

# Tutorial 12: Using SAM through ArrayTrack

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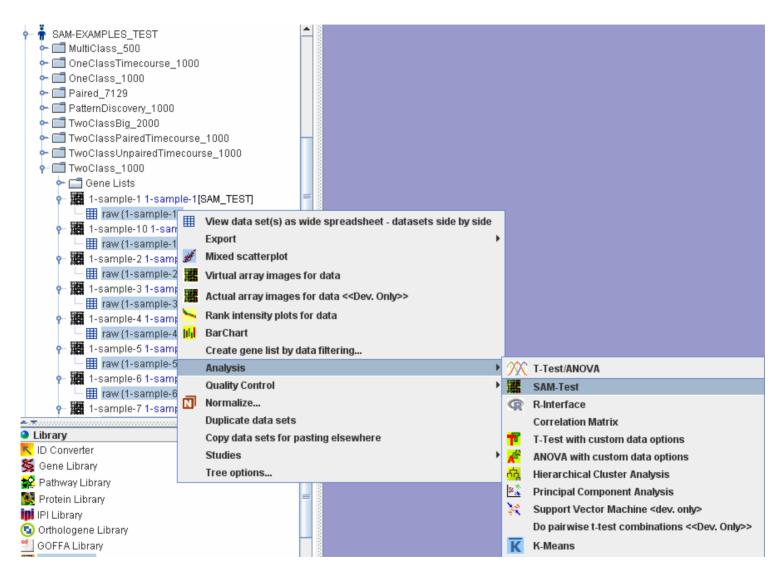
#### SAM-Test

SAM (Significance Analysis of Microarrays) is an analysis tool for Identifying statistical significant genes in a set of microarray experiments. <u>http://www-stat.stanford.edu/~tibs/SAM/</u>

Before using SAM-test tool in ArrayTrack, users should read SAM manual <a href="http://www-stat.stanford.edu/~tibs/SAM/sam.pdf">http://www-stat.stanford.edu/~tibs/SAM/sam.pdf</a>

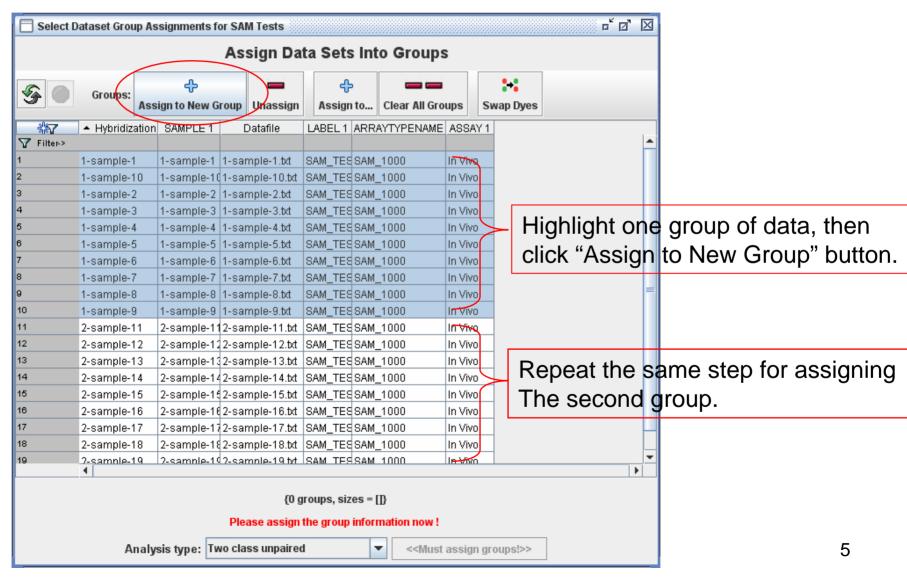
The SAM tool in ArrayTrack includes following analysis types:

- •Two class paired
- •Two class unpaired
- •One class
- •Multi class
- Survival
- •One class timecourse
- •Two class unpaired timecourse
- •Two class paired timecourse



Select all the data, right-click, choose "Analysis -> SAM-Test"

Select [	Dataset Group	Assignments for	SAM Tests				<b>e</b> <sup>k</sup>	d' 🛛
			Assign Dat	a Sets In	to Groups			
<b>%</b>	Groups: A	수 ssign to New Gro	up Unassign	႕ Assign to	Clear All Groups	<b>;+;</b> Swap Dyes		
₩7 7 Filter->	▲ Hybridizati	on SAMPLE 1	Datafile	LABEL 1	ARRAYTYPENAME	ASSAY 1		<b>^</b>
1 2	1-sample-1 1-sample-10	1-sample-1 1-sample-10	1-sample-1.txt 1-sample-10.txt	SAM_TEST SAM_TEST	SAM_1000 SAM_1000	In Vivo In Vivo		
3	1-sample-2 1-sample-3	1-sample-2 1-sample-3	1-sample-2.txt 1-sample-3.txt	SAM_TEST		In Vivo In Vivo		
5	1-sample-4	1-sample-4	1-sample-4.txt	SAM_TEST	SAM_1000	In Vivo		
6 7	1-sample-5 1-sample-6	1-sample-5 1-sample-6	1-sample-5.txt 1-sample-6.txt	SAM_TEST SAM_TEST	SAM_1000 SAM_1000	In Vivo In Vivo		
8 9	1-sample-7 1-sample-8	1-sample-7 1-sample-8	1-sample-7.txt 1-sample-8.txt	SAM_TEST SAM_TEST	SAM_1000 SAM_1000	In Vivo In Vivo		_
10 11	1-sample-9 2-sample-11	1-sample-9 2-sample-11	1-sample-9.txt 2-sample-11.txt	SAM_TEST SAM_TEST	SAM_1000 SAM_1000	In Vivo In Vivo		
12 13	2-sample-12 2-sample-13	2-sample-12 2-sample-13	2-sample-12.txt 2-sample-13.txt		SAM_1000 SAM_1000	In Vivo In Vivo		
14 15	2-sample-14 2-sample-15	2-sample-14 2-sample-15	2-sample-14.txt 2-sample-15.txt		SAM_1000 SAM_1000	In Vivo In Vivo		
16 17	2-sample-16 2-sample-17	2-sample-16 2-sample- <mark>Non</mark>	2-sample-16.txt		SAM_1000	In Vivo In Vivo		
18 19	2-sample-18 2-sample-19	2-sample-Two	o class unpaired		AM_1000 AM_1000	In Vivo In Vivo		
20	A Multiclass					In Your		
		Qua	o class paired antitative				F	irst s
		One	vival e class timecours	(	elected firstly			
	An	alysis type: Nor	le			gn groups!>>		



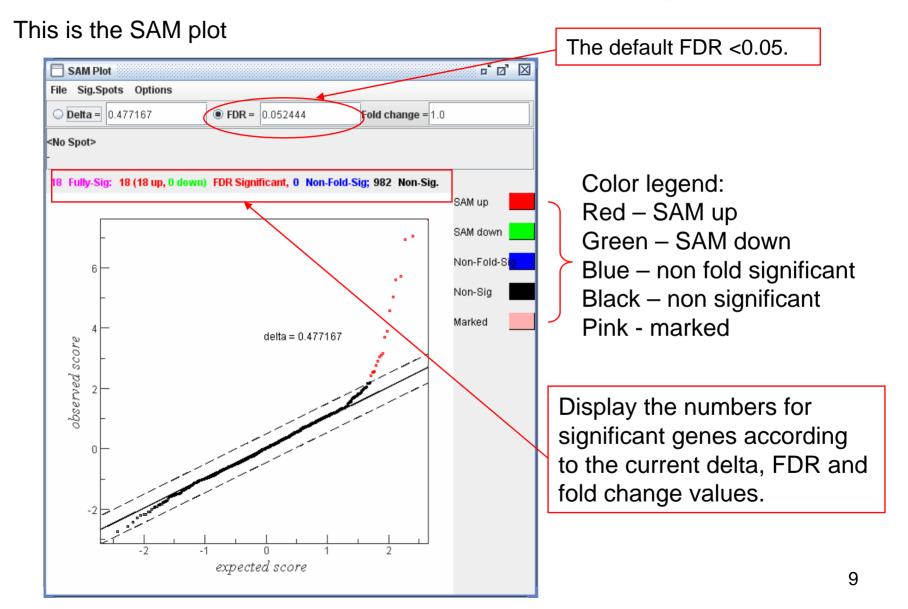
#### After assign data sets into two groups, click "Next" button.

Select [	Dataset Group /	Assignments for SAM	A Tests				r 0 ×	1
		As	sign Data	a Sets Ir	to Groups			
<b>5</b>	Groups: As	දා ssign to New Group	unassign	🕂 Assign to	Clear All Groups	;+; Swap Dyes	5	
Filter->	<ul> <li>Hybridizatio</li> </ul>	n SAMPLE 1	Datafile	LABEL	1 ARRAYTYPENAME	ASSAY 1		
1								<u> </u>
<b>6</b>	1-sample-1	1-sample-1	1-sample-1.	txt SAM_T	ESSAM_1000	In Vivo		
<u>6</u>	1-sample-10	1-sample-10	1-sample-10	0.txt SAM_T	ESSAM_1000	In Vivo		
ด้	1-sample-2	1-sample-2	1-sample-2.	txt SAM_T	ESSAM_1000	In Vivo		
ň –	1-sample-3	1-sample-3	1-sample-3.	txt SAM_T	ESSAM_1000	In Vivo		
ŏ –	1-sample-4	1-sample-4	1-sample-4.	txt SAM_T	ESSAM_1000	In Vivo		
<u>6</u>	1-sample-5	1-sample-5	1-sample-5.	txt SAM_T	ESSAM_1000	In Vivo		
<b>6</b>	1-sample-6	1-sample-6	1-sample-6.	txt SAM_T	ESSAM_1000	In Vivo		
<b>7</b>	1-sample-7	1-sample-7	1-sample-7.	txt SAM_T	ESSAM_1000	In Vivo		
1	1-sample-8	1-sample-8	1-sample-8.	txt SAM_T	ESSAM_1000	In Vivo	_	=
2	1-sample-9	1-sample-9	1-sample-9.	txt SAM_T	ESSAM_1000	In Vivo		-
	2-sample-11	2-sample-11	2-sample-11	I.txt SAM_T	ESSAM_1000	In Vivo		
0	2-sample-12	2-sample-12	2-sample-12	2.txt SAM_T	ESSAM_1000	In Vivo		
2	2-sample-13	2-sample-13	2-sample-13	3.txt SAM_T	ESSAM_1000	In Vivo		
2	2-sample-14	2-sample-14	2-sample-14	4.txt SAM_T	ESSAM_1000	In Vivo		
8	2-sample-15	2-sample-15	2-sample-16	5.txt SAM_T	ESSAM_1000	In Vivo		
8	2-sample-16	2-sample-16	2-sample-16	6.txt SAM_T	ESSAM_1000	In Vivo		
8	2-sample-17	2-sample-17	2-sample-17	7.txt SAM_T	ESSAM_1000	In Vivo		
	2-sample-18	2-sample-18	2-sample-18	3.txt SAM_T	ESSAM_1000	In Vivo		
8	2-sample-19	2-sample-19	2-sample-19	9.txt SAM_T	ESSAM_1000	In Vivo	-	
	2 comple 20	2 comple 20	2 compto 20	DAA COM T	Ed.0.0M 4000	Lo Mino		
					ssigned to 2 group			
			2 group	s, sizes = [1	0, 10]		The size of eac	h group is presente
		Before click 'I	next >', Please	e check the	group assignment a	jain!	in parenthesis.	• • •
		Analysis type	: Two class	unpaired	▼ Nex	t>)		

