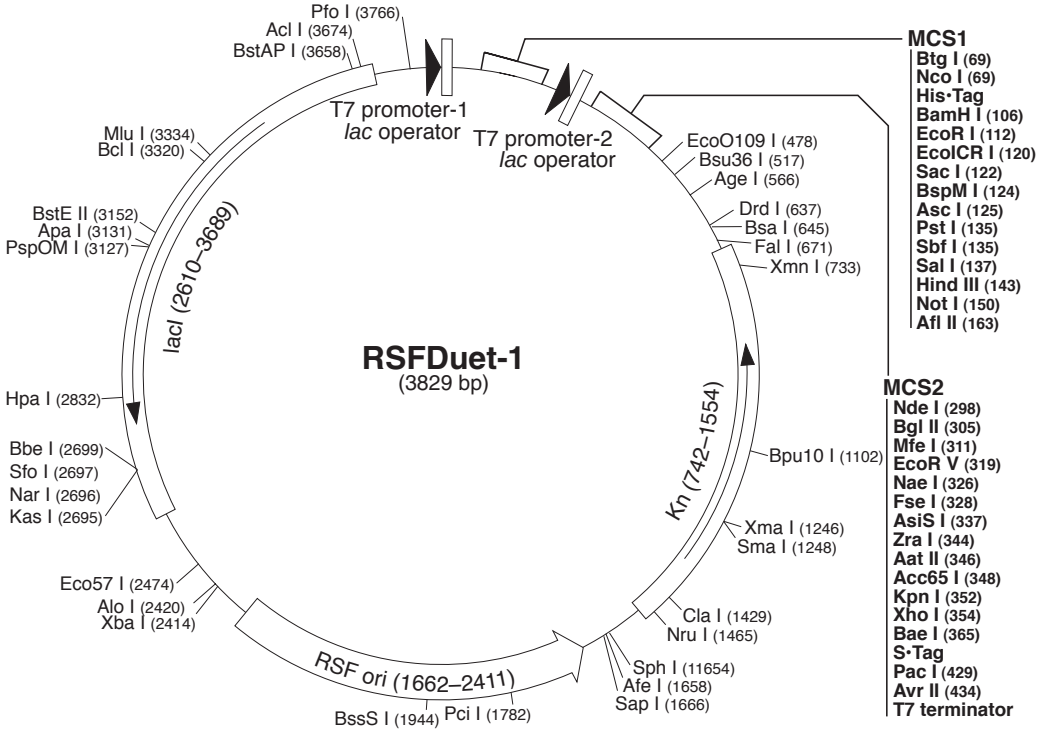
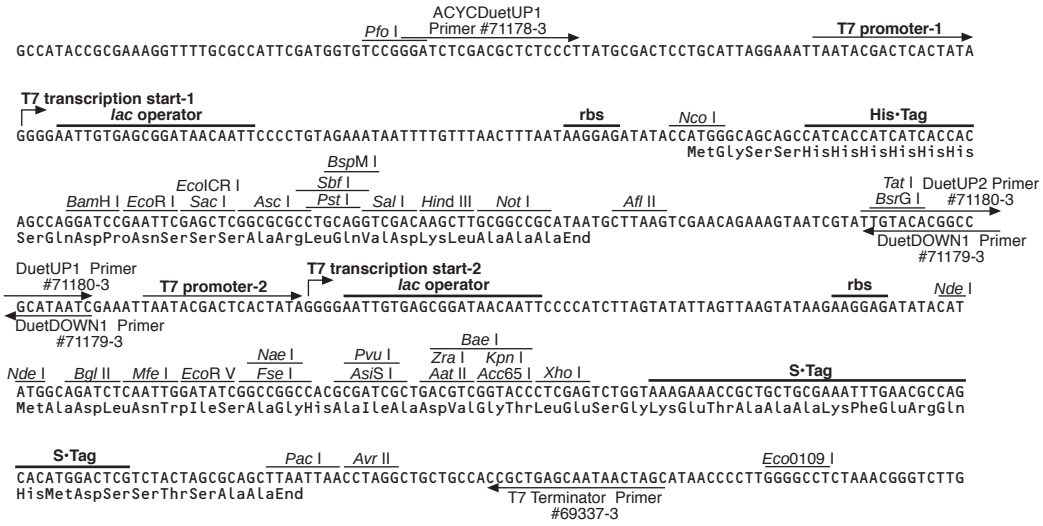
pRSFDuet-1 Vector

TB391 0903

	Cat. No.
pRSFDuet-1 DNA	71341-3
pRSFDuet-1 sequence landmarks	
T7 promoter-1	3582–3598
T7 transcription start-1	1
His•Tag® coding sequence	83–100
Multiple cloning sites-1 (*Nco* I–*Afl* II)	69–168
T7 promoter-2	214–230
T7 transcription start-2	231
Multiple cloning sites-2 (*Nde* I–*Avr* II)	297–438
S•Tag™ coding sequence	366–410
T7 terminator	462–509
kan (Kn^R) coding sequence	742–1554
RSF origin	1662–2411
lacI coding sequence	2610–3689

pRSFDuet™-1 is designed for the coexpression of two target ORFs. The vector contains two multiple cloning sites (MCS), each of which is preceded by a T7*lac* promoter and ribosome binding site (rbs). The vector also carries the RSF1030-derived RSF replicon, *lacI* gene and kanamycin resistance gene (Kn^R). This vector can be used in combination with pACYCDuet™-1 (Cat. No. 71147-3), pCDFDuet™-1 (Cat. Np. 71340-3), and pETDuet™-1 (Cat. No. 71146-3) in an appropriate host strain for the coexpression of 4 to 8 target proteins. ORFs inserted into MCS1 can be sequenced using the ACYCDuetUP1 Primer (Cat. No. 71178-3) and DuetDOWN1 Primer (Cat. No. 71179-3). ORFs inserted into MCS2 can be sequenced using the DuetUP2 Primer (Cat. No. 71180-3) and T7 Terminator Primer (Cat. No. 69337-3). Unique sites are shown on the circle map.





