

Supervised clustering reference region approach: Pipeline for generating the tissue kinetic classes & extracting the supervised reference region

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Main package contents

The package contains the set of MATLAB functions and scripts to 1) **generate the tissue kinetic classes** (for a given tracer X) and to 2) **extract the reference region with the supervised clustering method** (given the classes generated in 1)

It also contains several UTILITY functions (e.g. computeIDIF.m, dynPET_normalisation.m) that are ancillary to the main code.

Representative simulated data for testing the scripts can be made available upon request at

https://molecularneuroimaging.files.wordpress.com/2020/11/svca_testingdata.zip

Matlab main function functions

PART 1 – Generation of tissue kinetic classes

- Main Function: **superX_extractNormTACs.m**
- *Supporting script: exampleScript_normTACextraction.m*
- *Inputs:* dynamic PET, brain mask, white matter mask, gray matter mask, high binding mask and PET mid-frame times
- *Outputs:* normalised time activity curves for gray matter, white matter, image-derived blood input function, high binding gray matter at single subject
- Post-processing and QC

PART 2 - Supervised reference region extraction

- Main function: **superX_refExtraction.m**
- *Supporting script: exampleScript_refExtraction.m*
- *Inputs:* dynamic PET, kinetic classes, brain mask, mask of prospective reference area, PET mid-frame times
- *Outputs:* reference region time activity curve, reference region mask
- Post-processing and QC

PART 1 – Generation of tissue kinetic classes

INSTRUCTIONS:

- Select a group of N healthy controls (preferably $N > 10$) for which both dynamic PET and structural MRI images are available
- For each control, extract the normalised kinetic TACs using the function **superX_extractNormTACs.m**.
- Compute the median of the normalised TACs across the group of controls and define the tissue kinetic classes that will be used to apply the supervised clustering

NOTES:

- The pipeline works only if the PET images are acquired consistently with the same protocol (i.e. same tracer, injection type, length of acquisition, framing, etc)
- The sample used to generate the kinetic classes should be independent from the one to which the supervised clustering is applied

PART 1 – Generation of tissue kinetic classes

Inputs for `superX_extractNormTACs.m`:

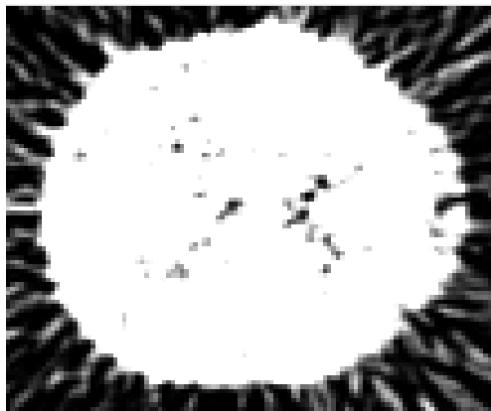
- *dynPET*: 4D matrix containing dynamic PET data
- *brainMask*: 3D matrix containing binary data specifying the brain voxels for the kinetic normalisation (ideally skull should not be included in the brainMask)
- *grayMatterMask*: 3D matrix containing binary brain data specifying the gray matter voxels (possibly >90% of probability), but excluding voxels with high specific binding or those used to define the highBindMask
- *whiteMatterMask*: 3D matrix containing binary data specifying white matter voxels (possibly >90% of probability)
- *highBindMask*: 3D matrix containing binary data specifying high binding gray matter voxels. In case of TSPO PET tracer, thalamus has been used as proxy for these voxels.
- *time*: 2D double with PET mid-frame times (minutes)

Notes:

- The first three dimensions of dynPET must match the dimensions of brain and tissue masks
- The length of the time vector must match with number of frames in dynPET.

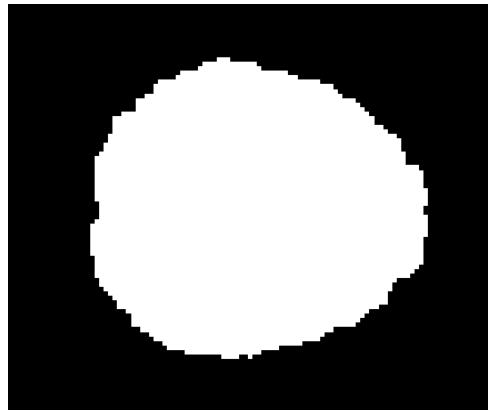
PART 1 – Generation of tissue kinetic classes