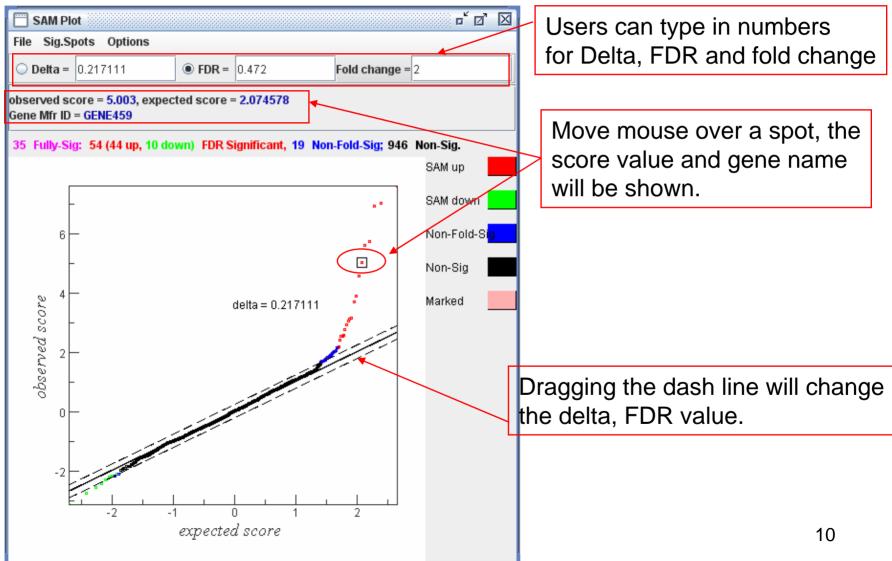
wo class unpaired probl	Assignments for SAM Tests	
	Significance Analysis of I	Microarrays (SAM)
AM-Tests Options		
est statistics:	◉ T-statistic 🛛 Wilcoxon	These values are de
lumber of permutations:	Limit to(100~1000): 100	
stimation of s0 factor:	Automatic      Used fixed percent	entile(%) 50
mputation engine:	Number of neighbors of KNN 10	
tandom number seed:	1234567 Ger	nerate Random Seed
alculate the fold change	's effect on FDR (may take a while)?	® No ○ Yes
iltering with a gene list— Only include genes from (	gene list <all genes=""></all>	
ene identifiers to include	]	
🖌 Genbank Acc 🛛 🖌	Gene Mfr ID	UNIGENEID 🗹 GENENAME
CLONEID	GEN_DESCR_MFR 🔽 REFSEQ	₽ SPOTID
Dataset Naming ☑ add sample name(s) t	Data option	is t backgrounds(raw datasets only)
	Apply lo	g (base 2) to expression values
add dye name(s) to hy	bridization names	spots flagged as bad

If you use the sample data to compare with the SAM results in Excel, make sure "apply log..." is Unchecked.

Genb	oank Acc Gene Mfr ID	LOCUSID	GENENAME	REFSEQ	SPOTID	Observed s	. P value	Numerato	r(r) Denominat	. Fold Chang	e q
	GENE703				4661482	-0.0454	0.9194	-0.0565	1.2451	0.8345	89 🔺
	GENE704				4661483	-0.1482	0.6589	-0.1534	1.0353	0.9822	84 📃
	GENE705				4661484	0.761	0.0832	0.9029	1.1864	2.2999	45
	GENE706				4661485	-0.3265	0.4395	-0.3862	1.1827	0.7524	75
	GENE707				4661486	0.0143	0.9672	0.0151	1.0555	0.9252	92
	GENE708				4661487	0.5624	0.1804	0.651	1.1576	1.1782	86
	GENE709				4661488	0.3802	0.3629	0.4447	1.1698	1.4819	91
1	GENE710				4661489	0.1115	0.7359	0.1147	1.029	1.0886	92
I	GENE711				4661490	-0.3565	0.3319	-0.3835	1.0757	0.8565	75
0	GENE712				4661491	0.1968	0.6172	0.2226	1.1314	1.1666	91
1	GENE713				4661492	0.1229	0.7774	0.1493	1.2145	1.0299	92
2	GENE714				4661493	-2.927	0	-3.4858	1.1909	0.0855	0
3	GENE715				4661494	-0.5412	0.1842	-0.6168	1.1398	0.5658	63
4	GENE716				4661495	-0.6519	0.0836	-0.7026	1.0778	0.6399	58
5	GENE717				4661496	-0.0329	0.9406	-0.0405	1.2304	1.0624	90
6	GENE718				4661497	-0.0177	0.9649	-0.0204	1.1509	1.1047	90
7	GENE719				4661498	-0.159	0.7136	-0.1913	1.2032	0.6056	84
8	GENE720				4661499	-0.5584	0.1313	-0.5993	1.0732	0.6625	63
9	GENE721				4661500	-0.5336	0.2539	-0.6746	1.2642	0.4535	63
0	GENE722				4661501	0.3128	0.4216	0.3501	1.1193	1.3825	91
1	GENE723				4661502	-0.6569	0.1064	-0.7384	1.124	0.5101	58
2	GENE724				4661503	0.2127	0.638	0.2648	1.2449	1.5199	91
3	GENE725				4661504	0.0909	0.8039	0.0987	1.0859	1.0696	92
4	GENE726				4661505	-1.6399	0.0006	-1.7439	1.0634	0.375	0
5	GENE727				4661506	0.3239	0.353	0.3405	1.0513	1.2123	91
6	GENE728				4661507	-0.2093	0.6108	-0.2434	1.1631	0.9654	80
8	GENE729				4661508	0.329	0.4524	0.3995	1.2143	1.4442	91 92 💌
8	GENE730		l		4661509	0.0478	0.9102	0.0569	1 1 9 1 4	1 1 5 9 7	<u>u</u> 7
Significance	Filtoring				1000 ge	enes					
-	-										
Target Fa	alse Discovery Rate (Fl	DR):	O De	elta =					Click		סוס
Select #	genes by la	argest SAM a	absolute score	• 🔻					Click	SAIVI	P10
Mean Chann	el Intensities >	Bad Flags	s <=								
Abs Fold Cha											
ADST OIL CIT	ange -		г					/			
				Apply Filte	ers Clea	ar Filters					
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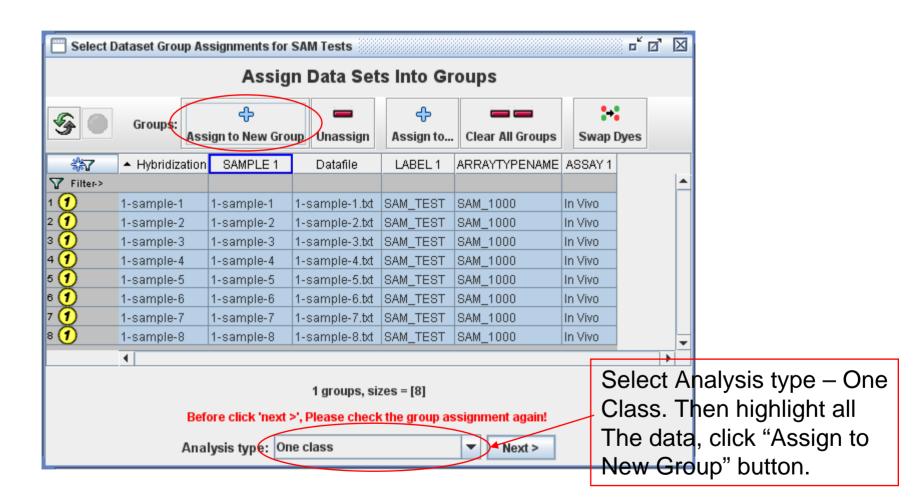


#### SAM Plot



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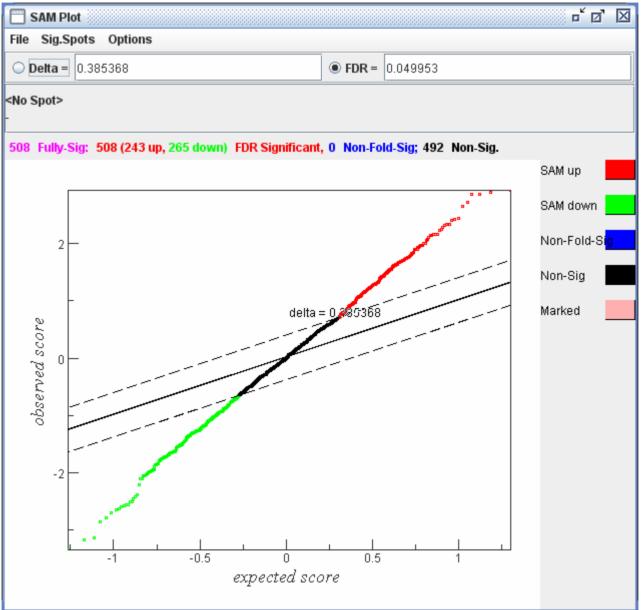
Right-click the selected data set, choose "Analysis" -> SAM-Test.



