

Transcriptomic and proteomic signatures of stemness and differentiation in the colon crypt

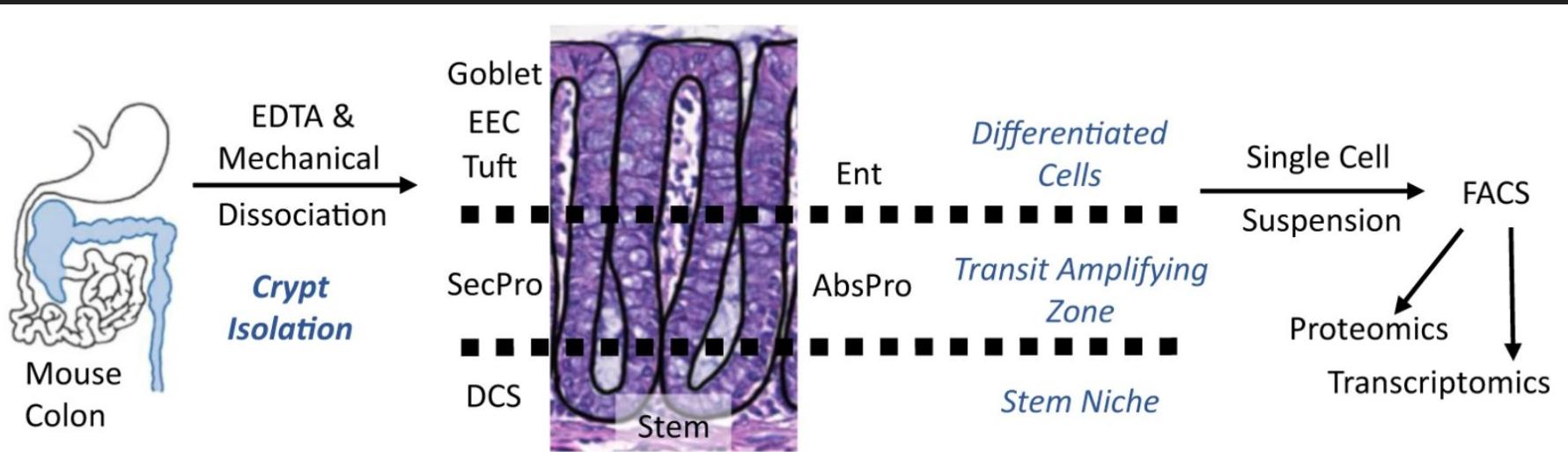
Bryan Jacobs, Alex Borowiec, and Kate Johnson

Background

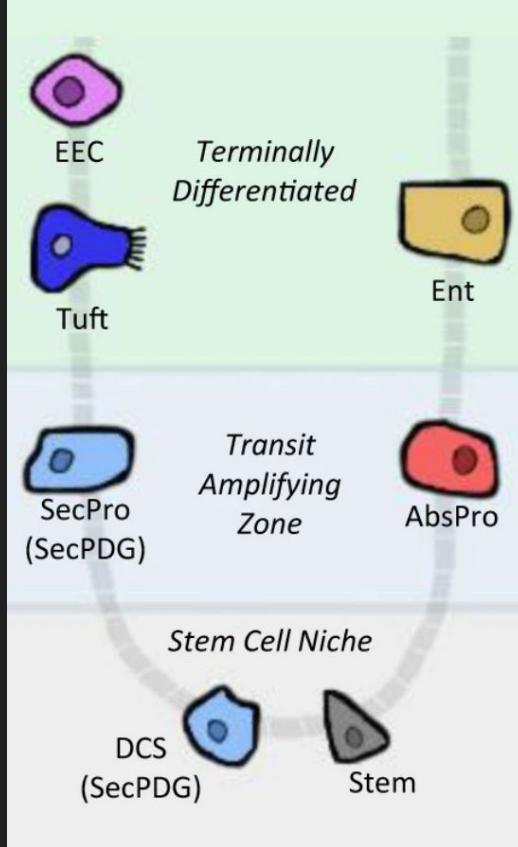
- Colonic epithelium is lined by crypts
- Stem cell niche at the base
- Differentiated cell types face the lumen

b

Cell Sorting Markers	
Dump Channel	Cd45+, Cd45+, Dead Cells
Cell Types Sorted from Live, Epcam+ Cells	
Stem Cells	Cd44highest, Cd24low, cKit-
AbsPro	Cd44med/hi, Cd24low, cKit-
SecPDG	Cd44hi, Cd24med, cKitmed
Tuft	cKit+, Cd24hi
Ent	Cd44low-, Cd24-
EEC	Cd44low-, Cd24+



Background



Author's main comparison
Stem versus progenitor

Our comparison
Stem versus progenitor

HPC Results — Default Parameters



Sample Name	% Aligned	M Aligned
SRR10913363	85.5%	19.4 M
SRR10913362	85.4%	20.5 M
SRR10913356	84.7%	20.5 M
SRR10913357	84.6%	17.5 M
SRR10913361	82.2%	14.1 M
SRR10913360	82.1%	16.3 M
SRR10913353	81.5%	16.3 M
SRR10913352	81.0%	17.6 M
SRR10913324	75.5%	15.4 M
SRR10913365	74.5%	17.5 M
SRR10913323	74.3%	16.2 M
SRR10913355	74.3%	20.0 M
SRR10913354	74.2%	21.9 M
SRR10913359	74.2%	18.1 M
SRR10913358	73.5%	19.5 M

All group members produced identical results for this

HPC Results — Modified Minimum PHRED Score

Default Parameters

$$Q = -10 \log_{10} P$$

- Confidence measure for accurately identifying the correct nucleotide
- Percent Limit = 40%

Min PHRED = 35

Bryan



HPC Results — Modified Sliding Window

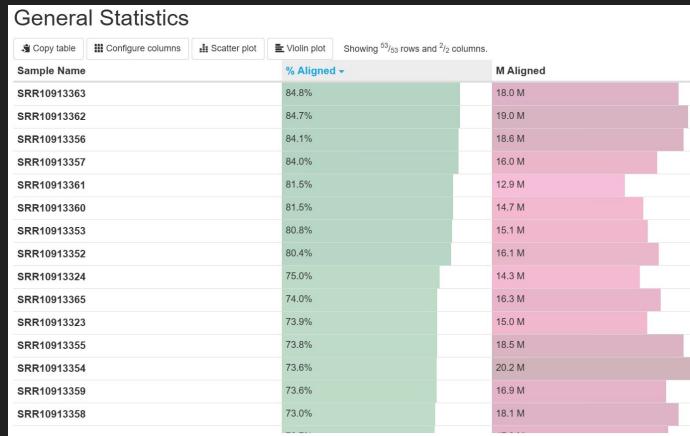
Default
Sliding Window 1



MULTI-MAPPING READS:

Number of reads mapped to multiple loci	1451385
% of reads mapped to multiple loci	6.40%
Number of reads mapped to too many loci	239543
% of reads mapped to too many loci	1.06%

Sliding Window 2



MULTI-MAPPING READS:

Number of reads mapped to multiple loci	1386581
% of reads mapped to multiple loci	6.52%
Number of reads mapped to too many loci	238377
% of reads mapped to too many loci	1.12%