Representative inputs



dynPET

4-D double



brainMask

91x109x91 double

time	
	26x1 double
1	1
2	0.5000
3	1.1250
4	1.3750
5	1.6250
6	1.8750
7	2.1250
8	2.3750
9	2.6250
10	2.8750
11	3.5000
12	4.5000
13	5.5000
14	7
15	9
16	11
17	13
18	15
19	18.5000
20	23.5000
21	28.5000
22	33.5000
23	38.5000
24	43.5000
25	48.5000
26	53.5000
	58.5000

time

26x1 double

grayMask

91x109x91 double



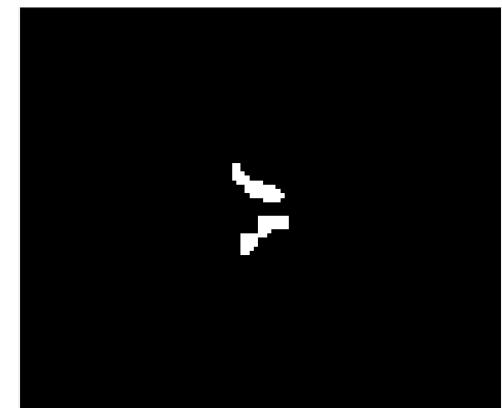
whiteMask

91x109x91 double



highBindMask

91x109x91 double



PART 1 – Generation of tissue kinetic classes

Representative outputs

The function returns 4 vectors (normGrayTAC, normWhiteTAC, normIDIF, normHighBindTAC) which represent the normalised time activity curves (TACs) for gray matter, white matter, image-derived blood input function and high binding gray matter, respectively.

normGrayTAC		
26x1 double		
1		
1	6.3083e-...	
2	-0.0100	
3	0.0952	
4	0.2085	
5	0.2394	
6	0.2507	
7	0.2681	
8	0.2772	
9	0.2776	
10	0.4929	
11	0.5081	
12	0.4960	
13	0.5627	
14	0.5509	
15	0.5468	
16	0.5236	
17	0.5123	
18	0.5647	
19	0.5143	
20	0.4715	
21	0.4287	
22	0.3675	
23	0.3221	
24	0.2747	
25	0.2355	
26	0.1973	

normWhiteTAC		
26x1 double		
1		
1	0.0285	
2	0.0186	
3	-0.2903	
4	-0.2664	
5	-0.2726	
6	-0.3041	
7	-0.2775	
8	-0.2775	
9	-0.2810	
10	-0.5129	
11	-0.5206	
12	-0.4738	
13	-0.5469	
14	-0.5531	
15	-0.5167	
16	-0.5049	
17	-0.4853	
18	-0.5080	
19	-0.4596	
20	-0.3939	
21	-0.3415	
22	-0.2771	
23	-0.2057	
24	-0.1613	
25	-0.1262	
26	-0.0625	

normHighBindTAC		
26x1 double		
1		2
1	0.2117	
2	-0.1036	
3	0.2720	
4	0.2840	
5	0.3745	
6	0.3742	
7	0.3649	
8	0.8104	
9	0.2820	
10	0.6334	
11	0.7382	
12	0.5086	
13	0.7467	
14	0.7719	
15	0.9222	
16	1.1450	
17	0.7422	
18	1.2108	
19	1.4336	
20	1.5524	
21	1.5088	
22	1.4713	
23	1.6568	
24	1.4878	
25	1.7556	
26	1.4162	

normIDIF		
26x1 double		
1		
1	1.6849	
2	4.2896	
3	4.9780	
4	2.0685	
5	0.6363	
6	0.2791	
7	1.0497	
8	0.2778	
9	0.4283	
10	-0.1421	
11	-0.5172	
12	-0.0928	
13	-0.5640	
14	-0.5146	
15	-0.5672	
16	-0.5470	
17	-0.5362	
18	-0.6341	
19	6.3431e-04	
20	-0.1728	
21	0.0201	
22	-0.0215	
23	0.1653	
24	0.7046	
25	0.8647	
26	0.6270	

PART 1 – Generation of tissue kinetic classes

Post-processing and QC

When the normalised TACs are extracted from each one of the healthy controls used to generate the tissue kinetic classes, it is possible to combine them and generate the final kinetic classes for the tracer under investigation.