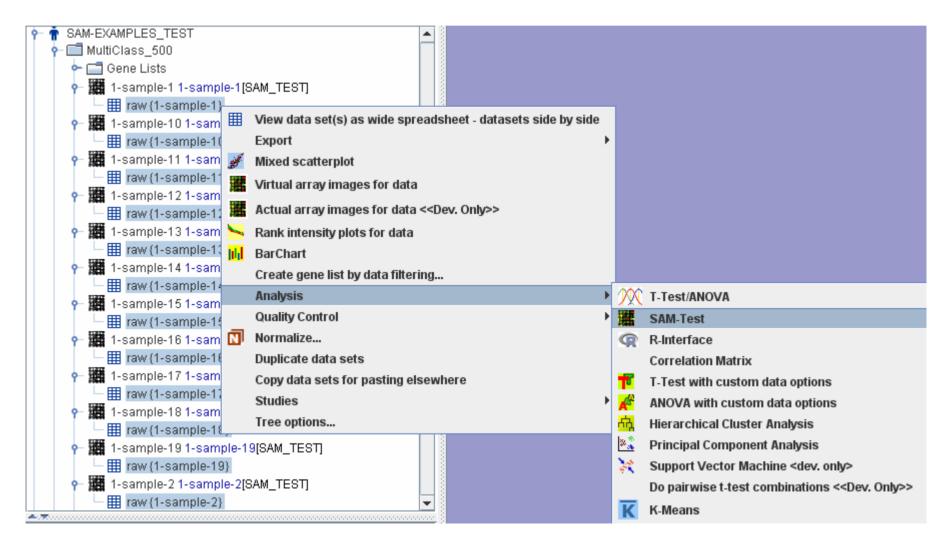
🔲 Select Dataset Group Assignments for SAM Tests	r 🛛 🖂								
One class problem									
Significance Analysis	of Microarrays (SAM)								
SAM-Tests Options									
Number of permutations: Limit to(100~1000): 100									
Estimation of s0 factor:  Automatic O Used fixed	percentile(%) 50								
Imputation engine: Number of neighbors of KNN	10								
Random number seed:     1234567       Generate Random Seed									
Calculate the fold change's effect on FDR (may take a wh	ile)? 🖲 No 🔾 Yes								
Filtering with a gene list									
Only include genes from gene list <a>all genes&gt;</a>									
Gene identifiers to include									
Construction of the second sec	ID UNIGENEID IZ GENENAME								
□ CLONEID □ GEN_DESCR_MFR   REFSEC	SPOTID								
	options								
Dataset Naming	btract backgrounds(raw datasets only)								
✓ add sample name(s) to hybridization names	ply log (base 2) to expression values								
add dye name(s) to hybridization names	clude spots flagged as bad								
< Back	Do Tests								

The value in the text boxes are default. Click "Do Tests" button.

🗂 SAM-Tests Results										
File	Selected-Spot	All-Spots								
	Gene Mfr ID	Observed score (d)	P value	Numerator(r)	Denominat	GEN	q-value(%)	REF	SPOTID	
1	GENE703	-0.3922		-0.5909	1.5068		5.9055		4661482	
2	GENE704	-0.4424	0.1608	-0.6147	1.3895		5.9055		4661483	
3	GENE705	-0.2094	0.5751	-0.2568	1.2264		10.9112		4661484	
4	GENE706	-0.468	0.3963	-0.741	1.5833		5.9055		4661485	
5	GENE707	-0.1579		-0.1847	1.17		10.9112		4661486	
6	GENE708	-0.7139	0.1181	-1.2662	1.7738		1.4605		4661487	
7	GENE709	-0.5778	0.0789	-0.8629	1.4934		2.496		4661488	
8	GENE710	-0.2387	0.6411	-0.3496	1.4643		10.334		4661489	
9	GENE711	-0.4595	0.2318	-0.7098	1.5445		5.9055		4661490	
10	GENE712	0.283	0.0852	0.3731	1.3186		14.253		4661491	
11	GENE713	-0.1933		-0.2218	1.1474		10.9112		4661492	
12	GENE714	-0.4287	0.3627	-0.6128	1.4294		5.9055		4661493	
13	GENE715	-0.5146	0.2931	-0.9513	1.8486		3.1854		4661494	
14	GENE716	-0.4829	0.1603	-0.7646	1.5834		3.1854		4661495	
15	GENE717	0.4599	0.0169	0.6108	1.3282		12.4867		4661496	
16	GENE718	-0.5346	0.3272	-1.0246	1.9166		3.1854		4661497	
17	GENE719	0.0668	0.5303	0.0971	1.454		14.253		4661498	
18	GENE720	-0.8768	0.1351	-1.6637	1.8975		0		4661499	
19	GENE721	-0.3551		-0.494	1.3915		6.6517		4661500	•
	•									
Sign	ificance Filtering			1000 ge	enes					
•	Target False Disc	covery Rate (FDR):		🔾 Delta =						
0	Select # genes	by largest	SAM absolu	te score 💌						
Mea	n Channel Intens	sities > Ba	d Flags <=							
			Apply Fil	ters Cle	ar Filters					
	Creat	te Sig. Gene List	<mark>₼</mark> д нса	PCA	K-M	leans	SAN	/I Plot		



Right-click the selected data set, choose "Analysis" ->SAM-Test

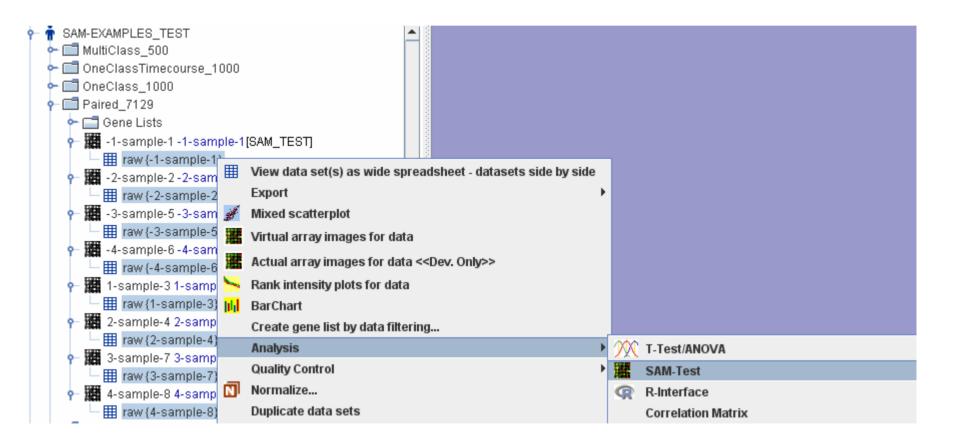


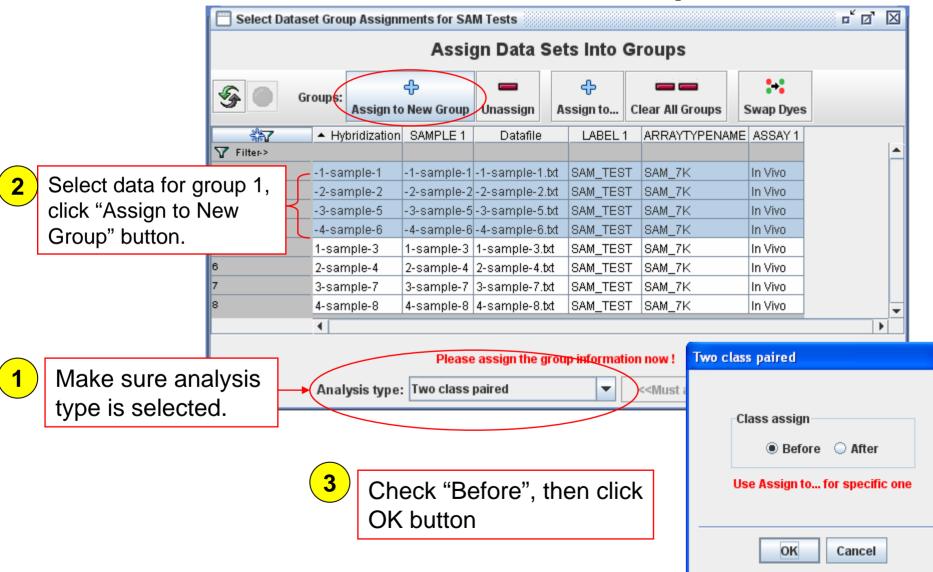
<b>S</b>			A33	yn Dala	Sets Into Gro	ups	
	Groups: As	ج) ssign to New Grou	p Unassign	수 Assign to	Clear All Groups	<b>;→;</b> Swap Dyes	
ak Tak	<ul> <li>Hybridizatio</li> </ul>	on SAMPLE 1	Datafile	LABEL 1	ARRAYTYPENAM	IE ASSAY 1	
🝸 Filter>							
1 🚺	1-sample-1	1-sample-1	1-sample-1.txt	SAM_TEST	SAM_500	In Vivo	
	1-sample-10	1-sample-10	1-sample-10.txt	SAM_TEST	SAM_500	In Vivo	
з 🧻	1-sample-11	1-sample-11	1-sample-11.txt	SAM_TEST	SAM_500	In Vivo	
4 🚺	1-sample-12	1-sample-12	1-sample-12.bt	SAM_TEST	SAM_500	In Vivo	
5 🚺	1-sample-13	1-sample-13	1-sample-13.txt	SAM_TEST	SAM_500	In Vivo	
5 1 6 2 7 2 8 2 9 2	1-sample-14	1-sample-14	1-sample-14.txt	SAM_TEST	SAM_500	In Vivo	
7 2	1-sample-15	1-sample-15	1-sample-15.bt	SAM_TEST	SAM_500	In Vivo	
8 2	1-sample-16	1-sample-16	1-sample-16.txt	SAM_TEST	SAM_500	In Vivo	
9 2	1-sample-17	1-sample-17	1-sample-17.bt	SAM_TEST	SAM_500	In Vivo	
	1-sample-18	1-sample-18	1-sample-18.txt	SAM_TEST	SAM_500	In Vivo	
	1-sample-19	1-sample-19	1-sample-19.txt	SAM_TEST	SAM_500	In Vivo	
12 3	1-sample-2	1-sample-2	1-sample-2.txt	SAM_TEST	SAM_500	In Vivo	
13 3	1-sample-20	1-sample-20	1-sample-20.txt	SAM_TEST	SAM_500	In Vivo	
14 3	1-sample-3	1-sample-3	1-sample-3.txt	SAM_TEST	SAM_500	In Vivo	
15 3	1-sample-4	1-sample-4	1-sample-4.txt	SAM_TEST	SAM_500	In Vivo	
16 3	1-sample-5	1-sample-5	1-sample-5.txt	SAM_TEST	SAM_500	In Vivo	
17 4	1-sample-6	1-sample-6	1-sample-6.txt	SAM_TEST	SAM_500	In Vivo	
18 4	1-sample-7	1-sample-7	1-sample-7.txt	SAM_TEST	SAM_500	In Vivo	
19 4	1-sample-8	1-sample-8	1-sample-8.txt	SAM_TEST	SAM_500	In Vivo	Choose the analysis type
20 4	1-sample-9	1-sample-9	1-sample-9.txt	SAM_TEST	SAM_500	In Vivo	
21 4	2-sample-21	2-sample-21	2-sample-21.txt	SAM_TEST	SAM_500	In Vivo	first, then assign the data
22 (5)	2-sample-22	2-sample-22	2-sample-22.txt	SAM_TEST	SAM_500	In Vivo	_
23 (5)	2-sample-23	2-sample-23	2-sample-23.txt	SAM_TEST	SAM_500	In Vivo	to multiple groups.
24 5	2-sample-24	2-sample-24	2-sample-24.txt	SAM_TEST	SAM_500	In Vivo	
	2-sample-25	2-sample-25	2-sample-25.txt	SAM_TEST	SAM_500	In Vivo	
26 (5)	2-sample-26	2-sample-26	2-sample-26.txt	SAM_TEST	SAM_500	In Vivo	
	•						
				5 groups, si	zes = [5, 6, 5, 5, 5]		
			Before click 'ne:	d >', Please (	check the group ass	igpment agai	<sup>in!</sup> 17