pRSFDuet-1 cloning/expression regions

pRSFDuet-1 Restriction Sites

TB391 0903

Enzyme	# Sites	Locations				
AatII	1	346				
Acc65I	1	348				
AccI	2	138	411			
AclI	1	3674				
AfeI	1	1658				
AflII	1	163				
AflIII	2	1782	3334			
AgeI	1	566				
AloI	1	2420				
ApaI	1	3131				
ApaLI	2	2085	3354			
AscI	1	125				
AseI	6	213	732	921	2592	2651
		3812				
AsiSI	1	337				
AvaI	2	354	1246			
AvrII	1	433				
BaeI	1	365				
BamHI	1	106				
BanI	4	348	2565	2695	3414	
BanII	3	122	1471	3131		
BbeI	1	2699				
BbsI	2	2849	3188			
BceAI	4	211	801	2850	3477	
BcgI	2	162	3014			
BciVI	4	728	1604	1974	2882	
BclI	1	3320				
BfrBI	2	1008	1274			
BglII	1	305				
BlpI	2	451	2309			
Bme1580I	3	2089	3131	3358		
BmrI	3	2536	3176	3413		
BpmI	2	3013	3502			
Bpu10I	1	1102				
BpuEI	5	515	1862	2160	2340	2526
BsaHI	3	343	2696	3379		
BsaI	1	645				
BsaWI	8	551	566	983	1804	1977
		2124	2512	3015		
BsaXI	2	655	2666			
BseYI	3	2075	2800	2935		
BsgI	2	3289	3489			
BsiEI	9	153	199	325	337	636
		1124	1698	2111	2555	
BsiHKAI	3	122	2089	3358		
BsmAI	7	645	1102	1604	2719	3106
		3232	3637			
BsmBI	2	1102	2719			
BsmI	2	1163	1240			
Bsp1286I	5	122	1471	2089	3131	3358
BspCNI	6	443	530	1094	2059	2322
		2755				
BspHI	2	725	1602			
BspLU11I	1	1782				
BspMI	1	124				
BsrBI	5	13	243	723	1608	1715
BsrDI	2	2927	3293			
BsrFI	4	324	566	1164	3648	
BsrGI	1	190				
BssHII	2	125	2923			
BssSI	1	1944				
BstAPI	1	3658				
BstEII	1	3152				
BstXI	3	3288	3411	3540		
BstYI	5	106	305	869	2558	3770
Bsu36I	1	517				
BtgI	1	69				
BtsI	5	543	1176	1263	2607	2975
ClaI	1	1429				
DrdI	1	637				
EaeI	5	150	196	322	326	2660
EagI	3	150	196	322		

Enzyme	# Sites	Locations				
EarI	5	1306	1562	1666	2442	3717
EciI	3	1833	1979	3549		
Ecl136II	1	120				
Eco57I	1	2474				
Eco57MI	3	2474	3013	3502		
EcoICRI	1	120				
EcoNI	2	1209	3802			
EcoO109I	1	478				
EcoRI	1	112				
EcoRV	1	319				
FalI	1	671				
FseI	1	328				
HaeII	5	1660	2019	2699	2942	3723
HincII	3	139	2392	2832		
HindIII	1	143				
HpaI	1	2832				
KasI	1	2695				
KpnI	1	352				
MfeI	1	311				
MluI	1	3334				
MslI	3	2968	2998	3286		
NaeI	1	326				
NarI	1	2696				
NcoI	1	69				
NdeI	1	298				
NgoMIV	1	324				
NotI	1	150				
NruI	1	1465				
NsiI	2	1010	1276			
NspI	2	1654	1786			
PacI	1	429				
PciI	1	1782				
PflMI	3	401	862	3759		
PfoI	1	3766				
PinAI	1	566				
PspOMI	1	3127				
PstI	1	135				
PvuI	1	337				
PvuII	2	2645	2738			
SacI	1	122				
SalI	1	137				
SapI	1	1666				
SbfI	1	135				
SfcI	4	29	131	226	3825	
SfoI	1	2697				
SmaI	1	1248				
SmlI	7	163	354	494	1877	2139
		2319	2541			
SphI	1	1654				
Sse8387I	1	135				
SspI	2	1197	1571			
StyI	4	69	433	473	2287	
TaqII	4	870	1684	2368	2541	
TatI	1	190				
TspGWI	3	1303	1315	2396		
Tth111I	2	637	2209			
XbaI	1	2414				
XcmI	3	2949	2967	3483		
XhoI	1	354				
XmaI	1	1246				
XmnI	1	733				
ZraI	1	344				

Enzymes that do not cut pRSFDuet-1:

AarI	AhdI	AleI	AlwNI	BbvCI
BglI	BmgBI	BmtI	BplI	BsaAI
BsaBI	BseRI	BsiWI	BsmFI	BspEI
Bst1107I	BstBI	BstZ17I	BtrI	DraI
DraIII	FspAI	FspI	MscI	NheI
NspV	PmeI	PmlI	PpiI	PpuMI
PshAI	PsiI	PsrI	RsrII	SacII
SanDI	ScaI	SexAI	SfiI	SgrAI
SnaBI	SpeI	SrfI	StuI	SwaI