INSTRUCTIONS:

- Copy normalised TACs into spreadsheet (e.g. Microsoft Excel)
- Check the shape of normalised TACs, investigate any outliers
- Calculate the median normalised TACs to use as kinetic classes
- Copy time and median normalised TACs to text editor (ex. Sublime Text) and save as .csv file for input to **superX_refExtraction.m**.
- Make sure the final file (e.g. *tissueClassesForSupervisedRef.csv*) includes numbers only, no headers, and that the column order follows the default one (i.e. normGrayTAC, normWhiteTAC, normIDIF, normHighBindTAC)

Extract kinetic tissue classes: Post-processing and QC

Step by step

Copy normalised TACs into spreadsheet (examples shown with Excel)

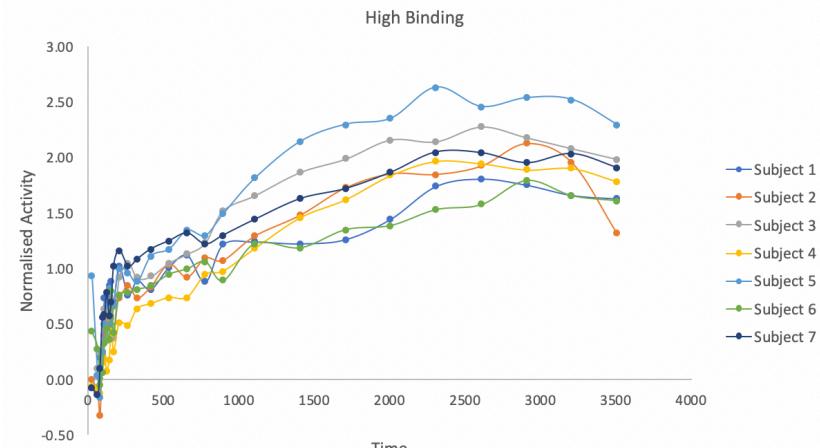
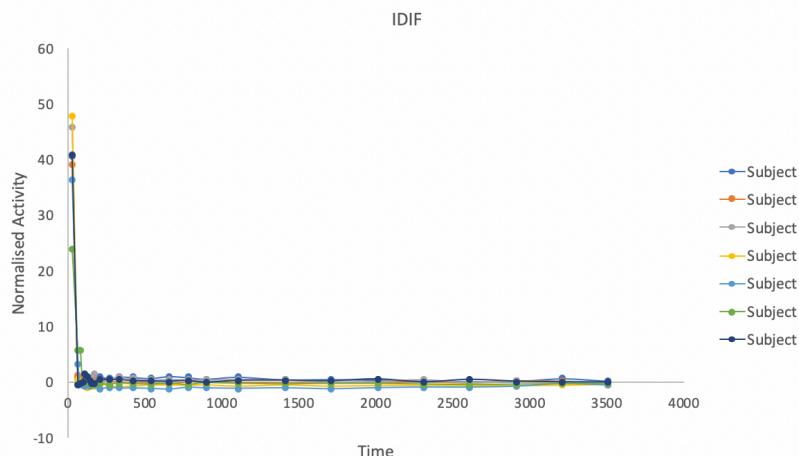
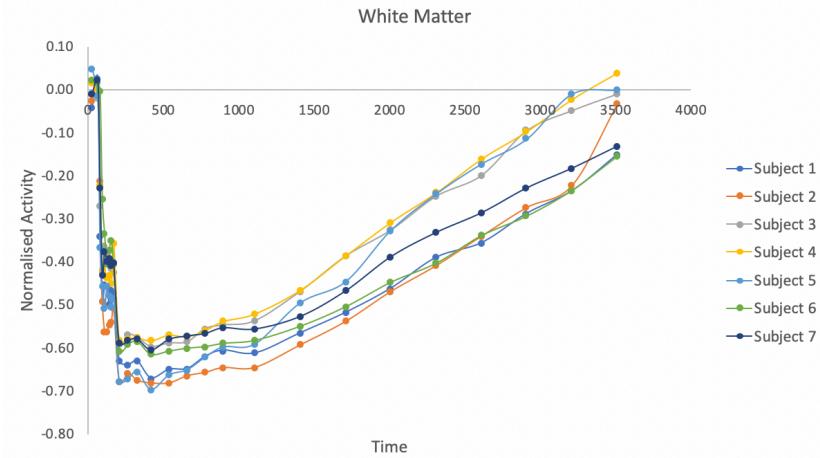
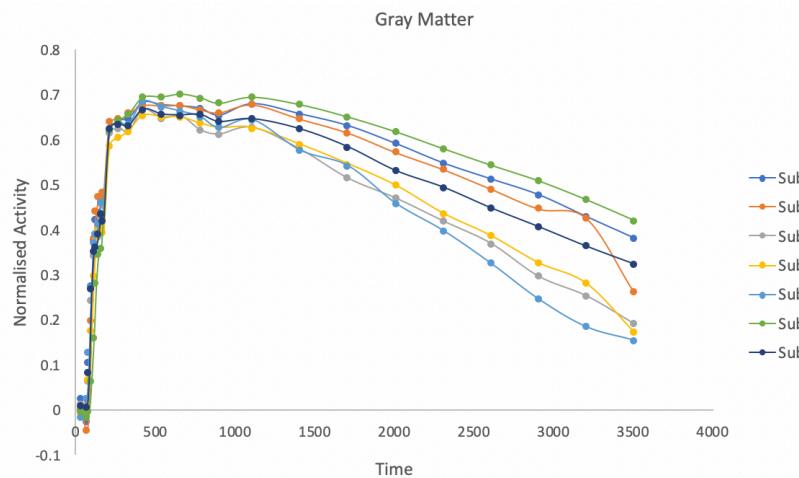
	A	B	C	D	E	F	G
1	White Matter						
2	Subject 1	Subject 2	Subject 3	Subject 4	Subject 5	Subject 6	Subject 7
3	-0.043797	-0.026048	-0.009566	0.0165246	0.0486461	0.0217729	-0.011814
4	0.024438	0.0190782	-0.020273	0.0017273	-0.014972	0.0148468	0.0217001
5	-0.342119	-0.214121	-0.270764	-0.223736	-0.367011	-0.002729	-0.228203
6	-0.491231	-0.493221	-0.426207	-0.418024	-0.456934	-0.254631	-0.430547
7	-0.454689	-0.562083	-0.365462	-0.430292	-0.508602	-0.334629	-0.37607
8	-0.456877	-0.563997	-0.384825	-0.439178	-0.457783	-0.40572	-0.39862
9	-0.497641	-0.54611	-0.37388	-0.430476	-0.480438	-0.372771	-0.392786
10	-0.46776	-0.540783	-0.416885	-0.451612	-0.506336	-0.352454	-0.408138
11	-0.472515	-0.509642	-0.426424	-0.35667	-0.484926	-0.404571	-0.404051