🔲 Select Dataset Group A	ssignments for SAM Tests 🛛 🖉 🛛											
Multiclass problem												
	Significance Analysis of Microarrays (SAM)											
SAM-Tests Options												
Number of permutations:												
Estimation of s0 factor:	on of s0 factor:  Automatic Used fixed percentile(%) 50 These va											
Imputation engine:	Number of neighbors of KNN 10											
Random number seed:												
Calculate the fold change'	s effect on FDR (may take a while)? <ul> <li>No</li> <li>Yes</li> </ul>											
Filtering with a gene list												
Only include genes from g	ene list <all genes=""></all>											
-Gene identifiers to include												
🖌 Genbank Acc 🛛 🖌	Gene Mfr ID 🔽 LOCUSID 🔄 UNIGENEID 🔽 GENENAME											
	GEN_DESCR_MFR REFSEQ SPOTID											
	Data options											
Dataset Naming	✓ Subtract backgrounds(raw datasets only)											
✓ add sample name(s) to	Apply log (base 2) to expression values											
add dye name(s) to hy	Exclude spots flagged as bad											
	< Back Do Tests											

🗂 SAM-Tests Results 🛛 🗖 🖂										
File	Selected-Spot	All-Spots								
	Gene Mfr ID	SPOTID	Observed score (d)	P value	Numerator(r)	Denominat	contrast-1	contras	31	
1	GENE199	4660478	0.0684	0.9532	0.0826	1.2088	0.6396	-0.1507		
2	GENE200	4660479	0.1082	0.6209	0.1897	1.7535	0.2063	0.5882	=	
3	GENE201	4660480	0.0632	0.9577	0.1119	1.7695	0.0768	-0.3344		
4	GENE202	4660481	0.0548		0.0973	1.7752	0.2543	0.1381		
5	GENE203	4660482	0.1619	0.1716	0.2372	1.4648	1.0482	0.0642	1	
6	GENE204	4660483	0.071	0.8664	0.1442	2.0316	-0.0939	0.1011	1	
7	GENE205	4660484	0.1747	0.1109	0.2407	1.3779	0.4285	0.4524	1	
8	GENE206	4660485	0.087	0.6966	0.1474	1.6948	-0.0446	0.045	1	
9	GENE207	4660486	0.1188	0.1882	0.1727	1.4537	0.0569	-0.0524		
10	GENE208	4660487	0.1523	0.1006	0.2093	1.3743	0.3434	-0.6046		
11	GENE209	4660488	0.105	0.4709	0.2001	1.9061	-0.4153	0.2514		
12	GENE210	4660489	0.0902	0.7682	0.1677	1.8595	-0.3386	0.0254		
13	GENE211	4660490	0.1079	0.6225	0.1816	1.6826	0.1645	0.428		
14	GENE212	4660491	0.1174	0.3474	0.2096	1.7856	0.2178	-0.7163		
15	GENE213	4660492	0.1563	0.1057	0.2266	1.4495	-0.2053	0.786		
16	GENE214	4660493	0.0683		0.1305	1.9108	0.0806	0.1939		
17	GENE215	4660494	0.2033	0.019		1.9027	0.1648	0.3296		
18	GENE216	4660495	0.1884		0.2603	1.3816	-1.5802	0.5408		
19	GENE217	4660496	0.1153	0.664	0.1646	1.4273	0.2295	0.4566	-	
	•							•		
Sign	ificance Filtering	J		500 gene	s					
•	Target False Disc	covery Rate (F	DR):	🔾 Delta =						
0 :	Select # genes	by	largest SAM absolute s	score 💌						
Mea	n Channel Intens	sities >	Bad Flags <=	]						
			Apply Filters	s Clear	Filters					
	Creat	te Sig. Gene L	ist 祝 HCA	💐 PCA	K-Mea	ns 🛛 🛃	SAM Plot			

Right-click the selected data set, choose "Analysis" ->SAM-Test.





×

	🔲 Select Datas	et Group Assign	ments for SA	M Tests				- d X				
		Assign Data Sets Into Groups										
	🚱 🔵 Gr	oup <b>s</b> : Assign to	슈 D New Group	Unassign	႕ Assign to	Clear All Groups	Swap Dyes					
		<ul> <li>Hybridization</li> </ul>	ization SAMPLE 1 Datafile LABEL 1 AF		ARRAYTYPEN	AME ASSAY 1						
	-1	-1-sample-1	-1-sample-1	-1-sample-1.t	xt SAM_TES	T SAM_7K	In Vivo					
	-2	-2-sample-2	-2-sample-2 -	-2-sample-2.t		T SAM_7K	In Vivo					
	-3	-3-sample-5	-3-sample-5	i-3-sample-5.t	xt SAM_TES	T SAM_7K	In Vivo					
		-4-sample-6	-4-sample-6	i-4-sample-6.t	xt SAM_TES	T SAM_7K	In Vivo					
1. Select data for grou	ıp 2,	1-sample-3	1-sample-3	1-sample-3.tx	t SAM_TES	T SAM_7K	In Vivo					
Click "Assign to New C	Group" H	2-sample-4	2-sample-4	2-sample-4.tx	t SAM_TES	T SAM_7K	In Vivo					
U		3-sample-7	3-sample-7	3-sample-7.tx	t SAM_TES	T SAM_7K	In Vivo					
Button.		4-sample-8	4-sample-8	4-sample-8.tx	t SAM_TES	T SAM_7K	In Vivo	-				
		•			_							
	Two class paired Before click 'next >', Please check the group assi											
		Dert		0 4551								
		Anal	ysis type: T	wo class pair	ed							
						Cla	nss assign					
							🔾 Before 🛛 🖲	After				
			heck " button	After", t	ck <sub>Use</sub>	Use Assign to for specific one						
							ОК Са	ncel				

	Select Dat	aset Grou	ıp Assignn	nents for SA	M Tests					r 0' 🛛		
		Assign Data Sets Into Groups										
	<b>G</b>	Groups:		+ New Group	unassign	As	🛟 ssign to	Clear All Groups	<b>\$</b> Swap Dyes			
		▲ Hy	bridization	SAMPLE 1	Datafile		LABEL 1	ARRAYTYPENAM	IE ASSAY1			
	-1				-1-sample-1.		SAM_TEST		In Vivo			
Data are assigned	-2		mple-2		-2-sample-2.txt		SAM_TEST		In Vivo			
Data are assigned	-3	-3-sa		-3-sample-5	-3-sample-5.	txt	SAM_TEST	E SAM_7K	In Vivo			
In pairs	-4	-4-sa	mple-6	-4-sample-6	-4-sample-6.	txt	SAM_TEST	E SAM_7K	In Vivo			
•	1	1-sam		1-sample-3	1-sample-3.t	xt	SAM_TEST	E SAM_7K	In Vivo			
	2	2-san	nple-4	2-sample-4	2-sample-4.t	xt	SAM_TEST	F SAM_7K	In Vivo			
	3	3-san	nple-7	3-sample-7	3-sample-7.t	xt	SAM_TEST	F SAM_7K	In Vivo			
	4	4-san	nple-8	4-sample-8	4-sample-8.t	xt	SAM_TEST	Γ SAM_7K	In Vivo	-		
		•										
	Before click 'next >', Please check the group assignment again!											
			Analy	/sistype: T	wo class pair	ed		▼ Next >	$\supset$			

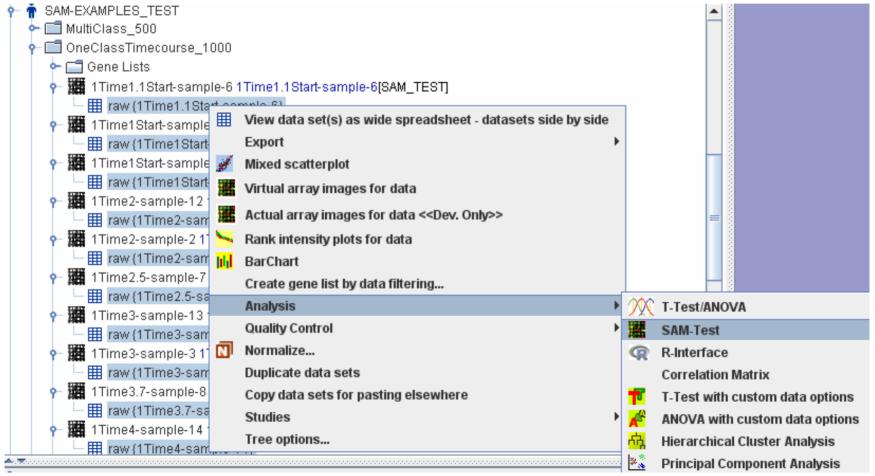
Click "Next" button.

Select Dataset Group Assignments for SAM Tests	<u>් න් න</u>	I							
Two class paired problem									
Significance Analysis of Microarrays (SAM)									
SAM-Tests Options	These valu	es are default							
Number of permutations: Limit to(100~1000): 100									
Estimation of s0 factor:   Automatic  Used fixed percentile(%) 50									
Imputation engine: Number of neighbors of KNN 10									
Random number seed: 1234567 Generate Random Seed									
Calculate the fold change's effect on FDR (may take a while)? (     No ) Yes									
Filtering with a gene list									
Only include genes from gene list <all genes=""></all>									
Gene identifiers to include									
Cenbank Acc Cene Mfr ID COUSID UNIGENEID	GENENAME								
CLONEID GEN_DESCR_MFR 🗹 REFSEQ 🗹 SPOTID									
Data options									
✓ add sample name(s) to hybridization names									
add dye name(s) to hybridization names  Apply log (base 2) to expression valu  Exclude spots flagged as bad	es								
		24							
< Back Do Tests									

<b>E</b>	SAM-Tests Results								
File	Selected-Spot	All-Spots							_
	Gene Mfr ID	SPOTID	Observed score (d)	Numerator(r)	Denominat	Fold Change	q-value(%)	localfdr	1
1	GENE870	4654020	0.4245	0.7516	1.7706	4.9263	80.8766	94.0522	-
2	GENE871	4654021	0.0677	0.1264	1.8682	3.0871	94.3878	100	=
3	GENE872	4654022	-0.2398	-0.4027	1.6793	0.8102	86.4446	94.1904	
4	GENE873	4654023	-0.0792	-0.1556	1.9641	1.3397	91.078	99.4605	
5	GENE874	4654024	0.0535	0.1033	1.9306	1.7484	94.3878	100	
6	GENE875	4654025	-0.2488	-0.5128	2.0609	1.1078	86.4446	93.8744	1
7	GENE876	4654026	-0.2298	-0.3941	1.715	0.823	86.4446	94.5365	
8	GENE877	4654027	-0.6001	-1.0456	1.7423	0.4787	77.8565	80.77	1
9	GENE878	4654028	-0.3534	-0.605	1.712	0.6823	86.4446	89.5737	1
10	GENE826	4653976	-0.0168	-0.0245	1.4572	1.0481	92.688	100	1
11	GENE827	4653977	-0.2985	-0.4591	1.5381	0.7289	86.4446	92.0149	1
12	GENE828	4653978	-0.1136	-0.2142	1.8858	1.3896	89.6273	98.399	1
13	GENE829	4653979	0.3222	0.5884	1.826	3.588	86.3568	95.7861	1
14	GENE830	4653980	0.0323	0.0533	1.6476	1.2171	94.3878	100	1
15	GENE831	4653981	0.0076	0.0122	1.5909	1.1026	94.3878	100	1
16	GENE832	4653982	-0.0855	-0.1446	1.6914	1.2943	91.078	99.2674	1
17	GENE833	4653983	0.3023	0.4876	1.6128	2.1278	86.4309	96.1364	-
	•							•	Ĩ
				7129 gene	s				
۲	iificance Filtering Target False Disc	covery Rate (		) Delta =					
$\circ$	Select # genes	by	largest SAM absolute s	core 💌					
Mea	an Channel Intens	sities >	Bad Flags <=						
Abs	Fold Change >								
			Apply Filters	Clear f	ilters				
	Crea	te Sig. Gene l	.ist 😽 HCA	<mark>≫≛</mark> PCA	K-Mea	ns 🖉 S	SAM Plot	>	