Alex

HPC Results — Modified Max Read Length

Default Parameters

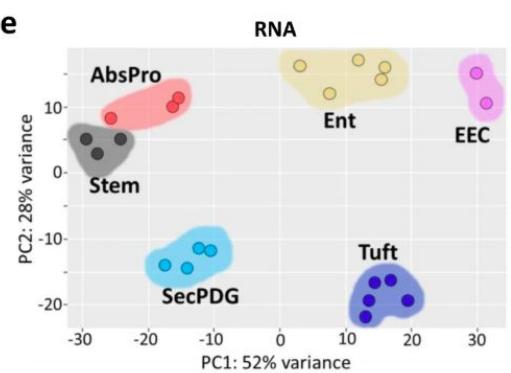


Max read length = 85

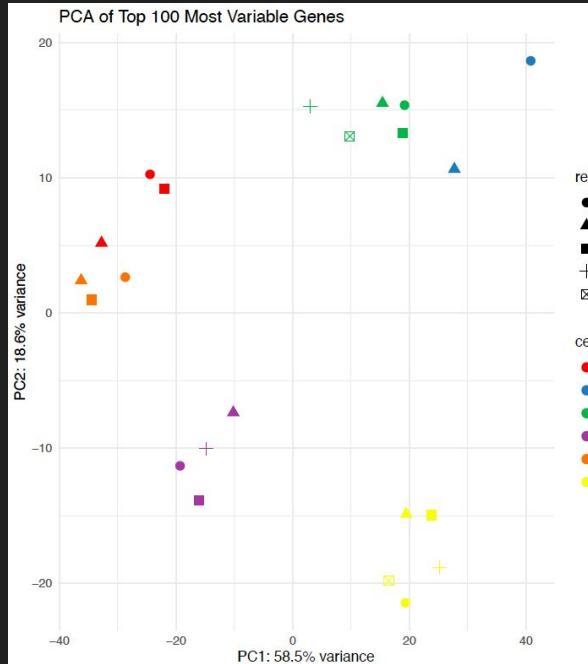
Kate

PCA — Default Parameters

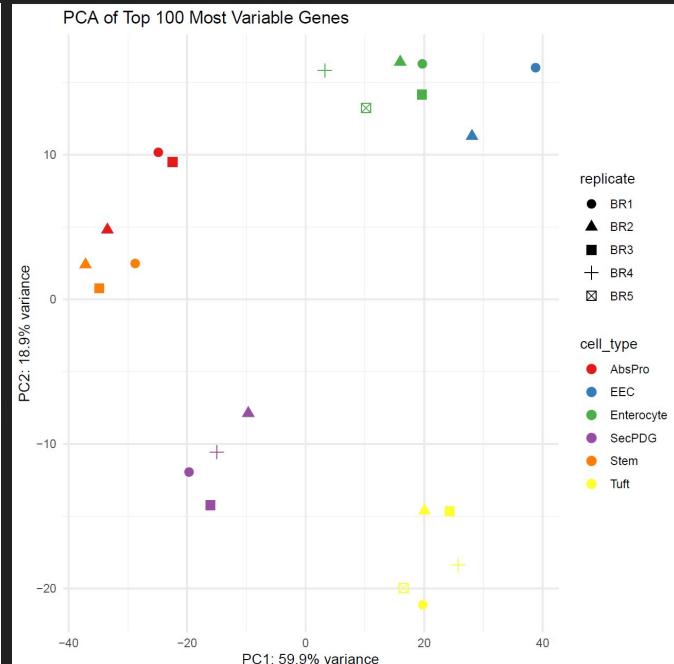
Original



Kate's PCA

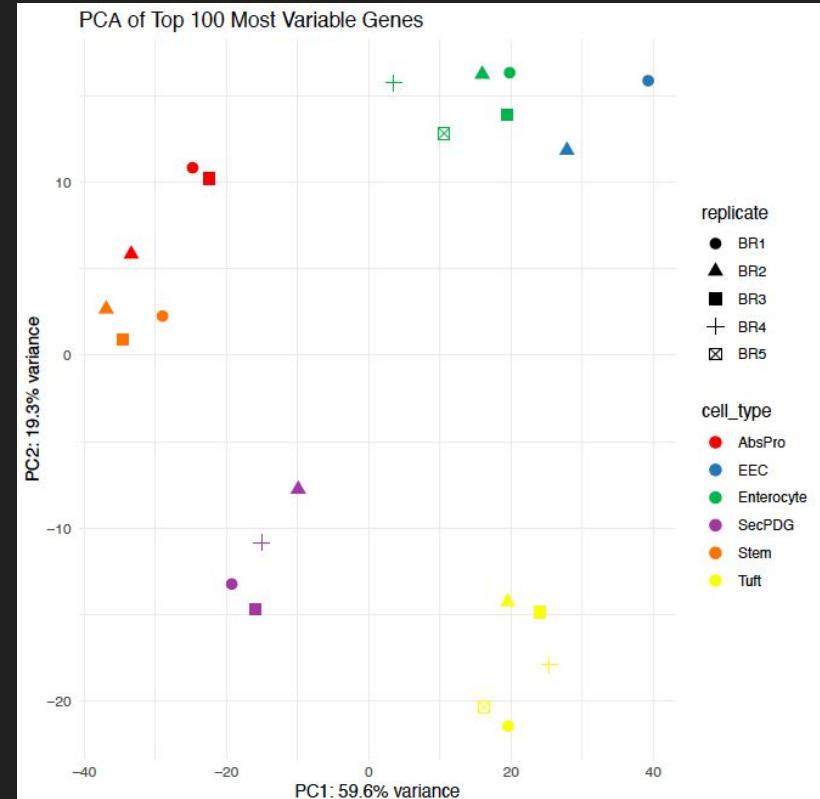
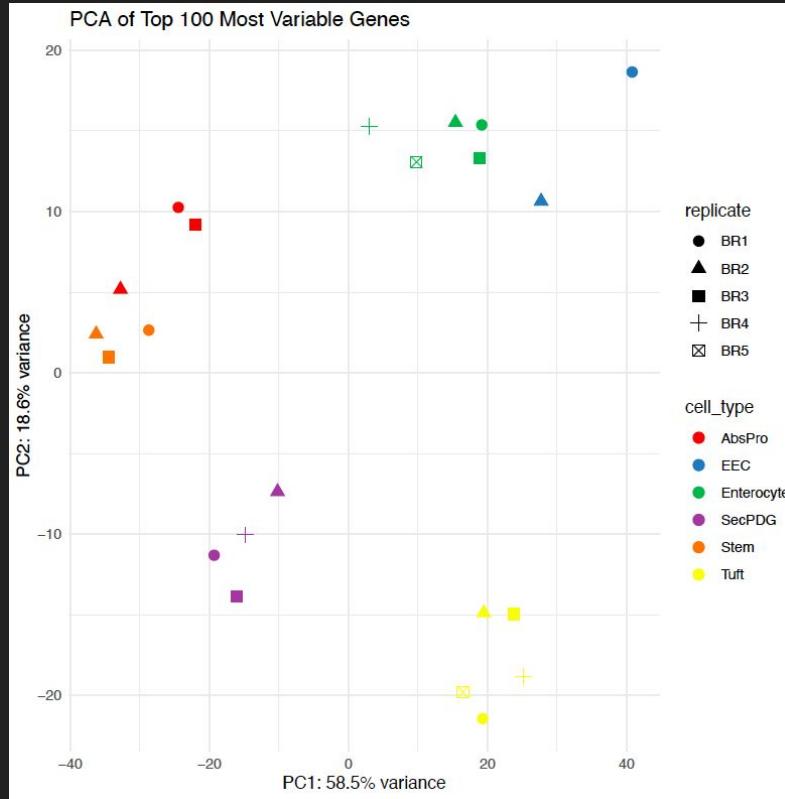


