigmented villonodular synovitis, or Hyperphosphatasemia with bone disease.
7. The pharmaceutical composition of claim 3, wherein the musculoskeletal disorder is selected from the group consisting of Loeys-Dietz Syndrome, Osteoarthritis, Marfan Syndrome, Aortic aneurysm (familial thoracic 3), and a Craniofacial abnormality.
8. The pharmaceutical composition of claim 1 or 2, wherein the disorder is a neoplasia.
9. The pharmaceutical composition of claim 8, wherein the neoplasia is selected from the group consisting of Osteosarcoma, Colon Cancer, Metastasis (General), Lung Cancer, Multiple Myeloma, Breast Cancer, Solid Tumors, Lymphoma, Pancreatic Cancer, Stomach Cancer, Epithelial Ovarian Cancer, Mammary Neoplasms, Oropharyngeal Carcinoma, Renal Cell Carcinoma, Chondrosarcoma, Esophageal Neoplasms, B-Cell Lymphoma, Cutaneous Lymphoma T-Cell, Leukemia, Thyroid Carcinoma, Skin carcinogenesis, Cholelithiasis, Glioma, Liver Cancer, Melanoma, Neuroblastoma, Polycystic Ovary Syndrome, Glioblastoma, Prostate Cancer, Cervical Cancer, Ovarian Cancer, Bladder Cancer, Squamous cell carcinoma, Kaposi Sarcoma, Oral Cavity Cancer, Leiomyosarcoma, Malignant Peripheral Nerve Sheath Tumor, and Ewing's Sarcoma.
10. The pharmaceutical composition of claim 8, wherein the neoplasia is selected from the group consisting of Multiple myeloma, Lung Cancer, Stomach Cancer, Breast Cancer, Kidney Cancer, Neoplasm Metastasis, Colorectal Neoplasms, Ovarian cancer, Osteosarcoma, Cholangiocarcinoma, Leukemia, Prostate cancer, Plasmacytoma, Pancreatic cancer, Cervical cancer, Lymphoma, Liver neoplasms, Neuroblastoma, Melanoma, Mastocytosis, Endometrial cancer, Bladder Cancer, Squamous cell carcinoma, Gallbladder carcinoma, Adenocarcinoma, Thyroid Cancer, prostate neoplasms, stomach cancer, liver carcinoma, pituitary neo-

plasms, hepatoblastoma, multiple myeloma, hepatocellular adenoma, breast carcinoma, carcinogenesis, leukemia, colon carcinoma, melanoma, metastasis (general), lung cancer, pancreatic cancer, Kaposi sarcoma, medulloblastoma, carcinoma of bladder, squamous cell carcinoma, mast cell neoplasm, glioma, mastocytoma, brain neoplasms, ovarian neoplasms, bone neoplasm, rhabdomyosarcoma, solid tumors, metastatic kidney cancer, and metastatic renal cell carcinoma

11. The pharmaceutical composition of claim 8, wherein the neoplasia is selected from the group consisting of a Neoplasm, Glioblastoma, Astrocytoma, Adenocarcinoma, Osteosarcoma, Squamous cell carcinoma, Metastasis, Ameloblastoma, Cholangiocarcinoma, Choriocarcinoma, Ovarian cancer, Prostate cancer, Lung Cancer, Larynx Cancer, Breast Cancer, Lip and Oral Cavity Carcinoma, Squamous intraepithelial lesion, Neuroblastoma, Non-Hodgkin lymphomas, Malignant Neoplasms, Skin Neoplasms, Neuroblastoma, Neoplasm Metastasis, Colorectal Cancer, Fibrosarcoma, Myeloid Leukemia, Myelofibrosis, Hodgkin Lymphomas, Non-Small Cell Lung Carcinoma, Cervical Cancer, Liver cancer, Extrapulmonary small cell carcinoma, Basal cell carcinoma, Kidney cancer, Pancreatic carcinoma, Mesothelioma, Gastric cancer, Polycystic Ovary Syndrome, Chordoma, Cholangiocarcinoma, Malignant ascites, Nasopharyngeal carcinoma, Head and Neck Carcinoma, Cancer metastasis, Prostate carcinoma, Fibrosarcoma, Liver carcinoma, Carcinoma of bladder, T-Cell Lymphoblastic Leukemia, Bladder Neoplasm, melanoma, Leukemia, acute myeloid leukemia, Hepatocellular carcinoma, Colorectal cancer, a Lymphoma, Mycosis fungoides, and Sezary syndrome.