#### SAM-Test: Timecourse

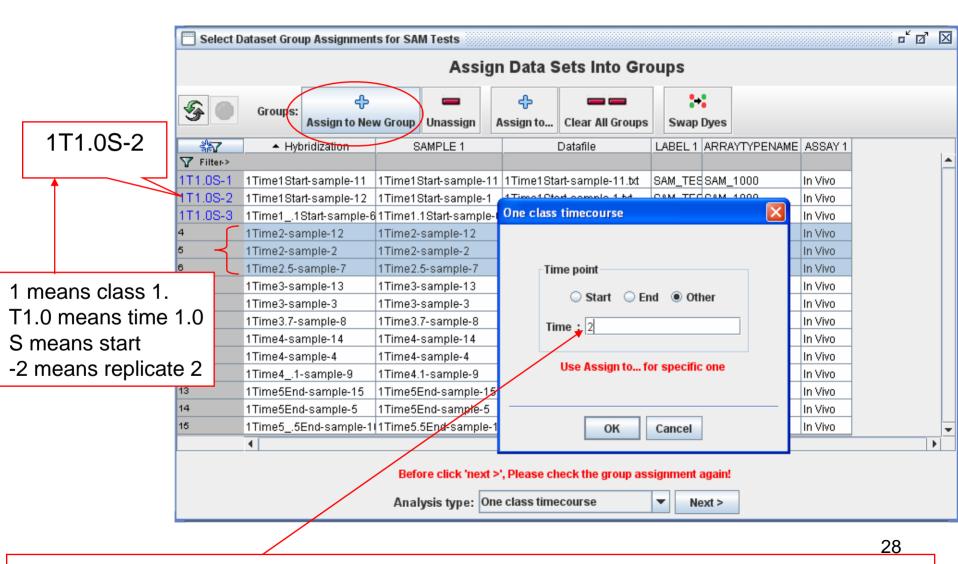
#### One class timecourse



#### SAM-Test: one class timecourse

Highlight the first group, click "Assign to New Group" button.

		Assign	n Data Sets Into Gro	oups			
<b>%</b>	Groups: 合 Assign to New	v Group Unassign	수 Clear All Groups	Swap Dyes			
	<ul> <li>Hybridization</li> </ul>	SAMPLE 1	Datafile	LABEL 1 ARRA	YTYPENAME	ASSAY 1	
🍸 Filter>							<b>_</b>
1	1Time1Start-sample-11	1Time1Start-sample-11	1Time1Start-sample-11.txt	SAM_TES SAM_	1000	In Vivo	
2 ┥	1Time1Start-sample-12	1Time1Start-sample-1	1Time1Start-sample-1.txt	SAM_TESSAM_	1000	In Vivo	
з (	_1Time11Start-sample-6	1Time1.1Start-sample-6	6 1Time1.1Start-sample-6.txt	SAM_TES SAM_	1000	In Vivo	
4	1Time2-sample-12	1Time2-sample-12	1Time2-sample-12.txt	SAM_TES SAM_	1000	In Vivo	
5	1Time2-sample-2	1Time2-sample-2	1Time2-sample-2.txt	SAM_TES SAM_	1000	In Vivo	
6	1Time2.5-sample-7	1Time2.5-sample-7	1Time2.5-sample-7.txt	SAM_TES SAM_	1000	In Vivo	
7	1Time3-sample-13	1Time3-sample-13	1Time3-sample-13.txt	SAM_TES SAM_	1000	In Vivo	
8	1Time3-sample-3	1Time3-sample-3	1Time3-sample-3.txt	SAM_TES SAM_		In Yéya	
9	1Time3.7-sample-8	1Time3.7-sample-8	1Time3.7-sample-8.txt	SAM_TESSAM_	One clas	s timecourse	
10	1Time4-sample-14	1Time4-sample-14	1Time4-sample-14.txt	SAM_TESSAM_	1		
11	1Time4-sample-4	1Time4-sample-4	1Time4-sample-4.txt	SAM_TESSAM_	1		
12	1Time41-sample-9	1Time4.1-sample-9	1Time4.1-sample-9.txt	SAM_TESSAM_	<u>1</u>	me point	
13	1Time5End-sample-15	1Time5End-sample-15	1Time5End-sample-15.txt	SAM_TESSAM_	1	me point	
14	1Time5End-sample-5	1Time5End-sample-5	1Time5End-sample-5.txt	SAM_TES SAM_	1	🖲 Start i 🔾 E	nd 🔾 Other
15	1Time55End-sample-1	1Time5.5End-sample-1	(1Time5.5End-sample-10.txt	SAM_TESSAM_	1		_
	•				Т	ime: 1	
		Diagona	opian the areas information (			,	
	<b>a</b>		ssign the group information i			Use Assign to t	for specific o
	Analy	sis type: One class tin	necourse 💌 <<	<must assign="" gr<="" td=""><td></td><td></td><td></td></must>			
				$\neg$			
"Start	" radio buttor	n type in nu	ımber "1" in th			ок	Cancel



For second group, check "Other" radio button and type in number "2" for time course.

Repeat the previous steps for assigning the other groups. For the last group, check "End" radio button and type in the time. Click OK button.

🔲 Select Dataset Group Assignments for SAM Tests									
		Assigr	n Data Sets Into Groups						
<b>\$</b>	Groups: 4	v Group Unassign A	Image: State of the state o						
** <b>*</b> 7	<ul> <li>Hybridization</li> </ul>	SAMPLE 1	One class timecourse						
🍸 Filter->									
1T1.0S-1	1Time1Start-sample-11	1Time1Start-sample-11	1Tim vo						
1T1.0S-2	1Time1Start-sample-12	1Time1Start-sample-1	1Tim vo						
1T1.0S-3	1Time11Start-sample-6	1Time1.1Start-sample-6	1Tim Time point ivo						
1T2.0-1	1Time2-sample-12	1Time2-sample-12	1Tim 🔾 Start 🖲 End 🔾 Other						
1T2.0-2	1Time2-sample-2	1Time2-sample-2	1Tim ivo						
1T2.0-3	1Time2.5-sample-7	1Time2.5-sample-7	1Tim Time: 5 ivo						
1T3.0-1	1Time3-sample-13	1Time3-sample-13	1Tim ivo						
1T3.0-2	1Time3-sample-3	1Time3-sample-3	1Tim Use Assign to for specific one ivo						
1T3.0-3	1Time3.7-sample-8	1Time3.7-sample-8	1Tim ivo						
1T4.0-1	1Time4-sample-14	1Time4-sample-14	1Tim ivo						
1T4.0-2	1Time4-sample-4	1Time4-sample-4	1Tim ivo						
1T4.0-3	1Time41-sample-9	1Time4.1-sample-9	1Tim OK Cancel ivo						
13	1Time5End-sample-15	1Time5End-sample-15	1Timivo						
14	1Time5End-sample-5	1Time5End-sample-5	1Time5End-sample-5.bt SAM_TES SAM_1000 In Vivo						
15	1Time55End-sample-1	1Time5.5End-sample-1	(1Time5.5End-sample-10.txt  SAM_TE\$ SAM_1000 In Vivo 🔤 🤤						
	•								
		Before click 'nevt >'	', Please check the group assignment again!						
		Dervie chen next >	, r rease encon the group assignment again						
		Analysis type: One	e class timecourse   Next >						

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This is what looks like after assigning all the groups.

		🔲 Select D	Dataset Group Assignment	ts for SAM Tests						r 0	$\boxtimes$
				Assi	gn Data S	Sets Into Gro	oups				
		<b>%</b>	Groups: Assign to New	w Group Unassign	႕ Assign to	Clear All Groups	Swap Dyes				
		**7	<ul> <li>Hybridization</li> </ul>	SAMPLE 1		Datafile	LABEL 1	ARRAYTYPENAME	ASSAY 1		
<b>T</b> :	~	Filter->									
Time 1	J	1T1.0S-1 1T1.0S-2		1Time1Start-sample-1		art-sample-11.txt		SAM_1000	In Vivo		
Start	Start			1Time1Start-sample-1		art-sample-1.txt		SAM_1000	In Vivo		
	C	1T1.0S-3	1Time11Start-sample-6					SAM_1000	In Vivo		
ſ		1T2.0-1		1Time2-sample-12		ample-12.txt		SAM_1000	In Vivo		
Time 2	$\prec$	1T2.0-2	1Time2-sample-2	1Time2-sample-2		ample-2.txt	SAM_TEST 8	SAM_1000	In Vivo		
	L	1T2.0-3	1Time2.5-sample-7	1Time2.5-sample-7	1Time2.5-	sample-7.txt	SAM_TEST S	SAM_1000	In Vivo		
	٢	1T3.0-1 11	1Time3-sample-13	1Time3-sample-13	1Time3-sa	1Time3-sample-13.txt		SAM_1000	In Vivo		
Time 3	$\prec$	1T3.0-2	1Time3-sample-3	1Time3-sample-3	1Time3-sa	ample-3.txt	SAM_TEST 8	SAM_1000	In Vivo		
	L	1T3.0-3	1Time3.7-sample-8	1Time3.7-sample-8	1Time3.7-:	sample-8.txt	SAM_TEST 8	SAM_1000	In Vivo		
	ſ	1T4.0-1	1Time4-sample-14	1Time4-sample-14	1Time4-sa	1Time4-sample-14.txt		SAM_1000	In Vivo		
Time 4	7	1T4.0-2	1Time4-sample-4	1Time4-sample-4	1Time4-sa	ample-4.txt	SAM_TEST S	SAM_1000	In Vivo		
		1T4.0-3	1Time41-sample-9	1Time4.1-sample-9	1Time4.1-:	sample-9.txt	SAM_TEST 8	SAM_1000	In Vivo		
	ſ	1T5.0E-1	1Time5End-sample-15	1Time5End-sample-1	15 1Time5En	d-sample-15.txt	SAM_TEST 8	SAM_1000	In Vivo		
Time 5	7	1T5.0E-2	1Time5End-sample-5	1Time5End-sample-5	5 1Time5En	d-sample-5.txt			In Vivo		
End	L	1T5.0E-3	1Time55End-sample-11						In Vivo		Ŧ
			•							•	
				Before click 'nex	t >', Please ch	neck the group ass	signment again	l.			
				Analysis type: 0	)ne class time	course	Next >	$\supset$			