12	-0.632032	-0.678456	-0.59382	-0.583455	-0.679753	-0.608184	-0.590298
13	-0.640099	-0.659673	-0.570199	-0.578123	-0.672547	-0.591762	-0.583035
14	-0.632487	-0.675878	-0.576304	-0.576141	-0.655993	-0.585327	-0.579834
15	-0.671112	-0.681975	-0.597633	-0.582954	-0.698348	-0.61545	-0.604962
16	-0.651411	-0.681941	-0.589633	-0.570001	-0.664474	-0.607867	-0.580416
17	-0.649529	-0.66527	-0.586793	-0.573695	-0.652242	-0.601244	-0.572554
18	-0.6209	-0.657399	-0.555882	-0.558855	-0.622782	-0.597348	-0.566726
19	-0.606924	-0.646244	-0.547886	-0.537989	-0.599155	-0.59003	-0.553802
20	-0.613183	-0.646508	-0.53737	-0.520355	-0.591976	-0.582828	-0.556375
21	-0.566389	-0.593165	-0.468978	-0.465611	-0.496286	-0.550028	-0.52711
22	-0.51883	-0.538508	-0.387112	-0.386126	-0.446873	-0.504847	-0.467835
23	-0.462402	-0.468828	-0.326358	-0.308169	-0.327569	-0.447896	-0.388732
24	-0.39104	-0.408679	-0.247927	-0.239801	-0.242464	-0.403549	-0.332521
25	-0.356918	-0.340152	-0.19926	-0.161325	-0.172891	-0.339558	-0.286853
26	-0.288714	-0.273532	-0.095371	-0.097571	-0.114722	-0.294225	-0.229333
27	-0.235906	-0.222062	-0.048508	-0.023112	-0.010095	-0.234193	-0.183519
28	-0.151449	-0.032733	-0.009852	0.0383558	-0.000636	-0.155339	-0.13251
29							
30	High Binding						
31	-0.075865	-0.0057661	-0.064838	-0.061368	0.9368241	0.4354282	-0.067016
32	-0.068584	-0.106016	0.0982566	-0.071332	0.0470216	0.2818305	-0.134368
33	0.1979963	-0.321883	0.0202804	-0.114942	-0.158393	-0.048439	0.1075707
34	0.2495334	0.2421173	0.2293307	0.1566126	0.259213	0.0568738	0.5680204