Alex's PCA



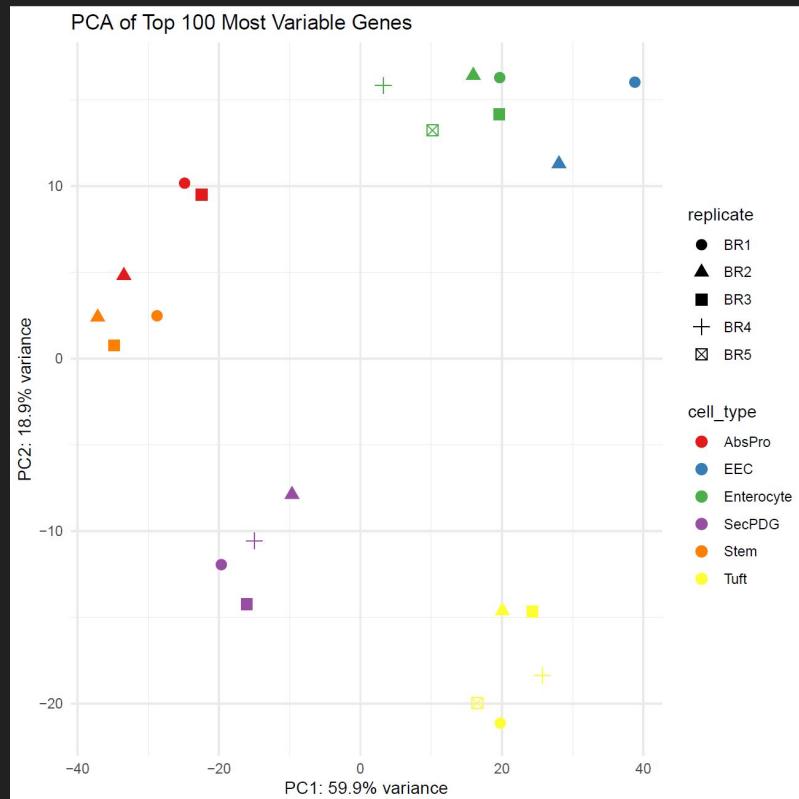
PCA — Modified Max Read Length

Kate

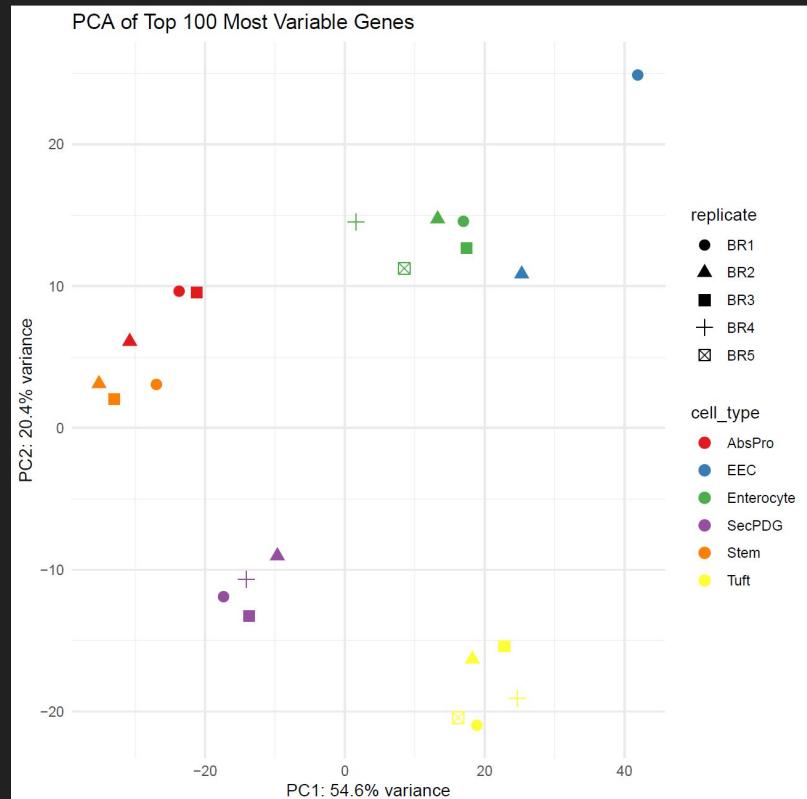


PCA — Modified Sliding Window

Alex



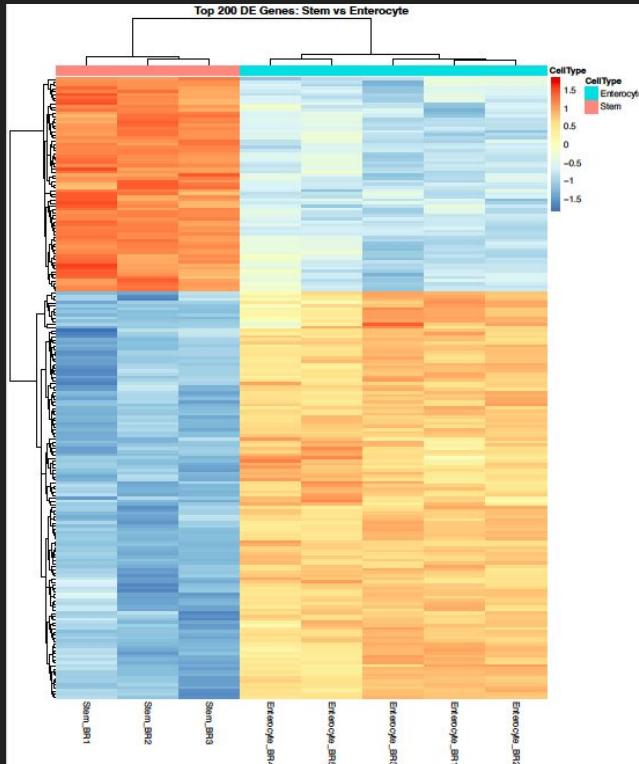
Default Parameters



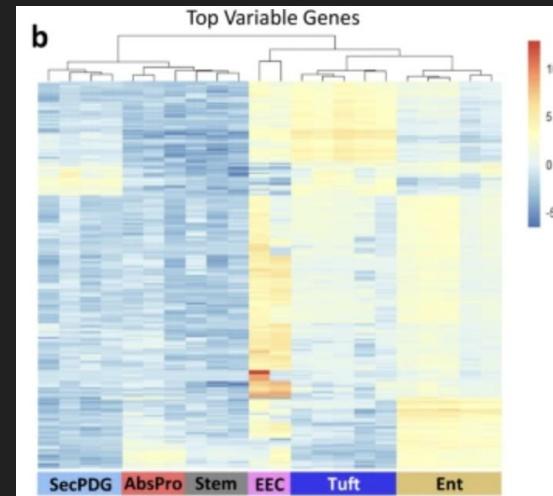
Sliding Window = 2

Heatmap — Default Parameters

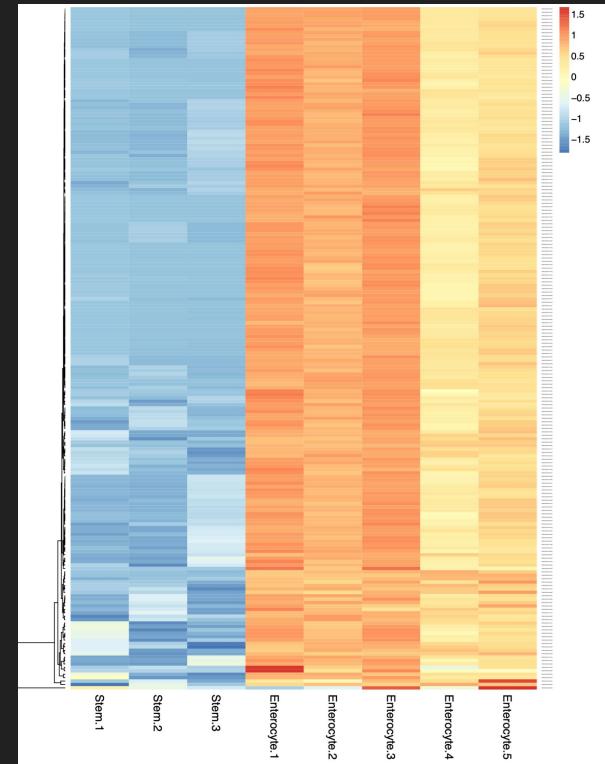
Kate



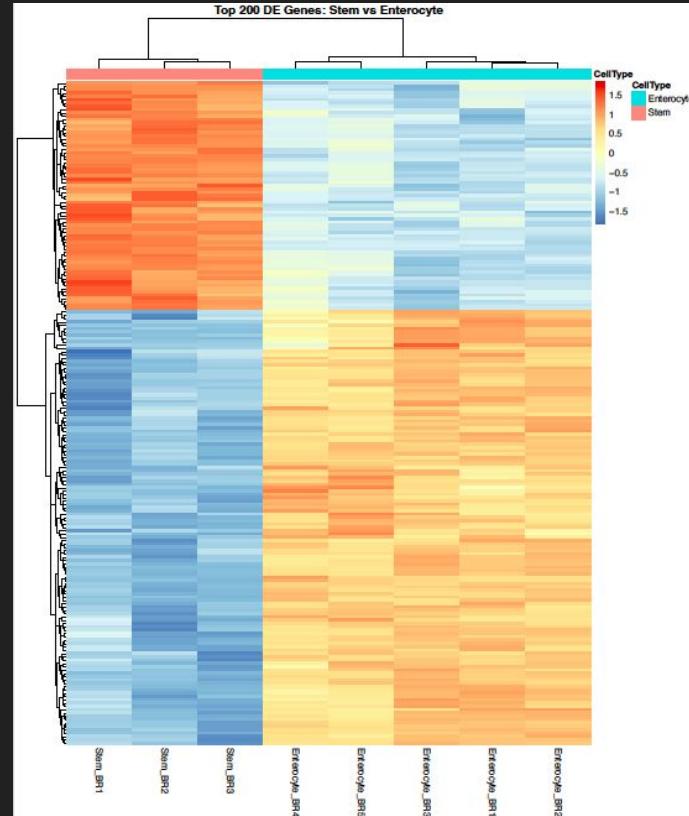
Original



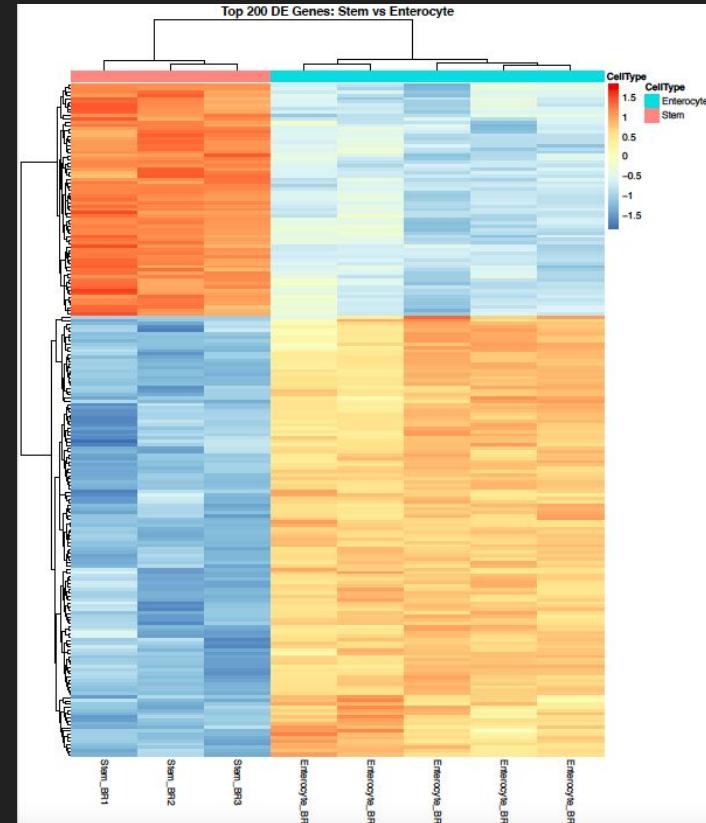
Bryan



Heatmap — Modified Max Read Length

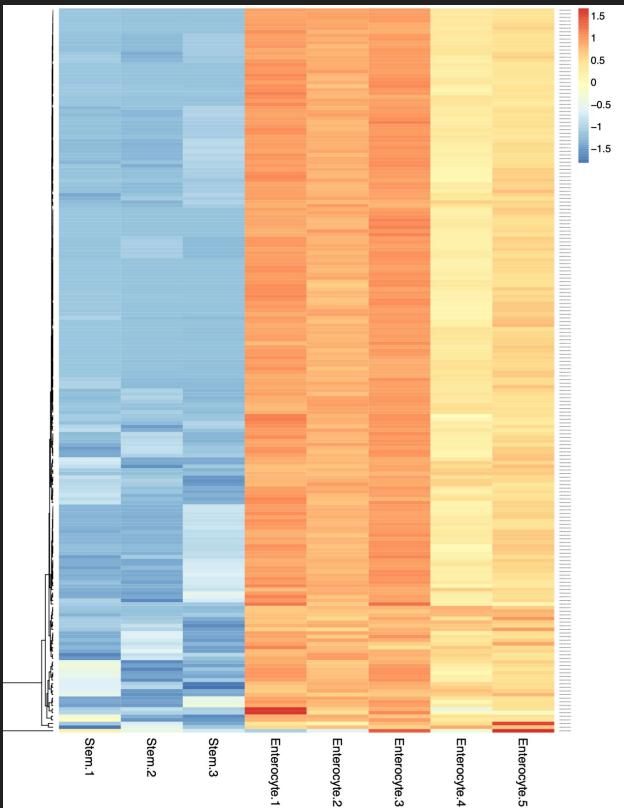