12. The pharmaceutical composition of claim 8, wherein the neoplasia is selected from the group consisting of Pancreatic cancer, Multiple self-healing squamous epithelioma (Ferguson-Smith disease), Pancreatic cancer, Gastrointestinal Stromal Tumors (GIST), Hereditary Nonpolyposis Colorectal Cancer (Lynch Syndrome), Metastasis, Colorectal Carcinoma, Bone neoplasms, Anaplastic carcinoma, Spindle-Cell carcinoma, Malignant Bone Neoplasm, Lung neoplasms, and Malignant brain neoplasm.

13. The pharmaceutical composition of claim 1 or 2, wherein the disorder is a neurological disorder.

14. The pharmaceutical composition of claim 13, wherein the neurological disorder is selected from the group consisting of Acute Thrombotic Stroke, Epilepsy, Multiple Sclerosis, and Alzheimer's Disease.

15. The pharmaceutical composition of claim 1 or 2, wherein the disorder is a cardiac disorder.

16. The pharmaceutical composition of claim 15, wherein the cardiac disorder is selected from the group consisting of Acute Myocardial Infarction, refractory heart failure, arrhythmias, pericarditis, myocarditis, sepsis-induced cardiomyopathy, Acute Decompensated Heart Failure, Refractory Idiopathic Pericarditis, Atherosclerosis, coronary artery disease, myocardial infarction, and cardiac remodeling.

17. The pharmaceutical composition of claim 15, wherein the cardiac disorder is atherosclerosis or abdominal aortic aneurism.

18. The pharmaceutical composition of claim 1 or 2, wherein the disorder is an inflammatory disorder.

19. The pharmaceutical composition of claim 18, wherein the inflammatory disorder is selected from the group consisting of Autoinflammatory Disease (AID), Cryopyrin

Associated Periodic syndrome (CAPS), Familial Mediterranean Fever (FMF), TNF-Receptor Associated Periodic Syndrome (TRAPS), Hyper-IgD Syndrome (HIDS), Systemic Lupus Erythematosus (SLE), and Fibrosis.

20. The pharmaceutical composition of claim 18, wherein the inflammatory disorder is selected from the group consisting of a cytokine storm, acute local inflammation, inflammatory bowel disease, Crohn's disease, sepsis, Experimental sepsis, and Castleman's disease

21. The pharmaceutical composition of claim 1 or 2, wherein the disorder is a digestive disorder.

22. The pharmaceutical composition of claim 21, wherein the digestive disorder is selected from the group consisting of Inflammatory Bowel Disease (IBD), Gastroesophageal reflux disease (GERD), and Non-HP-associated Peptic Ulcer Disease.

23. The pharmaceutical composition of claim 1 or 2, wherein the disorder is a respiratory disorder.

24. The pharmaceutical composition of claim 23, wherein the respiratory disorder is Asthma or Pulmonary Fibrosis.

25. The pharmaceutical composition of claim 1 or 2, wherein the disorder is a renal, metabolic, or ophthalmic disorder.

26. The pharmaceutical composition of claim 25, wherein the renal, metabolic, or ophthalmic disorder is selected from the group consisting of Chronic Kidney Disease, Type 2 Diabetes, an Eye Disease, Uveitis, Scleritis, Sjogren asthenia, and dry eye.

27. The pharmaceutical composition of claim 1 or 2, wherein the disorder is an autoimmune disorder.

28. The pharmaceutical composition of claim 27, wherein the autoimmune disorder is Systemic Lupus Erythematosus.

29. The pharmaceutical composition of claim 1 or 2, wherein the disorder is acute synovitis, chronic synovitis, inflammatory arthritis, or immune-mediated arthritides.

30. The pharmaceutical composition of claim 1 or 2, wherein the disorder is an autoimmune or inflammatory disorder.

31. The pharmaceutical composition of claim 29, wherein the autoimmune or inflammatory disorder is selected from the group consisting of an Allergy, Asthma, Multiple sclerosis, Psoriasis, Inflammatory bowel disease, Autoimmune experimental uveitis, Colitis, Hypersensitivity reaction disease, Diabetes, Crohn's disease, Systemic lupus erythematosus, Pulmonary sarcoidosis, Leishmaniasis, Experimental autoimmune encephalomyelitis, HTLV-1-associated myelopathy, Tropical spastic paraparesis, Scleroderma, an Idiopathic inflammatory myopathy, polymyositis, and dermatomyositis.

32. The pharmaceutical composition of any one of claims 1-31, wherein the RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease is the RNA-guided nuclease.

33. The pharmaceutical composition of any one of claims 1-31, wherein the RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease is DNA encoding the RNA-guided nuclease.