🚍 Select Dataset Group Assignments for SAM Tests 🛛 🗹	
One class timecourse problem	
Significance Analysis of Microarrays (SAM)	
SAM-Tests Options These value	les are default
Time summary method: <ul> <li>Slop</li> <li>Signed area</li> </ul>	
Number of permutations: Limit to(100~1000): 100	
Estimation of s0 factor:   Automatic  Used fixed percentile(%)  50	
Imputation engine: Number of neighbors of KNN 10	
Random number seed: 1234567 Generate Random Seed	
Calculate the fold change's effect on FDR (may take a while)? <ul> <li>No</li> <li>Yes</li> </ul>	
Filtering with a gene list	
Only include genes from gene list <a>all genes&gt;</a>	
Gene identifiers to include	
Genbank Acc Gene Mfr ID CUSID UNIGENEID GENENAME	
< Back Do Tests	31

One class time course result:

File	Selected-Spot	All-Spots							
	Gene Mfr ID	SPOTID	Observed score (d)	Numerator(r)	Denominat	q-value(%)	localfdr(%)	Expected	s
	GENE298	4661077	1.0215	0.698	0.6833	37.35	87.4649	0.9613	٦
	GENE508	4661287	0.9564	0.5229	0.5468	37.35	83.5332	0.9089	1
	GENE840	4661619	0.922	0.4409	0.4782	37.35	81.4801	0.8734	1
	GENE881	4661660	0.9181	0.8478	0.9235	37.35	81.2569	0.8585	1
	GENE247	4661026	0.8902	0.4638	0.5211	37.35	79.7323	0.8406	
	GENE585	4661364	0.8882	0.5565	0.6266	37.35	79.6336	0.8283	1
	GENE720	4661499	0.8859	0.4105	0.4634	37.35	79.5172	0.8095	1
	GENE317	4661096	0.8779	0.5037	0.5738	37.35	79.1376	0.7994	Ī
	GENE665	4661444	0.8471	0.5516	0.6512	41.5	77.9312	0.7843	1
I	GENE274	4661053	0.8441	0.4578	0.5424	41.5	77.84	0.7756	
	GENE49	4660828	0.8265	0.3842	0.4649	41.5	77.3926	0.7649	
2	GENE929	4661708	0.8238	0.3418	0.4149	41.5	77.3386	0.7606	
	GENE940	4661719	0.7902	0.3505	0.4435	53.3571	76.9903	0.749	
ł	GENE272	4661051	0.789	0.642	0.8136	53.3571	76.9886	0.7438	
;	GENE149	4660928	0.7673	0.342	0.4457	53.3571	77.0805	0.7338	
;	GENE264	4661043	0.7441	0.4767	0.6406	53.3571	77.42	0.7246	1
r	GENE62	4660841	0.7307	0.6164	0.8436	53.3571	77.7225	0.715	Ī
;	GENE953	4661732	0.7302	0.5165	0.7074	53.3571	77.7363	0.707	Ī
	GENE829	4661609	0.7240	0.455	0 6276	52 2671	77 975	0 7037	_
Sign	ificance Filtering	J		1000 ger	ies			•	
۲	Target False Dis	covery Rate (	FDR):	O Delta =					
$\bigcirc$	Select # genes	by	largest SAM absolute	score 💌					
Mea	in Channel Intens	sities >	Bad Flags <=						
			Apply Filter	rs Clear	Filters				
	Crea		List <mark>A HCA</mark>	×å PCA	K-Me		SAM Plot		

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#### SAM-Test: Two Class Unpaired Timecourse

👇 🛉 SAM-EXAMPLES_TEST		
- 📑 MultiClass_500		
⊷ 🗐 OneClass_1000		
- 🗐 Paired_7129		
🕶 🗐 PatternDiscovery_1000		
🔶 🗂 Quantitative_7129		
🔶 🗖 Survival_7129		
Ⴡ 🗂 TwoClassBig_2000		
TwoClassBlocked_7129		
🗠 🗂 TwoClassMissing_500		
TwoClassPairedTimecourse_1		
TwoClassUnpairedTimecourse		
🕂 🎢 1Time1.1Start-sample-6 1T		
── 🎹 raw {1Time1.1Start-sam		
👇 🗰 1Time1Start-sample-1 1Tir	▦	View data set(s) as wide spreadsheet - datasets side by side
── 🏢 raw {1Time1Start-sampl		Export •
- ITime1Start-sample-11 1T	H	Mixed scatterplot
☐ III raw {1Time1Start-sampl		Virtual array images for data
- ITime2-sample-12 1Time2	1	Actual array images for data < <dev. only="">&gt;</dev.>
III raw {1Time2-sample-12		
- Imme2-sample-2 + Hime2- me2-sample-2 + Hime2-sample-2 +		Rank intensity plots for data
raw {11ine2-sample-2}	լդի	BarChart
milline2.5-sample-7		Create gene list by data filtering
P III 1Time3-sample-13 1Time3		Analysis T-Test/ANOVA
- III raw (1Time3-sample-13		Quality Control SAM-Test
- 🗰 1Time3-sample-3 1Time3-	N	Normalize @ R-Interface
Taw (1Time3-sample-3)	_	Duplicate data sets Correlation Matrix
🕂 🎆 1Time3.7-sample-8 1Time		Copy data sets for pasting elsewhere T-Test with custom data options
🗌 🏢 raw (1Time3.7-sample-8		
1Timot comple 14 1Timot		
Library		Tree options 🖧 Hierarchical Cluster Analysis

Right-click the selected data set, choose "Analysis" ->SAM-Test.

#### SAM-Test: Two Class Unpaired Timecourse

Here the steps are similar as one class time course. User assign start time point, middle point and end time point for class one. And then repeat the steps for class two. Just remember if class one is control group, then class two will be treated group.

Assign Data Sets Into Groups								
	Groups:	දා Assign to New Gro	up Unassign	႕ Assign to	Clear All Groups	<b>\$</b> Swap Dyes		
*	<b>A</b>	Hybridization	SAMP	LE 1	Data	afile	LABEL 1	ARRAYTYF
Filter->								
	1Time1.1S	tart-sample-6	1Time1.1Start-s	ample-6	1Time1.1Start-sar	mple-6.txt	SAM_TEST	SAM_1000
	1Time1Sta	rt-sample-1	1Time1Start-sa	mple-1	1Time1Start-sam	ple-1.txt	SAM_TEST	SAM_1000
	1Time1Sta	rt-sample-11	1Time1Start-sa	mple-11	1Time1Start-sam	ple-11.txt	SAM_TEST	SAM_1000
	1Time2-sa	mple-12	1Time2-sample	-12	1Time2-sample-1		SAM_TEST	SAM_1000
	1Time2-sa	mple-2	There is a second				SAM_TEST	SAM_100
1Time2.5-sample-7			Two class unp	aired timeco	urse		SAM_TEST	SAM_100
	1Time3-sa	mple-13					SAM_TEST	SAM_100
	1Time3-sa	mple-3					SAM_TEST	SAM_100
	1Time3.7-s	sample-8	Class a	SAM_TEST	SAM_100			
	1Time4-sa	mple-14	Control     Treated			SAM_TEST	SAM_100	
	1Time4-sa	mple-4		Control 9 Treated			SAM_TEST	SAM_100
	1Time4.1-s	sample-9	-Time po	pint			SAM_TEST	SAM_100
	1Time5.5E	nd-sample-10		Ctart O Fau		.txt	SAM_TEST	SAM_100
	1Time5End	d-sample-15	•	Start 🔾 End	a Uner	t	SAM_TEST	SAM_100
	1Time5End	d-sample-5	Time :	1			SAM_TEST	SAM_100
	2Time1.1S	tart-sample-16		1		i.txt	SAM_TEST	SAM_100
	2Time1.1S	tart-sample-26	lleo	Acciento fo	r enocific ono	i.txt	SAM_TEST	SAM_100
	2Time1Sta	rt-sample-21	Use	Assign to fo	r specific one	đ	SAM_TEST	SAM_100
	2Time2-sa	mple-22					SAM_TEST	SAM_100
	2Time2.5-s	sample-17					SAM_TEST	SAM_100
	2Time2.5-s	sample-27		ок	Cancel		SAM_TEST	SAM_100
	2Time3-sa	mple-23					SAM_TEST	SAM_100
	2Time3.7-s	sample-18	2Time3.7-samp	le-18	2Time3.7-sample	-18.txt	SAM_TEST	SAM_100
	2Time3.7-s	sample-28	2Time3.7-samp	le-28	2Time3.7-sample	-28.txt	SAM_TEST	SAM_100
	2Time4-sa	mple-24	2Time4-sample	-24	2Time4-sample-2	4.txt	SAM_TEST	SAM_1000
	2Time4.1-s	sample-19	2Time4.1-samp	)le-19	2Time4.1-sample	-19.txt	SAM_TEST	SAM_1000
	2Time4.1-s	sample-29	2Time4.1-samp	le-29	2Time4.1-sample	-29.txt	SAM_TEST	SAM_1000
	1	ad example 20	OTHER COURSE		OTHER CERTIN		ONN TEOT	0000 4000
	•							
		Befor	e click 'next >', I	Please check t	the group assignm	ent again!		

Make sure the analysis type is Selected before assigning class.