35	0.7354738	0.4009215	0.6349112	0.1890723	0.5062884	0.3286651	0.5883339
36	0.7575932	0.5370474	0.3525928	0.0823014	0.6785083	0.4641524	0.793467
37	0.8484135	0.1714548	0.6417007	0.175026	0.8300458	0.3626408	0.5811561
38	0.8828373	0.5738843	0.5566352	0.4635527	0.5075217	0.7953991	0.7062488
39	0.6954732	0.663506	0.3759407	0.2562222	0.674419	0.4327139	1.0299987
40	1.0248312	0.7345249	0.9251493	0.5101782	0.9975793	0.7668605	1.1670893
41	0.7654859	0.8559919	1.0561067	0.4941816	0.9658977	0.7879528	1.021174
42	0.8977917	0.7393442	0.920211	0.6403344	0.8823756	0.8184038	1.0872093
43	0.818211	0.8424809	0.933529	0.6856814	1.1163049	0.8496611	1.1771064
44	1.0167083	0.10440379	0.10549376	0.37937534	1.1799022	0.9533117	1.2508904
45	1.1240627	0.9284446	1.1346113	0.7403431	1.3464176	1.0011311	1.3197876
46	0.8826055	1.0982068	1.2349777	0.9494884	1.3022891	1.0665653	1.2212612
47	1.2224107	1.080803	1.5275427	0.9803505	1.49725	0.8959914	1.3020889
48	1.2441927	1.2991511	1.6631091	1.1843289	1.8184048	1.2304117	1.4442212
49	1.2286659	1.4866077	1.87188	1.4605343	2.1472306	1.1918991	1.6300239
50	1.2666789	1.7322241	1.992107	1.6231075	2.3013271	1.3541085	1.7212097
51	1.4507184	1.8583501	2.1646249	1.841028	2.3558685	1.3926624	1.8682659
52	1.7458856	1.8532115	2.1486791	1.9689084	2.6350987	1.5347254	2.0507073
53	1.8125542	1.9285805	2.2814567	1.9740524	2.4614487	1.5799955	2.0433453
54	1.7572895	2.1313147	2.1846085	1.891679	2.5399302	1.7970393	2.9537476
55	1.6652955	1.9591345	2.0864748	1.9044693	2.5237689	1.6603195	2.035754
56	1.6331709	1.318818	1.9897125	1.7862945	2.2995518	1.6147746	1.9098394
57							