34. The pharmaceutical composition of any one of claims 1-31, wherein the RNA-guided nuclease or a nucleic acid encoding an RNA-guided nuclease is mRNA encoding the RNA-guided nuclease.

35. The pharmaceutical composition of any one of claims 1-34, wherein the RNA-guided nuclease is a Cas protein.

- 36.** The pharmaceutical composition of claim **35**, wherein the Cas protein is a Cas9 protein.
- 37.** The pharmaceutical composition of claim **35**, wherein the Cas9 protein is an *S. pyogenes* Cas9 polypeptide.
- 38.** The pharmaceutical composition of claim **35**, wherein the Cas9 protein is selected from the group consisting of esCas9, hfCas9, peCas9, and ARCas9.
- 39.** The pharmaceutical composition of any one of claims **1-38**, wherein the at least one guide RNA or a nucleic acid encoding at least one guide RNA is the at least one guide RNA.
- 40.** The pharmaceutical composition of any one of claims **1-38**, wherein the at least one guide RNA or a nucleic acid encoding at least one guide RNA is DNA encoding the at least one guide RNA.
- 41.** The pharmaceutical composition of any one of claims **1-38**, comprising a nucleic acid encoding both the RNA-guided nuclease and the at least one guide RNA.
- 42.** The pharmaceutical composition of any one of claims **1-41**, wherein the at least one guide RNA is a single guide RNA (sgRNA).
- 43.** The pharmaceutical composition of any one of claims **1-42**, wherein the at least one guide RNA targets a human gene.
- 44.** The pharmaceutical composition of any one of claims **1-42**, wherein the at least one guide RNA targets a canine gene.
- 45.** The pharmaceutical composition of any one of claims **1-42**, wherein the at least one guide RNA targets an equine gene.
- 46.** The pharmaceutical composition of any one of claims **1-42**, wherein the at least one guide RNA targets a feline gene.
- 47.** The pharmaceutical composition of any one of claims **1-42**, wherein the at least one guide RNA targets a mammalian gene.
- 48.** The pharmaceutical composition of any one of claims **1-47**, wherein the composition is formulated for parenteral administration.
- 49.** The pharmaceutical composition of any one of claims **1-47**, wherein the composition is formulated for intra-articular injection within a joint of the subject.
- 50.** The pharmaceutical composition of any one of claims **1-47**, wherein the composition is formulated for intradiscal injection.
- 51.** The pharmaceutical composition of any one of claims **1-47**, wherein the composition is formulated for peridiscal injection.
- 52.** The pharmaceutical composition of any one of claims **1-47**, wherein the composition is formulated for intravertebral injection.
- 53.** The pharmaceutical composition of any one of claims **1-47**, wherein the composition is formulated for epidural injection.
- 54.** The pharmaceutical composition of any one of claims **1-47**, wherein the composition is formulated for injection to the facet joints of the spine.
- 55.** The pharmaceutical composition of any one of claims **1-54**, wherein the one or more LNPs comprise one or more ionizable lipids selected from: 3-(didodecylamino)-N1,N1,4-tridodecyl-1-piperazineethanamine (KL10), N1-[2-(dido-decylamino)ethyl]-N1,N4,N4-tridodecyl-1,4-piperazinediethanamine (KL22), 14,25-ditridodecyl-15,18,21,24-tetraaza-octatriacontane (KL25), 1,2-dilinoleyoxy-N,N-

dimethylaminopropane (DLin-DMA), 2,2-dilinoleyl-4-dimethylaminomethyl-[1,3]-dioxolane (DLin-K-DMA), heptatriaconta-6,9,28,31-tetraen-19-yl 4-(dimethylamino)butanoate (DLin-MC3-DMA), 2,2-dilinoleyl-4-(2-dimethylaminoethyl)-[1,3]-dioxolane (DLin-KC2-DMA), 1,2-dioleyoxy-N,N-dimethylaminopropane (DODMA), 2-({8-[{3(beta.)-cholest-5-en-3-yloxy}octyl]oxy}-N,N-dimethyl-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA), (2R)-2-({8-[{3(beta.)-cholest-5-en-3-yloxy}octyl]oxy}-N,N-dimethyl-3-[(9Z,-12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA (2R)), (2S)-2-({8-[{3(beta.)-cholest-5-en-3-yloxy}octyl]oxy}-N,N-dimethyl-3-[(9Z,-12Z)-octadeca-9,12-dien-1-yloxy]propan-1-amine (Octyl-CLinDMA (2S)), LP01, ALC-0315, SM-102, a lipid including a cyclic amine group, and mixtures thereof.