Default Parameters

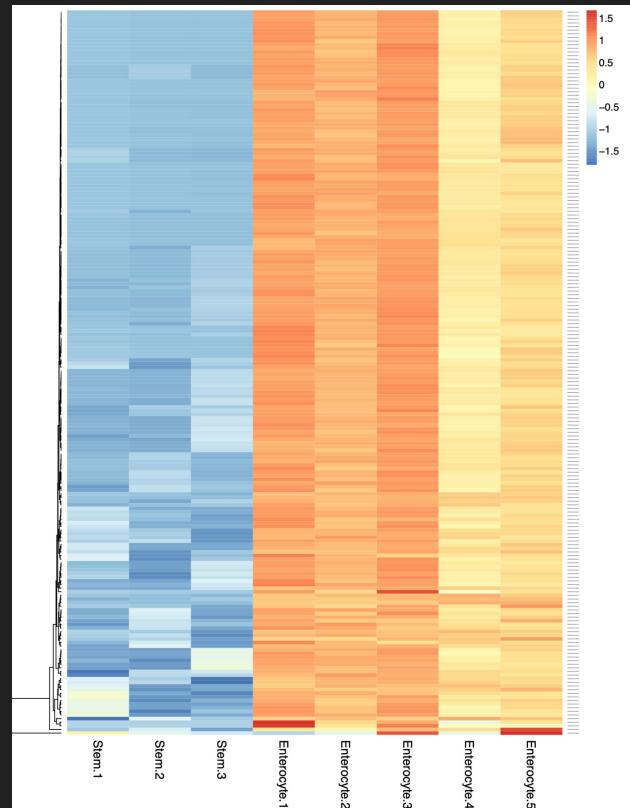


Max Read Length = 85

Heatmap — Modified Minimum PHRED Score Bryan



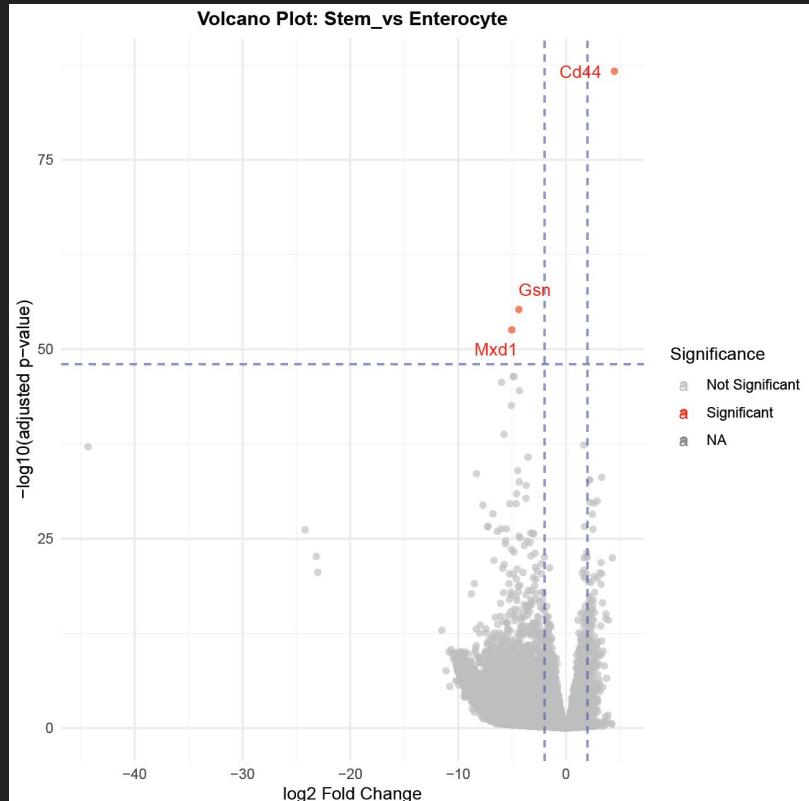
Default Parameters



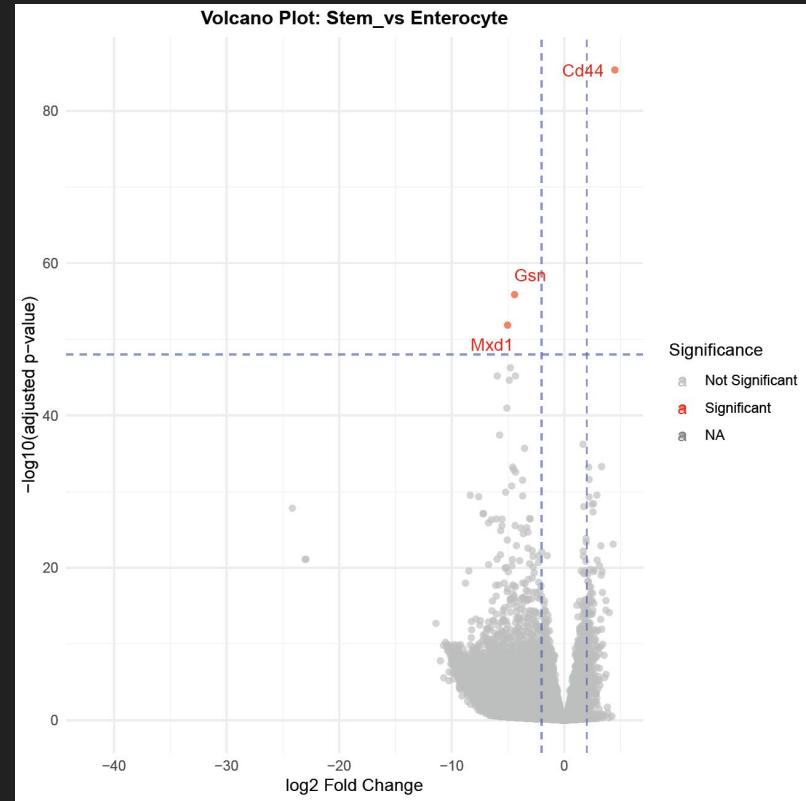
Min PHRED = 35

Volcano Plot — Modified Sliding Window

Alex



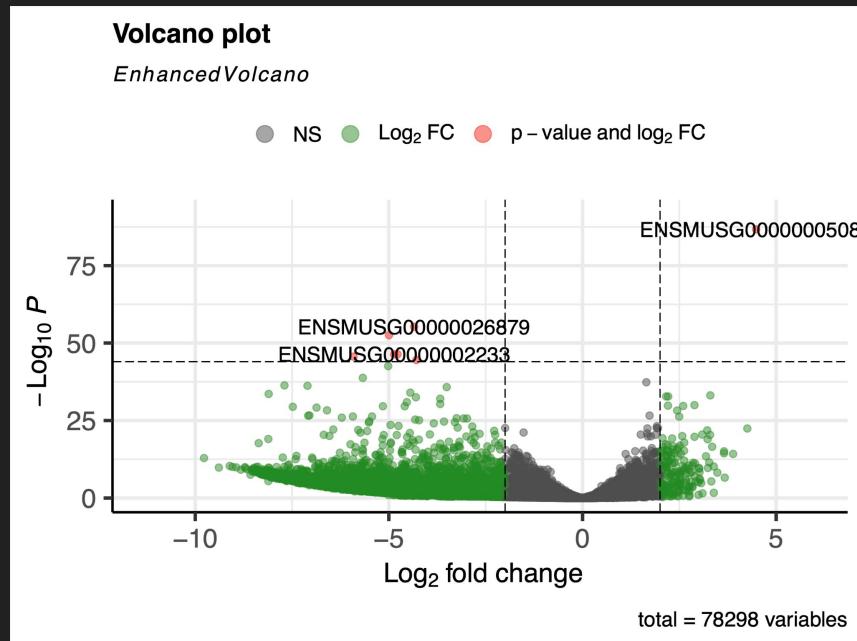
Default Parameters



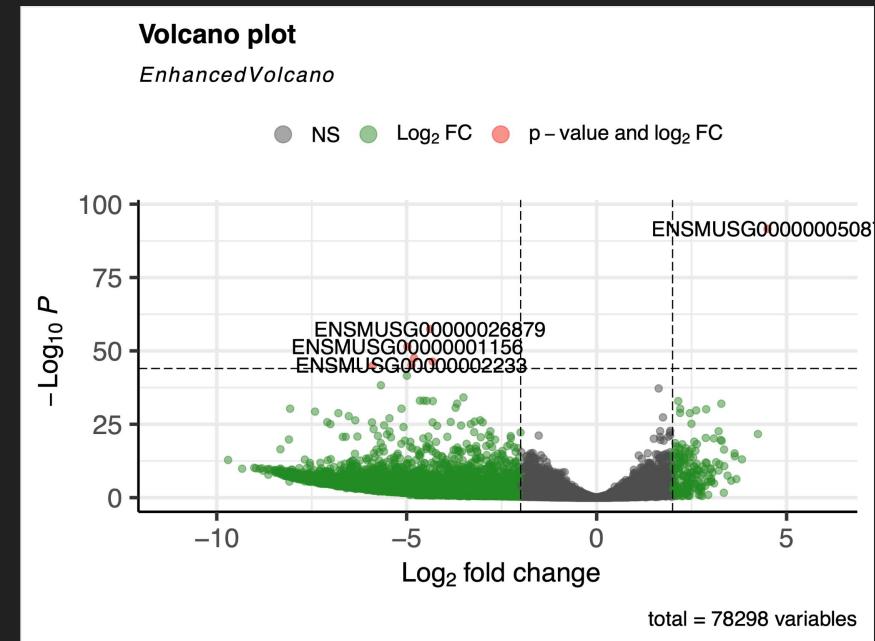
Sliding Window = 2

Volcano Plot — Modified Minimum PHRED Score

Bryan

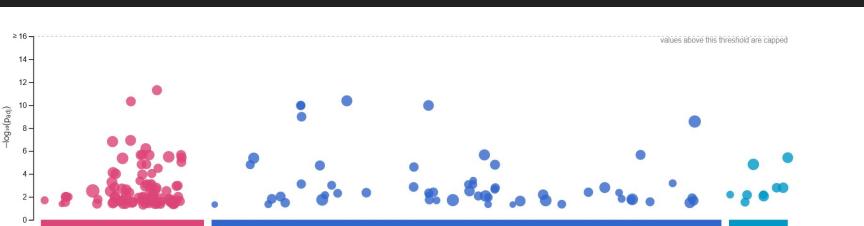