#### SAM-Test: Two Class Unpaired Timecourse

	📄 Select D	ataset Gro	up Assignments for	SAM Tests					• <b>•</b> • •
				Assign	Data Sets	Into Groups			
	<b>\$</b>	Groups:	ද්- Assign to New Gro	up Unassign	႕ Assign to	Clear All Groups	<b>}+</b> Swap Dyes		
	*7	-	Hybridization	SAM	PLE 1	Dataf	ile	LABEL 1	ARRAYTYPE
	🕎 Filter->								
(	1T1.0S-1	1Time1.1S	tart-sample-6	1Time1.1Start-	sample-6	1Time1.1Start-sam	ple-6.txt	SAM_TEST	SAM_1000
	1T1.0S-2	1Time1Sta	art-sample-1	1Time1Start-sa	ample-1	1Time1Start-sampl	e-1.txt	SAM_TEST	SAM_1000
	1T1.0S-3	1Time1Sta	art-sample-11	1Time1Start-sa	ample-11	1Time1Start-sampl	e-11.txt	SAM_TEST	SAM_1000
	1T2.0-1	1Time2-sa	ample-12	Two class unp	aired timecou	irse		SAM_TEST	SAM_1000
	1T2.0-2	1Time2-sa		nno cruss unp	anea timecou	1196		SAM_TEST	SAM_1000
Class and from start	1T2.0-3	1Time2.5-:	sample-7					SAM_TEST	SAM_1000
Class one from start	1T3.0-1	1Time3-sa	imple-13	Class				SAM_TEST	SAM_1000
to and time point	1T3.0-2	1Time3-sa	imple-3	Class a	ssign	$\frown$		SAM_TEST	SAM_1000
to end time point.	1T3.0-3	1Time3.7-:	sample-8		🔾 Control 🌔	Treated		SAM_TEST	SAM_1000
	1T4.0-1	1Time4-sa	ample-14					SAM_TEST	SAM_1000
	1T4.0-2	1Time4-sa	ample-4	Time po	int			SAM_TEST	SAM_1000
	1T4.0-3	1Time4.1-:	sample-9	۲	Start 🔾 End	Other		SAM_TEST	SAM_1000
	1T5.0E-1	1Time5.5E	nd-sample-10	Ĭ			0.txt	SAM_TEST	SAM_1000
	1T5.0E-2	1Time5En	d-sample-15	Time :	1		bat	SAM_TEST	SAM_1000
×	1T5.0E-3	1Time5En	d-sample-5				t	SAM_TEST	SAM_1000
	16	2Time1.19	itart-sample-16	Use	Assign to for	specific one	6.bd	SAM_TEST	SAM_1000
	17	2Time118	tart-sample-26		, in the second		:6.bd	SAM_TEST	SAM_1000
	18	211me1Sta	art-sample-21				.txt	SAM_TEST	SAM_1000
	19	2Time2-sa	ample-22					SAM_TEST	SAM_1000
Repeat the steps to	20	2Time2.5-	sample-17		ок	Cancel		SAM_TEST	SAM_1000
•	21	2Time2.5-	sample-27 🛛 📘	1				SAM_TEST	SAM_1000
assign class two.	22	2Time3-sa	imple-23	2Time3-sampl	e-23	2Time3-sample-23	.txt	SAM_TEST	SAM_1000
•	23	2Time3.7-:	sample-18	2Time3.7-sam	ple-18	2Time3.7-sample-1	8.txt	SAM_TEST	SAM_1000
Class two is treated.	24	2Time3.7-:	sample-28	2Time3.7-sam	ple-28	2Time3.7-sample-2	28.txt	SAM_TEST	SAM_1000
	25	2Time4-sa	ample-24	2Time4-sampl	e-24	2Time4-sample-24	.txt	SAM_TEST	SAM_1000
	26	2Time4.1-	sample-19	2Time4.1-sam	ple-19	2Time4.1-sample-1	9.txt	SAM_TEST	SAM_1000
	27	27 2Time4.1-sample-29		2Time4.1-sam	ple-29	2Time4.1-sample-2	29.txt	SAM_TEST	SAM_1000
	20	A		LOTING C CENT		DTHE STORE STORE	-1- 00 ++	OWN TEOT	A 4000
						the group assignment	nt again! Next >		

# SAM-Test: Two Class Unpaired

necourse Select Dataset Group Assignments for SAM Tests

Here is what lo after assigning

lere is what looks like		Assign Data Sets Into Groups								
fter assigning.	<b>%</b>	Groups: Assign to New	w Group Unassign As	႕ာ ssign to Clear All Groups	<b>;→;</b> Swap Dyes					
	*7	<ul> <li>Hybridization</li> </ul>	SAMPLE 1	Datafile	LABEL 1	ARRAYTYPENAME	ASSAY 1			
	🕎 Filter>									
(	1T1.0S-1	1Time1.1Start-sample-6	1Time1.1Start-sample-6	1Time1.1Start-sample-6.txt	SAM_TEST	SAM_1000	In Vivo			
	1T1.0S-2	1Time1Start-sample-1	1Time1Start-sample-1	1Time1Start-sample-1.txt	SAM_TEST	SAM_1000	In Vivo			
	1T1.0S-3	1Time1Start-sample-11	1Time1Start-sample-11	1Time1Start-sample-11.txt	SAM_TEST	SAM_1000	In Vivo			
	1T2.0-1	1Time2-sample-12	1Time2-sample-12	1Time2-sample-12.txt	SAM_TEST	SAM_1000	In Vivo			
	1T2.0-2	1Time2-sample-2	1Time2-sample-2	1Time2-sample-2.txt	SAM_TEST	SAM_1000	In Vivo			
	1T2.0-3	1Time2.5-sample-7	1Time2.5-sample-7	1Time2.5-sample-7.txt	SAM_TEST	SAM_1000	In Vivo			
	1T3.0-1	1Time3-sample-13	1Time3-sample-13	1Time3-sample-13.txt	SAM_TEST	SAM_1000	In Vivo			
Class one $\checkmark$	1T3.0-2	1Time3-sample-3	1Time3-sample-3	1Time3-sample-3.txt	SAM_TEST	SAM_1000	In Vivo			
	1T3.0-3	1Time3.7-sample-8	1Time3.7-sample-8	1Time3.7-sample-8.txt	SAM_TEST	SAM_1000	In Vivo			
	1T4.0-1	1Time4-sample-14	1Time4-sample-14	1Time4-sample-14.txt	SAM_TEST	SAM_1000	In Vivo			
	1T4.0-2	1Time4-sample-4	1Time4-sample-4	1Time4-sample-4.txt	SAM_TEST	SAM_1000	In Vivo			
	1T4.0-3	1Time4.1-sample-9	1Time4.1-sample-9	1Time4.1-sample-9.txt	SAM_TEST	SAM_1000	In Vivo			
	1T5.0E-1	1Time5.5End-sample-10	1Time5.5End-sample-10	1Time5.5End-sample-10.txt	SAM_TEST	SAM_1000	In Vivo			
	1T5.0E-2	1Time5End-sample-15	1Time5End-sample-15	1Time5End-sample-15.txt	SAM_TEST	SAM_1000	In Vivo			
N N	1T5.0E-3	1Time5End-sample-5	1Time5End-sample-5	1Time5End-sample-5.txt	SAM_TEST	SAM_1000	In Vivo			
(	2T1.0S-1	2Time1.1Start-sample-16	2Time1.1Start-sample-16	2Time1.1Start-sample-16.txt	SAM_TEST	SAM_1000	In Vivo			
	2T1.0S-2	2Time1.1Start-sample-26	2Time1.1Start-sample-26	2Time1.1Start-sample-26.txt	SAM_TEST	SAM_1000	In Vivo			
	2T1.0S-3	2Time1Start-sample-21	2Time1Start-sample-21	2Time1Start-sample-21.txt	SAM_TEST	SAM_1000	In Vivo			
	2T2.0-1	2Time2-sample-22	2Time2-sample-22	2Time2-sample-22.txt	SAM_TEST	SAM_1000	In Vivo			
	2T2.0-2	2Time2.5-sample-17	2Time2.5-sample-17	2Time2.5-sample-17.txt	SAM_TEST	SAM_1000	In Vivo			
	2T2.0-3	2Time2.5-sample-27	2Time2.5-sample-27	2Time2.5-sample-27.txt	SAM_TEST	SAM_1000	In Vivo			
	2T3.0-1	2Time3-sample-23	2Time3-sample-23	2Time3-sample-23.txt	SAM_TEST	SAM_1000	In Vivo			
Class two 🖌	2T3.0-2	2Time3.7-sample-18	2Time3.7-sample-18	2Time3.7-sample-18.txt	SAM_TEST	SAM_1000	In Vivo			
	2T3.0-3	2Time3.7-sample-28	2Time3.7-sample-28	2Time3.7-sample-28.txt	SAM_TEST	SAM_1000	In Vivo			
	2T4.0-1	2Time4-sample-24	2Time4-sample-24	2Time4-sample-24.txt	SAM_TEST	SAM_1000	In Vivo			
	2T4.0-2	2Time4.1-sample-19	2Time4.1-sample-19	2Time4.1-sample-19.txt	SAM_TEST	SAM_1000	In Vivo			
	2T4.0-3	2Time4.1-sample-29	2Time4.1-sample-29	2Time4.1-sample-29.txt	SAM_TEST	SAM_1000	In Vivo			
	2T5.0E-1	2Time5.5End-sample-20	2Time5.5End-sample-20	2Time5.5End-sample-20.txt	SAM_TEST	SAM_1000	In Vivo			
	2T5.0E-2	2Time5.5End-sample-30	2Time5.5End-sample-30	2Time5.5End-sample-30.txt	SAM_TEST	SAM_1000	In Vivo			
	2T5.0E-3	2Time5End-sample-25	2Time5End-sample-25	2Time5End-sample-25.txt	SAM_TEST	SAM_1000	In Vivo			
		•					•			
			Before click 'post s'	Please check the group assi	Inmont adaint					
Olial, Nave button to			Dervie click liext >,	ricuse check the group assi	ginnen ayalli:					
Click Next button to g	jet SA	livi-test —	Analysis type: Two	class unpaired timecourse	Next >					

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<ul> <li>SAM-EXAMPLES_TEST</li> <li>MultiClass_500</li> <li>OneClassTimecourse_1000</li> <li>OneClass_1000</li> <li>Paired_7129</li> <li>PatternDiscovery_1000</li> <li>Quantitative_7129</li> <li>Survival_7129</li> <li>Gene Lists</li> <li>(1.3, 1)-sample-6 (1.3, 1)-sample-7 (1.3</li></ul>	View data set(s) as wide spreadsheet - datasets side by side Export Mixed scatterplot Virtual array images for data		
👇 🖬 (57.8, 1)-sample-5 (57.8	Create gene list by data filtering		
- Ⅲ raw {(57.8, 1)-sample	Analysis	100	T-Test/ANOVA
	Quality Control		SAM-Test
• III (623.0, 1)-sample-8 (62 N	Normalize	C C	
🗏 🏢 raw {(623.0, 1)-samp	Duplicate data sets		Correlation Matrix
∽ 🗂 TwoClassBig_2000	Copy data sets for pasting elsewhere	-	
TwoClassBlocked_7129	Studies		ANOVA with custom data options
• 🗂 TwoClassMissing_500	Tree options	A <sup>2</sup>	
Ⴡ 🗂 TwoClassPairedTimecours(	The options.	<mark>-</mark> ф	Hierarchical Cluster Analysis

Right-click the selected data set, choose "Analysis" -> SAM-Test

This data set has two groups. The first number in parenthesis represent time and the second number in parenthesis (1, or 0) represents "died" (1) or "censored"(0) group.

Select the first data – (1.3, 1)-sample-6, click "Assign to New Group" button. Type 1.3 in the time area.