	A	B	C	D	E	F	G
58	Gray Matter						
59	0.0269417	0.0052137	0.0142718	-0.004813	-0.016051	-0.001711	0.0097415
60	-0.025294	-0.043205	-0.009492	-0.007924	0.0280084	-0.015723	0.0087293
61	0.1059337	0.0212017	0.0662779	0.0693659	0.1288999	-0.003267	0.0826982
62	0.2767004	0.198063	0.2450832	0.1761792	0.2718333	0.0662723	0.2687109
63	0.3777326	0.3807195	0.3424416	0.298121	0.372613	0.1618628	0.3520452
64	0.4237469	0.4415494	0.3930522	0.3759944	0.3919283	0.2834855	0.3636479
65	0.4379321	0.4743905	0.3864022	0.4016075	0.4153912	0.3470403	0.3921893
66	0.4620634	0.471713	0.426677	0.4104903	0.4622577	0.3593483	0.4358272
67	0.4627741	0.4846159	0.4552881	0.3934914	0.4632939	0.4375514	0.4190937
68	0.6205225	0.6398068	0.614687	0.5867878	0.6220362	0.6230967	0.6258487
69	0.6432289	0.6463461	0.6257269	0.6044946	0.6393896	0.6441275	0.6343329
70	0.6480623	0.65889736	0.6183474	0.6199125	0.6354758	0.6579685	0.631438
71	0.6837558	0.6776969	0.6672821	0.6537749	0.686117	0.694782	0.6677058
72	0.6780577	0.676804	0.6492427	0.6507216	0.674362	0.6960513	0.6565886
73	0.6753753	0.6773716	0.652908	0.649501	0.6642386	0.7018848	0.6550199
74	0.67035	0.6678876	0.6223119	0.6365407	0.6524346	0.6939709	0.655578
75	0.6550293	0.6616503	0.6132019	0.6270356	0.6281295	0.6817541	0.6402353
76	0.6803034	0.6786224	0.6276443	0.6267277	0.6453422	0.6950083	0.6462208
77	0.6576333	0.6471232	0.57903	0.5894265	0.5774656	0.6782345	0.6236679
78	0.6314671	0.6162357	0.5148959	0.5465536	0.5429908	0.650989	0.5840911
79	0.5918811	0.5731557	0.4708996	0.4989185	0.4597695	0.6181702	0.5317616
80	0.5485915	0.5352083	0.419922	0.436822	0.398859	0.5805709	0.49484865
81	0.5131269	0.4916838	0.3701444	0.3871942	0.3260338	0.5440436	0.4491203
82	0.4780209	0.4479788	0.2974034	0.3273095	0.2466095	0.5090356	0.4074691
83	0.4295831	0.4266565	0.2540536	0.2815986	0.1851431	0.4672867	0.3644581
84	0.3820998	0.2623576	0.192672	0.1738703	0.1553364	0.4216018	0.3251094
85							
86	IDIF						
87	40.685695	39.129195	45.920365	47.955825	36.531047	23.88702	40.994746
88	0.9300584	1.2367198	-0.294266	0.484381	3.4271803	5.6959665	-0.319741
89	-0.32746	0.695452	0.0790962	-0.411103	-0.480832	5.7281121	-0.277211
90	0.9034667	0.8325146	0.1824783	-0.223161	-0.496277	-0.264587	0.1531716
91	0.8344187	0.099906	0.7652991	0.2917687	-0.581773	0.0185605	1.4558792
92	-0.046711	0.4356364	0.10533665	-1.02728	-0.392924	0.2390088	1.0068063
93	0.8661091	0.1584863	0.8451378	-0.509536	-0.558968	-0.2407278	0.2235237
94	0.6764426	0.2764716	0.2720327	-0.231048	-0.440554	-0.467954	-0.086524
95	1.387482	0.239006	1.4583024	-0.784038	-0.321187	-0.107825	-0.150117
96	1.1675312	0.4981386	0.2424525	0.379999	-1.134994	-0.174017	0.4720251
97	0.7785901	0.3461232	0.6488153	-0.472404	-0.842758	-0.358474	0.4445351
98	1.0662521	1.173622	0.9843838	-1.79378	-0.128743	-0.14423	0.4921713
99	0.9938724	0.0540088	0.7535766	-0.623913	-1.006907	-0.267188	0.2801218
00	0.8149112	-0.05617	0.5257531	-0.416826	-1.076171	-0.336393	0.1987327
01	1.1503417	-0.198114	0.6498035	-0.411854	-1.193837	-0.192976	0.1753421
02	0.9608051	0.139035	0.5755656	-0.460565	-0.885597	-0.404532	0.2935834
03	0.6864341	0.2065945	0.5768762	-0.402481	-0.968227	-0.087356	-0.002822
04	1.0248608	0.0840713	0.6161286	-0.624247	-1.059597	-0.201166	0.3354283
05	0.5763306	0.0251441	0.5553264	-0.427904	-0.97625	-0.299554	0.3656905
06	0.6056433	-0.076171	0.2504956	-0.625852	-1.140258	-0.211306	0.277689
07	0.5135966	-0.020178	0.3126389	-0.513719	-0.956089	-0.222534	0.5801475
08	0.3425217	0.1745227	0.6034928	-0.468041	-0.841093	-0.321697	0.0663471
09	0.2441232	-0.9505	0.1010285	-0.51295	-0.849619	-0.412716	0.4945981
10	0.2656292	0.3286665	0.2717752	-0.539866	-0.682384	-0.347764	0.1882622
11	0.752482	0.2377182	0.1808765	-0.443561	-0.22156	-0.258467	0.066582