**56.** The pharmaceutical composition of claim **55**, wherein the ionizable lipid is DLin-KC2-DMA.

**57.** The pharmaceutical composition of claim **55**, wherein the ionizable lipid is DLin-MC3-DMA.

**58.** The pharmaceutical composition of claim **55**, wherein the ionizable lipid is SM-102.

**59.** The pharmaceutical composition of claim **55**, wherein the ionizable lipid is ALC-0315.

**60.** The pharmaceutical composition of claim **55**, wherein the ionizable lipid is LP01.

**61.** The pharmaceutical composition of any one of claims **1** to **54**, wherein the one or more LNPs comprise one or more helper lipid selected from: 1,2-dilinoleoyl-sn-glycero-3-phosphocholine (DLPC), 1,2-dimyristoyl-sn-glycero-phosphocholine (DMPC), 1,2-dioleoyl-sn-glycero-3-phosphocholine (DOPC), 1,2-dipalmitoyl-sn-glycero-3-phosphocholine (DPPC), 1,2-distearoyl-sn-glycero-3-phosphocholine (DSPC), 1,2-diundecanoyl-sn-glycero-phosphocholine (DUPC), 1-palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine (POPC), 1,2-di-O-octadecenyl-sn-glycero-3-phosphocholine (18:0 Diether PC), 1-oleoyl-2-cholesterylhemisuccinoyl-sn-glycero-3-phosphocholine (OChemsPC), 1-hexadecyl-sn-glycero-3-phosphocholine (C16 Lyso PC), 1,2-dilinolenoyl-sn-glycero-3-phosphocholine, 1,2-diarachidonoyl-sn-glycero-3-phosphocholine, 1,2-didocosahexaenoyl-sn-glycero-3-phosphocholine, 1,2-dioleoyl-sn-glycero-3-phosphoethanolamine (DOPE), 1,2-diphytanoyl-sn-glycero-3-phosphoethanolamine (ME 16.0 PE), 1,2-distearoyl-sn-glycero-3-phosphoethanolamine, 1,2-dilinoleoyl-sn-glycero-3-phosphoethanolamine, 1,2-dilinolenoyl-sn-glycero-3-phosphoethanolamine, 1,2-diarachidonoyl-sn-glycero-3-phosphoethanolamine, 1,2-didocosahexaenoyl-sn-glycero-3-phosphoethanolamine, 1,2-dioleoyl-sn-glycero-3-phospho-rac-(1-glycerol) sodium salt (DOPG), sphingomyelin (SM), and mixtures thereof.

**62.** The pharmaceutical composition of any one of claims **1** to **54**, wherein the one or more LNPs comprise one or more PEG lipids selected from: PEG-modified phosphatidylethanolamines, PEG-modified phosphatidic acids, PEG-modified ceramides, PEG-modified dialkylamines, PEG-modified diacylglycerols, PEG-modified dialkylglycerols, and mixtures thereof. For example, a PEG lipid may be PEG-c-DOMG, PEG-DMG, PEG-DLPE, PEG-DMPE, PEG-DPPC, PEG-DMA or a PEG-DSPE lipid.

**63.** The pharmaceutical composition of any one of claims **1** to **54**, wherein the one or more LNPs comprise one or more structural lipids selected from: cholesterol, fecosterol, stigmasterol, stigmastanol, sitosterol, D-sitosterol, lupeol, betu-

lin, ursolic acid, oleanolic acid, campesterol, fucosterol, brassicasterol, ergosterol, 9, 11-dehydroergosterol, tomatidine, tomatine,  $\alpha$ -tocopherol, and mixtures thereof.

**64.** The pharmaceutical composition of claim **63**, wherein the structural lipid is cholesterol.

**65.** The pharmaceutical composition of claim **63**, wherein the one or more LNPs comprise a structural lipid and a corticosteroid (e.g., prednisolone, dexamethasone, prednisone, and hydrocortisone), or a combination thereof.

**66.** The pharmaceutical composition of claim **65**, wherein the corticosteroid is dexamethasone.

**67.** The pharmaceutical composition of any one of claims **1** to **66**, wherein the LNPs comprise a plurality of particles with a diameter over 100 nm.

**68.** The pharmaceutical composition of any one of claims **1** to **66**, wherein the LNPs comprise a plurality of particles with a diameter between about 60 nm and about 120 nm.

**69.** The pharmaceutical composition of any one of claims **1** to **68**, wherein the plurality of LNPs comprise an LNP system selected from any one of LNP001 to LNP240.

**70.** A method for treating a disease or disorder in a subject in need thereof, the method comprising administering to the subject a pharmaceutical composition of any one of claims **1** to **69**.

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