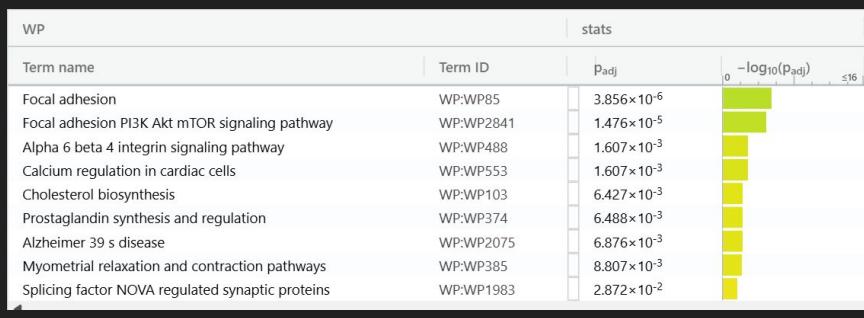
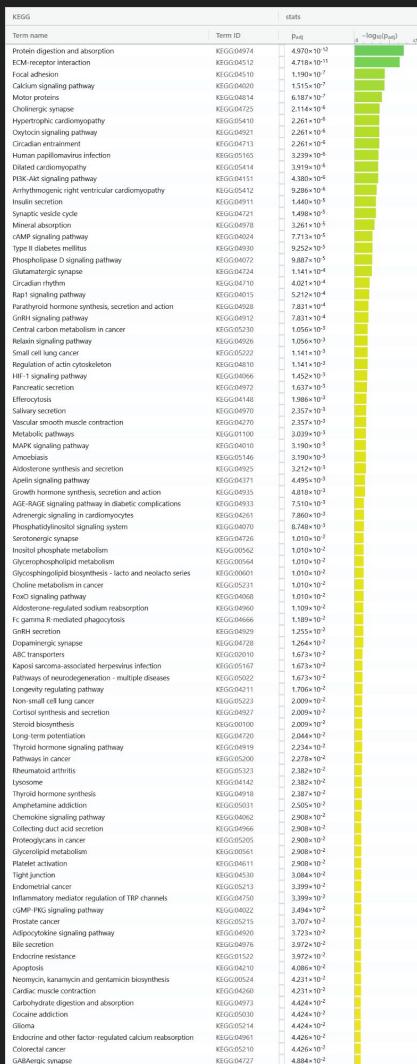


Default Parameters



Min PHRED = 35

Alex - g:Profiler default parameters Downregulated



version
date
organism

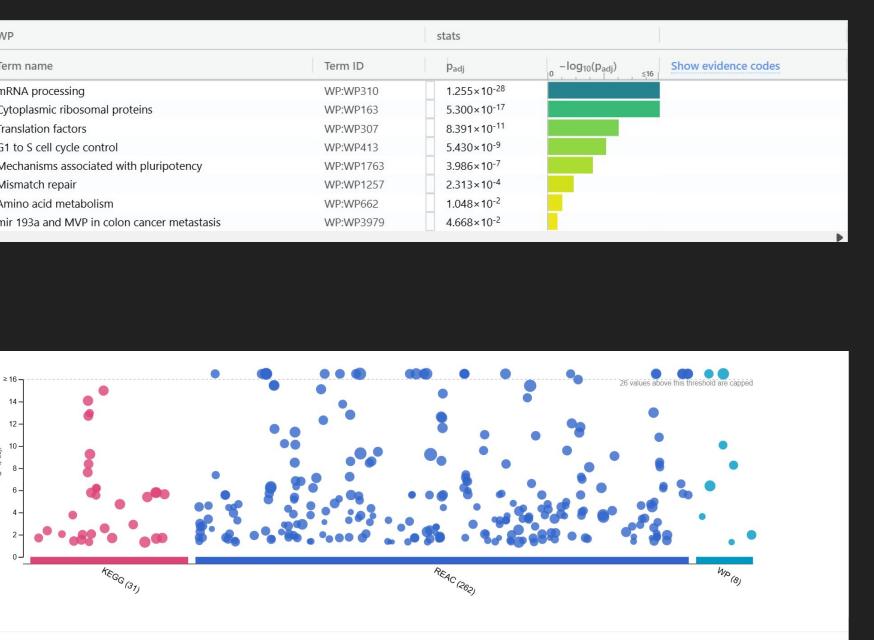
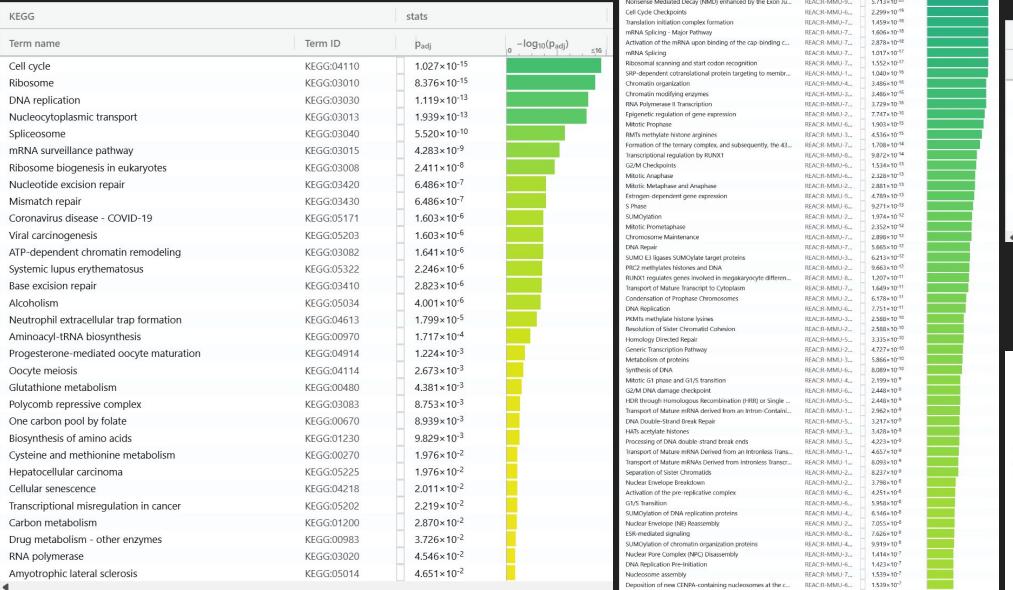
e112_eg59_p19_25aa4782

4/27/2025, 2:59:03 PM

mmusculus

g:Profiler

Alex - g:Profiler default parameter Upregulated

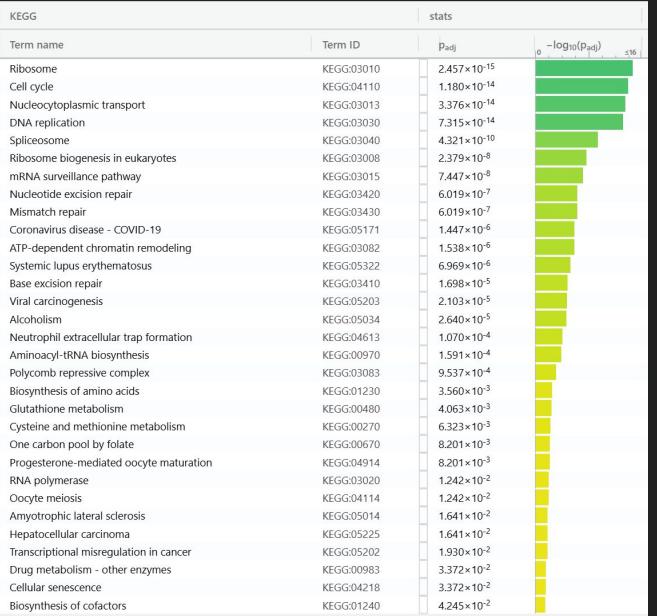


g:Profiler

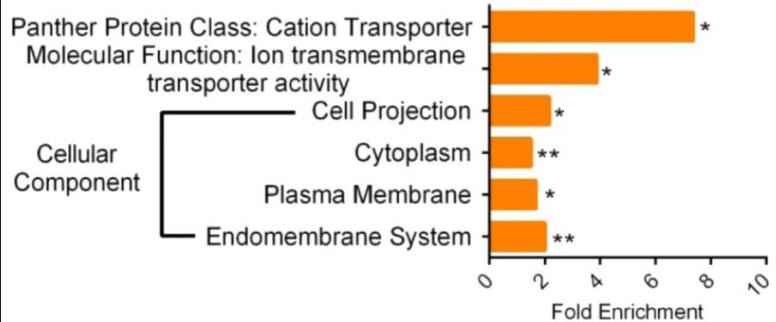
Alex - g:Profiler new parameter Downregulated