Extract kinetic tissue classes: Post-processing and QC

Step by step

Check shape of normalised TACs, investigate any outliers



Extract kinetic tissue classes: Post-processing and QC

Step by step

Calculate median normalised TACs to use as kinetic classes

Subject 1	Subject 2	Subject 3	Subject 4	Subject 5	Subject 6	Subject 7	Median
-0.043797	-0.026048	-0.009566	0.0165246	0.0486461	0.0217729	-0.011814	=Median(B3:H3)
0.024438	0.0190782	-0.020273	0.0017273	-0.014972	0.0148468	0.0217001	

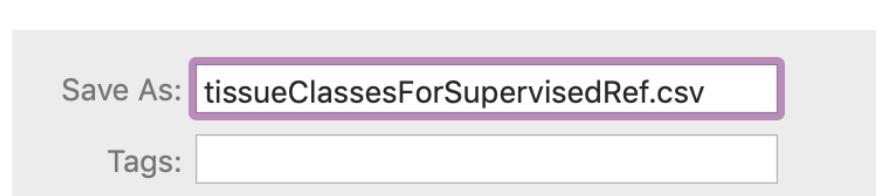
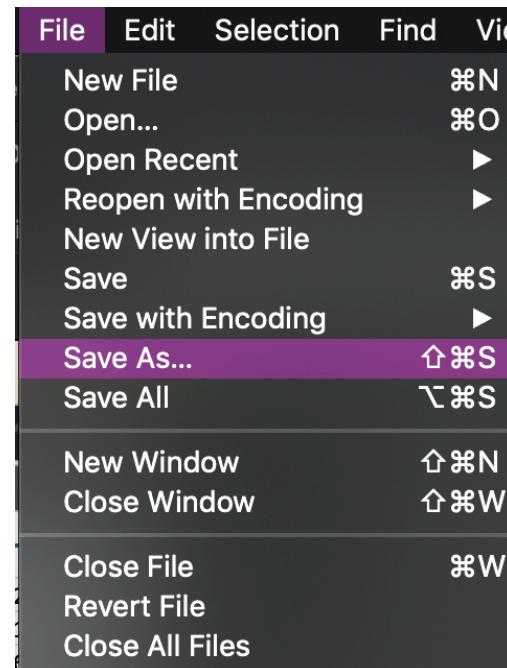
Subject 1	Subject 2	Subject 3	Subject 4	Subject 5	Subject 6	Subject 7	Median
-0.043797	-0.026048	-0.009566	0.0165246	0.0486461	0.0217729	-0.011814	-0.009566
0.024438	0.0190782	-0.020273	0.0017273	-0.014972	0.0148468	0.0217001	0.0148468
-0.342119	-0.214121	-0.270764	-0.22376	-0.367011	-0.002729	-0.228203	-0.228203
-0.491231	-0.493221	-0.426207	-0.418024	-0.456934	-0.254631	-0.430547	-0.430547
-0.454689	-0.562083	-0.365462	-0.430292	-0.508602	-0.334629	-0.37607	-0.430292
-0.456877	-0.563997	-0.384825	-0.439178	-0.457783	-0.40572	-0.39862	-0.439178
-0.497641	-0.54611	-0.37388	-0.430476	-0.480438	-0.372771	-0.392786	-0.430476
-0.46776	-0.540783	-0.416885	-0.451612	-0.506336	-0.352454	-0.408138	-0.451612
-0.472515	-0.509642	-0.426424	-0.35667	-0.484926	-0.404571	-0.404051	-0.426424
-0.632032	-0.678456	-0.59382	-0.583455	-0.679753	-0.608184	-0.590298	-0.608184
-0.640099	-0.659673	-0.570199	-0.578123	-0.672547	-0.591762	-0.583035	-0.591762
-0.632487	-0.675878	-0.576304	-0.576141	-0.655993	-0.585327	-0.579834	-0.585327
-0.671112	-0.681975	-0.597633	-0.582954	-0.698348	-0.61545	-0.604962	-0.61545
-0.651411	-0.681941	-0.589633	-0.57001	-0.664474	-0.607867	-0.580416	-0.607867
-0.649529	-0.66527	-0.586793	-0.573695	-0.652242	-0.601244	-0.572554	-0.601244
-0.6209	-0.657399	-0.555882	-0.558855	-0.622782	-0.597348	-0.566726	-0.597348
-0.606924	-0.646244	-0.547886	-0.537989	-0.599155	-0.59003	-0.553802	-0.59003
-0.613183	-0.645608	-0.53737	-0.520355	-0.591976	-0.582828	-0.556375	-0.582828
-0.566389	-0.593165	-0.468978	-0.465611	-0.496286	-0.550028	-0.52711	-0.52711
-0.51883	-0.538508	-0.387112	-0.386126	-0.446873	-0.504847	-0.467835	-0.467835
-0.462402	-0.468828	-0.326358	-0.308169	-0.327569	-0.447896	-0.388732	-0.388732
-0.39104	-0.408679	-0.247927	-0.239801	-0.24264	-0.403549	-0.332521	-0.332521
-0.356918	-0.340152	-0.19926	-0.161325	-0.172891	-0.339538	-0.286853	-0.286853
-0.288714	-0.273532	-0.095371	-0.097571	-0.114722	-0.294225	-0.229333	-0.229333
-0.235906	-0.222062	-0.048508	-0.023112	-0.010095	-0.234193	-0.183519	-0.183519
-0.151449	-0.032733	-0.009852	0.0383558	-0.000636	-0.155339	-0.13251	-0.032733