📋 Select D	🗂 Select Dataset Group Assignments for SAM Tests 🛛 🗖 🖂									
	Assign Data Sets Into Groups									
<b>%</b>	Groups:	Assign to	New Group	unassign	Clear I	All Groups	<b>\$</b> → Swap Dyes			
*7	Hybrid	ization	SAM	PLE 1		Survival		×	ME ASSAY1	
Y       Filter.>         1       2         2       3         4       5         6       6	(1.3, 1)-sar (1151.0 0) (1326.3, 1) (145.0, 1)-s (506.1, 1)-s (57.8, 1)-sa	sample-2 -sample-1 sample-4 sample-7	(1.3, 1)-sam (1151.0, 0)- (1326.3, 1)- (145.0, 1)-s (506.1, 1)-s (57.8, 1)-sa	sample-2 sample-1 ample-4 ample-7	(1.3, 1)-s (1151.0, (1326.3, (145.0, 1 (506.1, 1 (57.8, 1)	C Ti	Died	Censored	In Vivo In Vivo In Vivo In Vivo In Vivo In Vivo	
7         (605.4, 0)-sample-3         (605.4, 0)-sample-3         (605.4, 0)           8         (623.0, 1)-sample-8         (623.0, 1)-sample-8         (623.0, 1)							ок	Cancel	In Vivo In Vivo	
Before click 'next >', Please check the group assignment again!										
	Analysis type. Survival   Next >									

Repeat the previous steps to assign the other data of group 1, type in the time.

Select D	Select Dataset Group Assignments for SAM Tests									$\boxtimes$			
	Assign Data Sets Into Groups												
<b>∳</b> ●	Groups:		႕ New Group	unassign			;+; Swap D						
*7	Hybrid	-	-	Survival				×	1	ARRAYTYPENAME	ASSAY 1		
	(1.3, 1)-sar	nple-6	(1.3, 1)-sam						ŝТ	SAM_7K	In Vivo		Ê
2 3	(1151.0, 0)- (1326.3, 1)-		(1151.0, 0)-s (1326.3, 1)-s		🖌 Died	Censo	red		ST ST	SAM_7K SAM_7K	In Vivo In Vivo		
4 5	(145.0, 1)-s (506.1, 1)-s	ample-4	(145.0, 1)-sa (506.1, 1)-sa	T	ime : 132	6.3			ST PT	SAM_7K SAM_7K	In Vivo In Vivo		
6	(57.8, 1)-sa	ample-5	(57.8, 1)-san						ST	SAM_7K	In Vivo		
7 8	(605.4, 0)-s (623.0, 1)-s		(605.4, 0)-sa (623.0, 1)-sa		ок	Cance	el		ST ST	SAM_7K SAM_7K	In Vivo In Vivo		-
	Before click 'next >', Please check the group assignment again!												
			Analysis ty	ype: Surviv	<i>r</i> al		-	N	ext >	<b>,</b>			

This image shows all the data of group 1 has been assigned.

🔲 Select D	🔲 Select Dataset Group Assignments for SAM Tests								í 🗹	X		
	Assign Data Sets Into Groups											
<b>%</b>	Groups:		႕ New Group	unassign	Clear All Groups		<b>.→</b> Ip Dyes					
*7	Hybrid	ization	SAMP	'LE 1	Datafile		LABE	L1	ARRAYTYPENAME	ASSAY 1		
🕎 Filter->												
(1.3,1)	(1.3, 1)-san	nple-6	(1.3, 1)-sam	ple-6	(1.3, 1)-sample-6.txt		SAM_TE	EST	SAM_7K	In Vivo		
2				(1151.0, 0)-sample-:	SAM_TEST		SAM_7K	In Vivo				
(1326.3,1)	(1326.3, 1)-	-sample-1	(1326.3, 1)-s	ample-1	(1326.3, 1)-sample-1.txt		SAM_TEST		SAM_7K	In Vivo		
(145.0,1)	(145.0, 1)-s	ample-4	(145.0, 1)-sa	imple-4	(145.0, 1)-sample-4.txt		SAM_TEST		SAM_7K	In Vivo		
(506.1,1)	(506.1, 1)-s	ample-7	(506.1, 1)-sa	imple-7	(506.1, 1)-sample-7.txt		SAM_TE	EST	SAM_7K	In Vivo		
(57.8,1)	(57.8, 1)-sa	ample-5	(57.8, 1)-san	nple-5	(57.8, 1)-sample-5.txt SA			EST	SAM_7K	In Vivo		
7	(605.4, 0)-s	ample-3	(605.4, 0)-sa	imple-3	(605.4, 0)-sample-3.	.txt	SAM_TE	EST	SAM_7K	In Vivo		
(623.0,1)	(623.0, 1)-s	ample-8	(623.0, 1)-sa	imple-8	(623.0, 1)-sample-8.	.txt	SAM_TE	EST	SAM_7K	In Vivo		
	•										•	-
	Before click 'next >', Please check the group assignment again!											
			Analysis ty	ype: Surviv	/al		-	Next >	>			

Select the 2<sup>nd</sup> data which is in group 0, click "Assign to New Group" button. Check "Censored" check box and type in the time.

🔲 Select D	🗂 Select Dataset Group Assignments for SAM Tests 🛛 🗖 🖂											
	Assign Data Sets Into Groups											
<b>%</b>	Groups:		႕ New Group	<b>ma</b> Unassign	Clear All Groups		<b>→</b> p Dyes					
*7	Hybrid	ization	SAMF	PLE 1	Datafile		LABEL	1 ARRA	YTYPENAME	ASSAY 1		
🍸 Filter>												
(1.3,1)	(1.3, 1)-sar	nple-6	(1.3, 1)-sam	iple-6	Survival				ĸ	In Vivo		
2	(1151.0,0)	sample-2	(1151.0, 0)-:	sample-2					ĸ	In Vivo		
(1326.3,1)	(1326.3, 1)-	sample-1	(1326.3, 1)-	sample-1					ĸ	In Vivo		
(145.0,1)	(145.0, 1)-s	ample-4	(145.0, 1)-s:	ample-4					ĸ	In Vivo		
(506.1,1)	(506.1, 1)-s	ample-7	(506.1, 1)-s:	ample-7	🗌 Died	🖌 C	ensored		ĸ	In Vivo	1	
(57.8,1)	(57.8, 1)-sa	mple-5	(57.8, 1)-sai	mple-5	Time 1 dd	54.0		_	ĸ	In Vivo		
7	(605.4, 0)-s	ample-3	(605.4, 0)-sa	ample-3	Time : 11	51.0			ĸ	In Vivo		
(623.0,1)	(623.0, 1)-s	ample-8	(623.0, 1)-s:	ample-8					ĸ	In Vivo		
	Image: Concel       Image: Before click 'next >', F								<b>▼</b> 			
	Analysis type: Survival   Next >											

Do the same steps for the other data of group 0.

🗍 Select D	🔲 Select Dataset Group Assignments for SAM Tests 📃 🗖 🖂													
	Assign Data Sets Into Groups													
<u></u>	Groups:		} New Group	<b>unassign</b>		All Groups		<b>→</b> p Dyes						
*7	Hybrid	ization	SAMF	PLE 1		Datafile		LABE	L1	ARRAYTYPENA	ME	ASSAY 1		
🍸 Filter>						Survival								
(1.3,1)	(1.3, 1)-sar	nple-6	(1.3, 1)-sam	iple-6	(1.3, 1)-	Survivat						In Vivo		
(1151.0,0)	(1151.0, 0)-	sample-2	(1151.0, 0)-:	sample-2	(1151.0	(1151.0 In				In Vivo				
(1326.3,1)	(1326.3, 1)-	sample-1	(1326.3, 1)-	sample-1	(1326.3							In Vivo		
(145.0,1)	(145.0, 1)-s	ample-4	(145.0, 1)-s:	ample-4	(145.0,	[	Die	d 🔽 C	enso	red		In Vivo		
(506.1,1)	(506.1, 1)-s	ample-7	(506.1, 1)-s:	ample-7	(506.1,		_					In Vivo		
(57.8,1)	(57.8, 1)-sa	mple-5	(57.8, 1)-sai	mple-5	(57.8, 1	Tin	ne: 6	05.4				In Vivo		
7	(605.4 0) s	ample-3	(605.4, 0)-s:	ample-3	(605.4,							In Vivo		
(623.0,1)	(623.0, T)-s	ample-8	(623.0, 1)-s:	ample-8	(623.0,							In Vivo		
	▲     OK     Cancel													
	Before click 'next >', Please check the group assignment again!													
	Analysis type: Survival   Next >								Next	>				

Here is what looks like after assigning all the data. Click "Next" button.

Select D	ataset Group Assignm	nents for SAM Tests				o <sup>r</sup>	ø	X	
	Assign Data Sets Into Groups								
<b>%</b>	Image: Groups:     Image: Groups								
*7	Hybridization	SAMPLE 1	Datafile	LABEL 1	ARRAYTYPENAME	ASSAY 1			
🍸 Filter>								-	
(1.3,1)	(1.3, 1)-sample-6	(1.3, 1)-sample-6.txt	SAM_TEST	SAM_7K	In Vivo				
(1151.0,0)	(1151.0, 0)-sample-2	(1151.0, 0)-sample-2	(1151.0, 0)-sample-2.txt	SAM_TEST	SAM_7K	In Vivo			
(1326.3,1)	(1326.3, 1)-sample-1	(1326.3, 1)-sample-1	(1326.3, 1)-sample-1.txt	SAM_TEST	SAM_7K	In Vivo			
(145.0,1)	(145.0, 1)-sample-4	(145.0, 1)-sample-4	(145.0, 1)-sample-4.txt	SAM_TEST	SAM_7K	In Vivo		≡	
(506.1,1)	(506.1, 1)-sample-7	(506.1, 1)-sample-7	(506.1, 1)-sample-7.txt	SAM_TEST	SAM_7K	In Vivo			
(57.8,1)	(57.8, 1)-sample-5	(57.8, 1)-sample-5	(57.8, 1)-sample-5.txt	SAM_TEST	SAM_7K	In Vivo			
(605.4,0)	(605.4, 0)-sample-3	(605.4, 0)-sample-3	(605.4, 0)-sample-3.bt	SAM_TEST	SAM_7K	In Vivo			
(623.0,1)	(623.0, 1)-sample-8	(623.0, 1)-sample-8	(623.0, 1)-sample-8.txt	SAM_TEST	SAM_7K	In Vivo		•	
	•								
	Before click 'next >', Please check the group assignment again!								
		Analysis type: Surviv	/al	▼ Next >	•				

Select Dataset Group Assignments for SAM Tests	- d I
-Survival problem	
Significance Analysis of Microarrays (SAM)	
SAM-Tests Options	These values are default
Number of permutations: Limit to(100~1000): 100	These values are deladit
Estimation of s0 factor:	
Imputation engine: Number of neighbors of KNN 10	
Random number seed: 1234567 Generate Random Seed	
Calculate the fold change's effect on FDR (may take a while)? $\$ $\blacksquare$ No $\$ $\bigcirc$ Yes	
- Filtering with a gene list	
Only include genes from gene list <a>l genes&gt;</a>	
Gene identifiers to include	
Cenbank Acc Cene Mfr ID COUSID UNIGENEID	GENENAME
CLONEID GEN_DESCR_MFR 🗹 REFSEQ 🗹 SPOTID	
Data options	
Dataset Naming	nly)
✓ add sample name(s) to hybridization names Apply log (base 2) to expression values	s
add dye name(s) to hybridization names	
< Back Do Tests	44