Alex - g:Profiler new parameter Upregulated



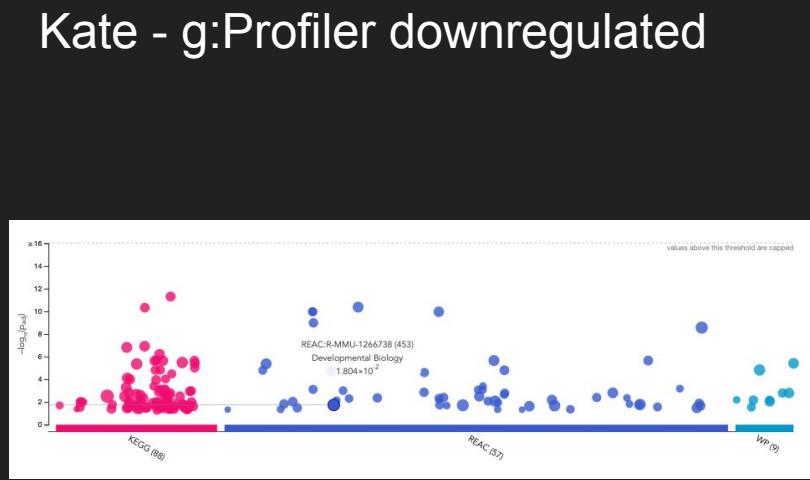
Comparing GO terms from paper



		Mus musculus (REF)		Client Text Box Input (▼ Hierarchy_ NEW! ⓘ)				
PANTHER Protein Class	#	#	expected	Fold Enrichment	+/-	raw P value	FDR	
transporter	1044	15	4.88	3.08	+	9.95E-05	1.96E-02	
Analysis Type:		PANTHER Overrepresentation Test (Released 20240807)						
Annotation Version and Release Date:		PANTHER version 19.0 Released 2024-06-20						
Analyzed List:		Client Text Box Input (Mus musculus)						
Reference List:		Mus musculus (all genes in database)						
Test Type:		FISHER						
Correction:		FDR						
PANTHER GO-Slim Molecular Function		Mus musc	Client Text	Client Text	Client Text	Client Text	Client Text	Client Box Input (FDR)
transition metal ion binding (GO:0046914)		93	4	0.43	+	9.21	9.53E-04	1.92E-01
inorganic anion transmembrane transporter activity (GO:0015103)		94	3	0.44	+	6.83	9.76E-03	2.46E-01
channel activity (GO:0015267)		385	6	1.8	+	3.34	9.43E-03	2.48E-01
Displaying all results; click here to display only significant results								
		Mus musculus (REF)		Client Text Box Input (Hierarchy_ NEW! ⓘ)				
PANTHER GO-Slim Cellular Component	#	#	expected	Fold Enrichment	+/-	raw P value	▲ FDR	
membrane	4688	39	21.90	1.78	+	1.38E-04	6.73E-02	
exocytic vesicle	98	4	.46	8.74	+	1.16E-03	1.42E-01	
secretory vesicle	166	5	.78	6.45	+	1.10E-03	1.79E-01	
neuron projection	385	7	1.80	3.89	+	2.21E-03	1.80E-01	
cytoplasm	5502	40	25.70	1.56	+	1.86E-03	1.82E-01	
Golgi membrane	88	4	.41	9.73	+	7.75E-04	1.89E-01	
transport vesicle	128	4	.60	6.69	+	3.07E-03	2.15E-01	
endomembrane system	1426	15	6.66	2.25	+	3.53E-03	2.16E-01	
plasma membrane bounded cell projection	667	9	3.12	2.89	+	4.03E-03	2.19E-01	
cell projection	690	9	3.22	2.79	+	5.02E-03	2.45E-01	

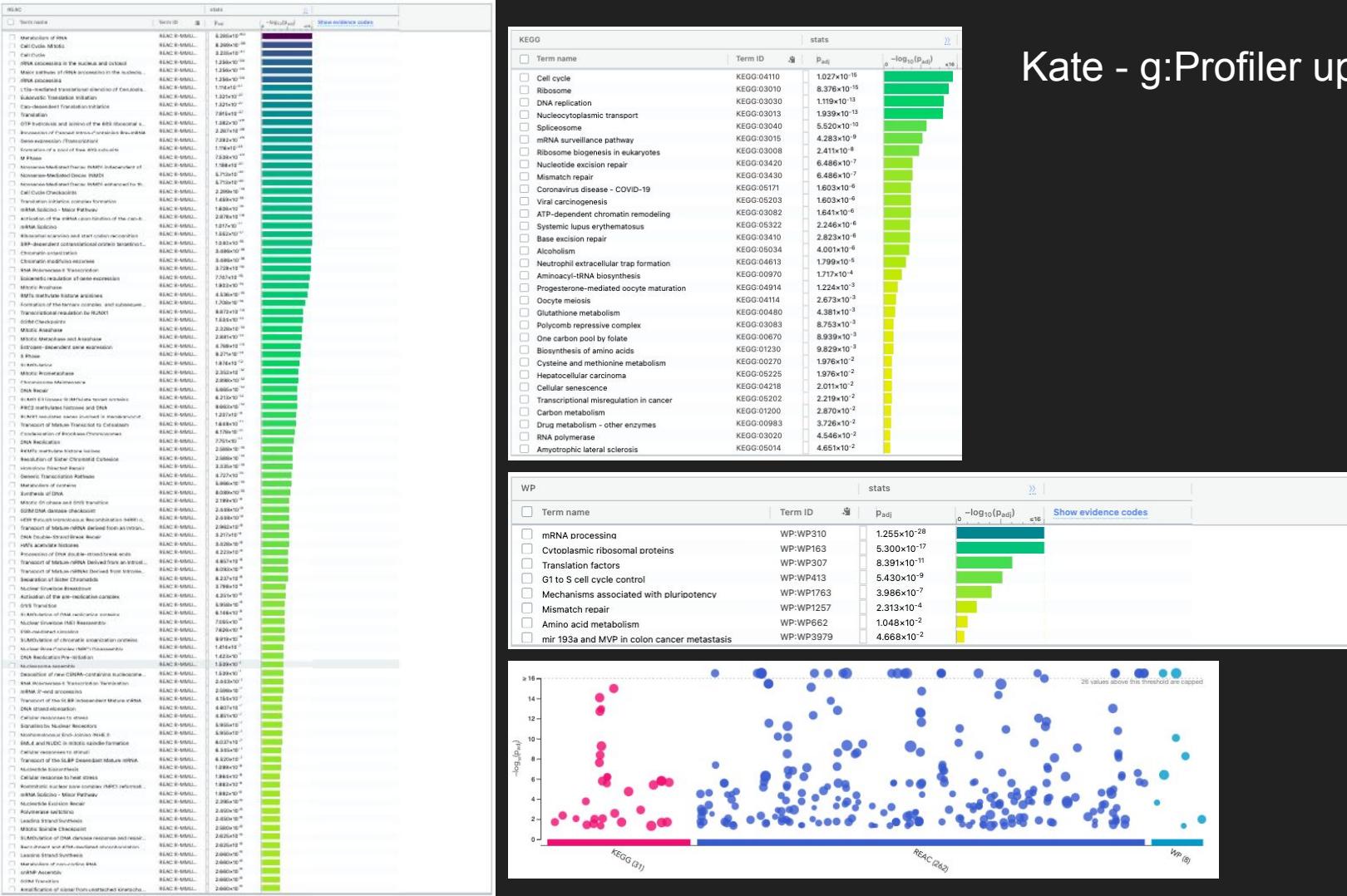
All group members worked on this analysis

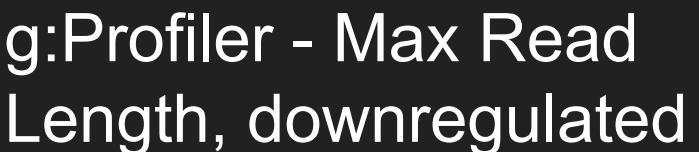
REAC		stats	
Term name	Term ID	P-value	-log ₁₀ (P-value)
Extracellular matrix organization	REACR-MMU-...	4.195x10 ⁻¹¹	
Collagen chain trimerization	REACR-MMU-...	1.071x10 ⁻¹²	
Ion channel transport	REACR-MMU-...	1.071x10 ⁻¹²	
Collagen biosynthesis and modifying enzymes	REACR-MMU-...	1.071x10 ⁻¹²	
Collagen formation	REACR-MMU-...	1.027x10 ⁻¹⁰	
Transport of small molecules	REACR-MMU-...	2.888x10 ⁻⁶	
Stimuli-sensing channels	REACR-MMU-...	2.184x10 ⁻⁴	
Nervous system development	REACR-MMU-...	2.187x10 ⁻⁶	
Axon guidance	REACR-MMU-...	4.218x10 ⁻⁶	
D-linked glycosylation	REACR-MMU-...	1.575x10 ⁻⁶	
Assembly of collagen fibrils and other multimeric a...	REACR-MMU-...	1.575x10 ⁻⁶	
Decoration of the extracellular matrix	REACR-MMU-...	1.839x10 ⁻⁶	
Interactin cell surface interactions	REACR-MMU-...	2.503x10 ⁻⁵	
NCAM1 interactions	REACR-MMU-...	3.885x10 ⁻⁴	
Termination of D-glycan biosynthesis	REACR-MMU-...	6.573x10 ⁻⁶	
Collagen degradation	REACR-MMU-...	7.836x10 ⁻⁴	
NCAM signaling for neurite out-growth	REACR-MMU-...	8.334x10 ⁻⁴	
Molecules associated with elastic fibres	REACR-MMU-...	8.334x10 ⁻⁴	
ECM proteolysis	REACR-MMU-...	9.885x10 ⁻⁴	
Integration of energy metabolism	REACR-MMU-...	1.382x10 ⁻³	
SLC-mediated transmembrane transport	REACR-MMU-...	1.556x10 ⁻³	
D-linked glycosylation of mucins	REACR-MMU-...	1.651x10 ⁻³	
D-glycosylation of TSR domain-containing proteins	REACR-MMU-...	2.226x10 ⁻³	
Muscle contraction	REACR-MMU-...	3.177x10 ⁻³	
Regulation of insulin secretion	REACR-MMU-...	3.975x10 ⁻³	
LYCAM interactions	REACR-MMU-...	3.975x10 ⁻³	
G alpha (12/13) signalling events	REACR-MMU-...	4.345x10 ⁻³	
Serotonin Neurotransmitter Release Cycle	REACR-MMU-...	4.345x10 ⁻³	
Ion homeostasis	REACR-MMU-...	4.835x10 ⁻³	
Elastic fibre formation	REACR-MMU-...	4.912x10 ⁻³	
RAC1 GTPase cycle	REACR-MMU-...	6.496x10 ⁻³	
Invasodrome formation	REACR-MMU-...	6.810x10 ⁻³	
Dopamine Neurotransmitter Release Cycle	REACR-MMU-...	7.181x10 ⁻³	
NRAGE signals death through JNK	REACR-MMU-...	8.392x10 ⁻³	
Neuronal System	REACR-MMU-...	8.392x10 ⁻³	
Cardiac conduction	REACR-MMU-...	9.487x10 ⁻³	
Non-intern membrane-ECM interactions	REACR-MMU-...	1.128x10 ⁻²	
Transport of bile salts and organic acids, metal ion...	REACR-MMU-...	1.295x10 ⁻²	
CDC42 GTPase cycle	REACR-MMU-...	1.496x10 ⁻²	
Signaling through L1	REACR-MMU-...	1.496x10 ⁻²	
Signaling by Receptor Tyrosine Kinases	REACR-MMU-...	1.656x10 ⁻²	
Developmental Biology	REACR-MMU-...	1.804x10 ⁻²	
Ion transport by P-type ATPases	REACR-MMU-...	1.804x10 ⁻²	
Signaling by PDGF	REACR-MMU-...	1.835x10 ⁻²	
Metabolism of Iodois	REACR-MMU-...	1.931x10 ⁻²	
Lewis blood group biosynthesis	REACR-MMU-...	2.066x10 ⁻²	
RHO GTPase cycle	REACR-MMU-...	2.082x10 ⁻²	
Transport of inorganic cations/anions and amino a...	REACR-MMU-...	2.151x10 ⁻²	
Phospholipid metabolism	REACR-MMU-...	2.345x10 ⁻²	
Synthesis of PIPs at the plasma membrane	REACR-MMU-...	2.705x10 ⁻²	
Transmission across Chemical Synapses	REACR-MMU-...	3.279x10 ⁻²	
Cell death signaling via NRAGE, NRIF and NADE	REACR-MMU-...	3.279x10 ⁻²	
Blood vessel smooth muscle biosynthesis	REACR-MMU-...	4.384x10 ⁻²	
RDS and IRS induction in photoreceptors	REACR-MMU-...	4.384x10 ⁻²	
Nitric oxide stimulates quinolinate cyclase	REACR-MMU-...	4.684x10 ⁻²	
PTK8 promotes HIF1A stabilization	REACR-MMU-...	4.734x10 ⁻²	
AMPK inhibits cREBP transcriptional activation ac...	REACR-MMU-...	4.734x10 ⁻²	