Extract kinetic tissue classes: Post-processing and QC

Step by step

Copy time and median normalised TACs to text editor (ex. Sublime Text) and save as .csv file for input to superX_refExtraction.m (include numbers only, no headers)

1	0	0	0	0	0
2	30	-0.004582126	1.7518357	0.035228315	0.198516617
3	67.5	0.000808174	2.340508787	0.015479132	0.166424023
4	82.5	0.044076598	3.915327885	-0.109787447	0.131591296
5	97.5	0.137005025	3.191793414	-0.263130267	0.22406747
6	112.5	0.200190332	1.89055942	-0.262720142	0.359391883
7	127.5	0.240943484	0.999664187	-0.27334293	0.388853879
8	142.5	0.248640415	0.648784286	-0.284935325	0.434767515
9	157.5	0.273047785	0.434044577	-0.284348217	0.481113621
10	172.5	0.281788833	0.338190907	-0.289656256	0.469251687
11	210	0.490902899	0.470997139	-0.508203538	0.756309242
12	270	0.508409508	0.029633732	-0.510123085	0.781172987
13	330	0.511427397	0.009261626	-0.485578379	0.756245568
14	420	0.591305895	-0.25122628	-0.546229935	0.869647106
15	540	0.581500977	-0.166638079	-0.528153569	0.919543391
16	660	0.573917805	-0.438504286	-0.503460947	1.015185125
17	780	0.558083275	-0.284180688	-0.489592357	1.084206778
18	900	0.546464203	-0.377196991	-0.460516006	1.171197355
19	1110	0.580696674	-0.237903736	-0.487592247	1.417688001
20	1410	0.53376023	-0.279010515	-0.417362764	1.498264708
21	1710	0.490233066	-0.217494617	-0.358893756	1.597540686
22	2010	0.448313718	-0.068072602	-0.294762596	1.721857155
23	2310	0.400141979	-0.01296629	-0.219503992	1.692795046
24	2610	0.347925931	-0.171584264	-0.162110276	1.715687193
25	2910	0.296886491	0.001972575	-0.099577861	1.677772233
26	3210	0.250408795	0.242393793	-0.049227475	1.669815724
27	3510	0.207903615	0.319790021	-0.004906596	1.631756106



PART 2 – Reference Region Extraction

INSTRUCTIONS:

- Select both dynamic PET and structural MRI images for the subject to which the supervised clustering method will be applied
- Run the function **superX_refExtraction.m** and extract the reference region TAC and the reference region mask

NOTES:

- The pipeline works only if the PET images are acquired with the same PET protocol (e.g. same tracer, injection type, length of acquisition, framing, etc)
- The sample used to generate the kinetic classes should be independent from one to which the supervised clustering will be applied
- The same criteria used to generate the *brainMask* for the definition of tissue kinetic classes must be applied for the reference region extraction.

PART 2 – Reference Region Extraction

Inputs for `superX_refExtraction.m`:

- dynPET: 4D matrix containing dynamic PET data
- kineticClasses: string with path to .csv file containing the kinetic tissue classes.
Columns of .csv file should include mid-frame times, gray matter class, white matter, blood class, high binding gray matter class in that order!!!
- brainMask: 3D matrix containing binary data specifying brain voxels
- refMaskExtraction: 3D matrix containing binary data specifying prospective voxels to use for the selection of the reference region (ex: cerebellar or cortical gray matter)
- time: 2D double with PET mid-frame times (in minutes)

Notes:

- The first three dimensions of dynPET must match dimensions of brain and tissue masks
- The length of time vector must match with number of frames in dynPET.

PART 2 – Reference Region Extraction

Representative outputs

The function returns the supervised reference region TAC (RefTAC) interpolated into the PET mid-frame time and the 3D matrix containing a binary mask with the voxels corresponding to the supervised reference region (RefMask)

RefTAC	
	26x1 double
1	1
1	0.0105
2	0.5443
3	3.4109
4	4.4971
5	4.8110
6	5.0187
7	5.0000
8	5.0841
9	5.2069
10	5.2684
11	5.2383
12	5.1371
13	4.9247
14	4.7297
15	4.4947
16	4.2951
17	4.1434
18	3.8141
19	3.4650
20	3.1420
21	2.8557
22	2.5725
23	2.3432
24	2.1749
25	2.0201
26	1.8834
27	



RefTAC
26x1 double

RefMask
91x109x91 double

PART 2 – Reference Region Extraction

Post-processing and QC

Visualise supervised reference region TAC and compared them with other ROI TACs. Hopefully it should be the one with the fastest wash-out