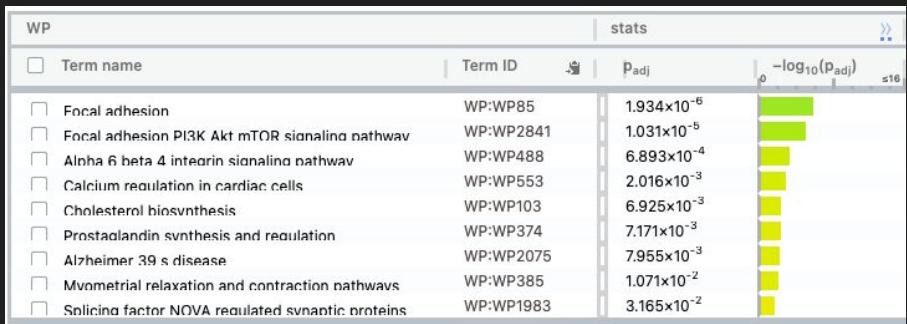
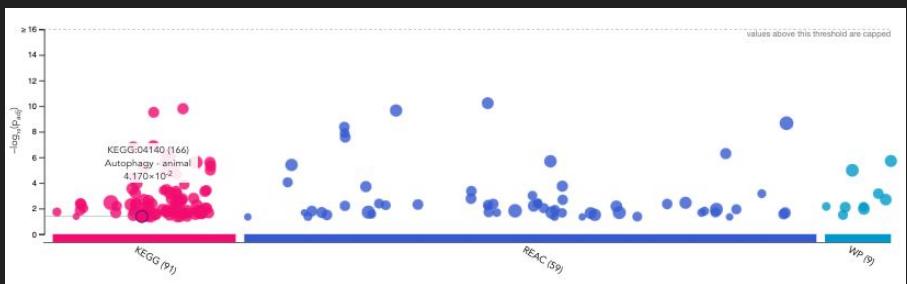
WP	Term name	Term ID	Pedi	$-\log_{10}(P_{adj})$	Show evidence codes
<input type="checkbox"/>	Focal adhesion	WPWP85	3.856e-06	4.8	
<input type="checkbox"/>	Focal adhesion PI3K Akt mTOR signaling pathway	WPWP2841	1.476e-05	4.6	
<input type="checkbox"/>	Alpha 6 beta 4 integrin signaling pathway	WPWP488	1.607e-03	4.4	
<input type="checkbox"/>	Calcium regulation in cardiac cells	WPWP553	1.607e-03	4.4	
<input type="checkbox"/>	Cholesterol biosynthesis	WPWP103	6.427e-03	4.2	
<input type="checkbox"/>	Prostaglandin synthesis and regulation	WPWP374	6.498e-03	4.2	
<input type="checkbox"/>	Alzheimer 39 + disease	WPWP2075	6.876e-03	4.1	
<input type="checkbox"/>	Mymetrical relaxation and contraction pathways	WPWP385	8.807e-03	4.0	
<input type="checkbox"/>	Spliginic factor.Nova ren listed synaptic proteins	WPWP1983	2.872e-02	3.9	

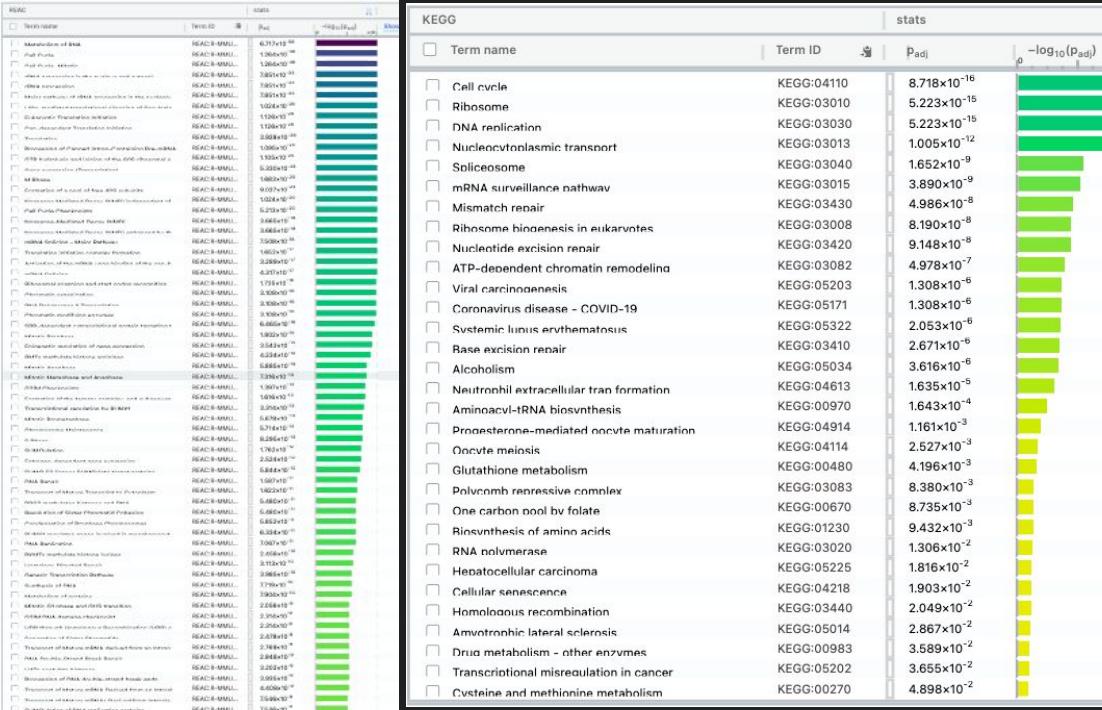
Kate - g:Profiler upregulated





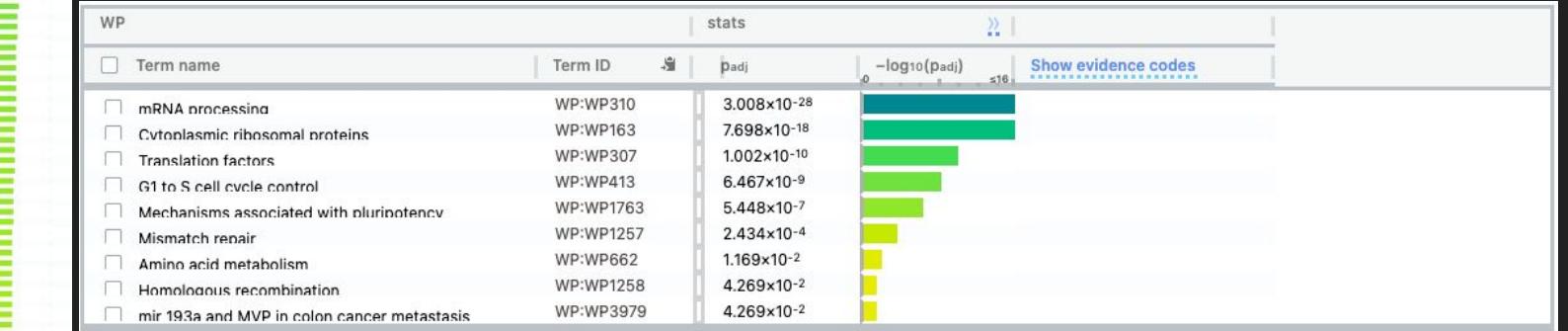
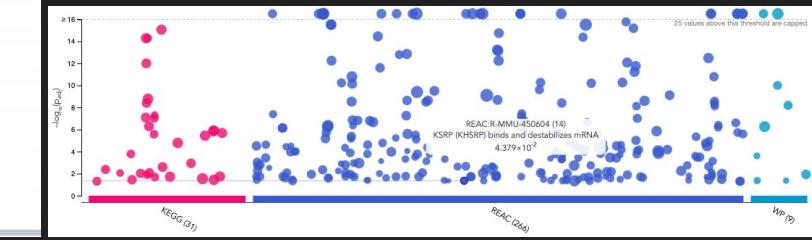
Kate





g:Profiler - Max Read Length, upregulated

Kate



GSEA — Modified Minimum PHRED Score

Bryan

Stem cells

Enterocytes

Enrichment in phenotype: 0 (3 samples)

- 291 / 807 gene sets are upregulated in phenotype **0**
- 280 gene sets are significant at FDR < 25%
- 45 gene sets are significantly enriched at nominal pvalue < 1%
- 45 gene sets are significantly enriched at nominal pvalue < 5%
- [Snapshot](#) of enrichment results
- Detailed [enrichment results in html](#) format
- Detailed [enrichment results in TSV](#) format (tab delimited text)
- [Guide to interpret results](#)

Enrichment in phenotype: 4 (5 samples)

- 516 / 807 gene sets are upregulated in phenotype **4**
- 105 gene sets are significantly enriched at FDR < 25%
- 21 gene sets are significantly enriched at nominal pvalue < 1%
- 52 gene sets are significantly enriched at nominal pvalue < 5%
- [Snapshot](#) of enrichment results
- Detailed [enrichment results in html](#) format
- Detailed [enrichment results in TSV](#) format (tab delimited text)
- [Guide to interpret results](#)

Default Parameters

Enrichment in phenotype: 0 (3 samples)

- 287 / 807 gene sets are upregulated in phenotype **0**
- 277 gene sets are significant at FDR < 25%
- 53 gene sets are significantly enriched at nominal pvalue < 1%
- 53 gene sets are significantly enriched at nominal pvalue < 5%
- [Snapshot](#) of enrichment results
- Detailed [enrichment results in html](#) format
- Detailed [enrichment results in TSV](#) format (tab delimited text)
- [Guide to interpret results](#)

Enrichment in phenotype: 4 (5 samples)

- 520 / 807 gene sets are upregulated in phenotype **4**
- 118 gene sets are significantly enriched at FDR < 25%
- 19 gene sets are significantly enriched at nominal pvalue < 1%
- 52 gene sets are significantly enriched at nominal pvalue < 5%
- [Snapshot](#) of enrichment results
- Detailed [enrichment results in html](#) format
- Detailed [enrichment results in TSV](#) format (tab delimited text)
- [Guide to interpret results](#)

Min PHRED = 35

GSEA — Modified Minimum PHRED Score (cont.)

Bryan

Stem

Enterocyte

Default
Parameters

	GS follow link to MsigDB									
	GS DETAILS	SIZE	ES	NES	NOM P-VAL	FDR P-VAL	RANK	MAX	LEADING EDGE	
1 REACTOME_DEADENYLATION_OF_MRNA	Details... 23	0.88	3.36	0.000	0.000	0.000	2548	tags=10%, list=3%, signal=1%		
2 REACTOME_PROCESSING_OF_INTRONLESS_PRE_MRNAS	Details... 20	0.90	3.21	0.000	0.000	0.000	5068	tags=10%, list=1%, signal=11%		
3 REACTOME_KRPP_KHSPB BINDS AND DESTABILIZES_MRNA	Details... 10	0.81	3.25	0.000	0.000	0.000	4020	tags=10%, list=1%, signal=11%		
4 REACTOME_PONA_DEPENDENT_LONG_PATCH_BASE_EXCISION_REFINER	Details... 21	0.87	3.19	0.000	0.000	0.000	3564	tags=10%, list=1%, signal=1%		
5 REACTOME_MRNA_CAPPING	Details... 20	0.70	3.14	0.000	0.000	0.000	5125	tags=10%, list=1%, signal=1%		
6 REACTOME_PHOSPHORYLATION_OF_THE_APC_C	Details... 20	0.78	3.06	0.000	0.000	0.000	9265	tags=10%, list=1%, signal=1%		
7 REACTOME_DNA_STRAND_ELONGATION	Details... 24	0.91	3.00	0.000	0.000	0.000	2759	tags=10%, list=1%, signal=1%		
8 REACTOME_BUTYRATE_RESPONSE_FACTOR_1_RF1 BINDS AND DESTABILIZES_MRNA	Details... 17	0.85	2.96	0.000	0.000	0.000	4820	tags=10%, list=1%, signal=1%		
9 REACTOME_PROGRESSIVE_SYNTHESIS_ON_THE_LAGGING_STRA	Details... 15	0.91	2.91	0.000	0.000	0.000	3729	tags=10%, list=1%, signal=1%		
10 REACTOME_PROGRESSIVE_SYNTHESIS_ON_C_STRAAND_OF_THE_TELOMERE	Details... 18	0.82	2.76	0.000	0.000	0.000	2471	tags=10%, list=1%, signal=1%		
11 REACTOME_MISMATCH_REPAIR	Details... 15	0.87	2.71	0.000	0.000	0.000	3471	tags=10%, list=1%, signal=1%		
12 REACTOME_FORMATION_OF_SPLICENCE_ASSOCIATED_HETERODIMERIC_POU_SA	Details... 16	0.86	2.64	0.000	0.000	0.000	3319	tags=10%, list=1%, signal=1%		
13 REACTOME_FORMATION_OF_ATP_BY_CHLOROMIC_COUP	Details... 20	0.88	2.51	0.000	0.000	0.000	18034	tags=10%, list=1%, signal=1%		
14 REACTOME_MRNA_GROWTH_BY_3_TO_5_EXORIBONUCLEASE	Details... 16	0.90	2.50	0.000	0.000	0.000	5020	tags=10%, list=1%, signal=1%		
15 REACTOME_INITIATION_OF_NUCLEAR_ENVELOPE_NE_INFORMATION	Details... 17	0.79	2.55	0.000	0.000	0.000	3237	tags=10%, list=1%, signal=1%		

	GS follow link to MsigDB									
	GS DETAILS	SIZE	ES	NES	NOM P-VAL	FDR P-VAL	RANK	MAX	LEADING EDGE	
1 REACTOME_CLASS_I_PEROXISOMAL_MEMBRANE_PROTEIN_IMPORT	Details... 19	0.81	1.67	0.000	0.000	0.010	1693	tags=11%, list=3%, signal=1%		
2 REACTOME_SYNTHESIS_OF_BILE_ACIDS_AND_BILE_SALTS_VIA_7ALPHA_HYDROXYCHOLESTEROL	Details... 22	-0.70	-1.46	0.000	0.002	0.419	6416	tags=36%, list=1%, signal=41%		
3 REACTOME_NEGATIVE_REGULATION_OF_FLTs	Details... 15	-0.72	-1.44	0.004	0.003	0.709	5403	tags=13%, list=10%, signal=15%		
4 REACTOME_SYNTHESIS_OF_BILE_ACIDS_AND_BILE_SALTS	Details... 32	-0.66	-1.41	0.003	0.006	0.885	6417	tags=16%, list=6%, signal=2%		
5 REACTOME_FATTY_ACIDS	Details... 22	-0.68	-1.41	0.008	0.006	0.685	13495	tags=73%, list=24%, signal=8%		
6 REACTOME_Olfactory_SIGNALING_PATHWAY	Details... 72	-0.62	-1.39	0.000	0.008	0.941	13764	tags=69%, list=24%, signal=1%		
7 REACTOME_WNT_LIGAND_BIOGENESIS_AND_TRAFFICKING	Details... 25	-0.64	-1.35	0.009	0.014	0.998	14637	tags=60%, list=20%, signal=7%		
8 REACTOME_AMINE_LIGAND_BINDING_RECEPORS	Details... 43	-0.61	-1.32	0.003	0.023	1.000	18467	tags=74%, list=33%, signal=11%		
9 REACTOME_PL3K CASCADE_FGFR1	Details... 21	-0.63	-1.32	0.041	0.024	1.000	16334	tags=57%, list=29%, signal=1%		
10 REACTOME_FORMATION_OF_THE_CORNIFIED_ENVELOPE	Details... 90	-0.58	-1.32	0.001	0.025	1.000	17021	tags=40%, list=30%, signal=6%		
11 REACTOME_GLYCOCYANATE_METABOLISM_AND_GLYCINE_DEGRADATION	Details... 18	-0.64	-1.32	0.029	0.025	1.000	12356	tags=39%, list=22%, signal=10%		
12 REACTOME_PHOSPHOLIPASE_C_MEDIATED CASCADE_FGFR1	Details... 16	-0.65	-1.32	0.032	0.026	1.000	16334	tags=76%, list=29%, signal=10%		
13 REACTOME_FGFR1_LIGAND_BINDING_AND_ACTIVATION	Details... 17	-0.65	-1.31	0.027	0.026	1.000	12356	tags=76%, list=29%, signal=10%		
14 REACTOME_NEUREXINS_AND_NEUROLIGINS	Details... 31	-0.61	-1.31	0.015	0.029	1.000	9598	tags=39%, list=17%, signal=17%		
15 REACTOME_EICOSANOIDS	Details... 17	-0.65	-1.30	0.026	0.030	1.000	13495	tags=71%, list=24%, signal=6%		

	GS follow link to MsigDB									
	GS DETAILS	SIZE	ES	NES	NOM P-VAL	FDR P-VAL	RANK	MAX	LEADING EDGE	
1 REACTOME_POSTMEiotic_NUCLEAR_POLE_COMPLEX_NEC_INFORMATION	Details... 22	0.60	4.19	0.000	0.000	0.000	3624	tags=10%, list=2%, signal=1%		
2 REACTOME_PHOSPHORYLATION_OF_THE_AP_C	Details... 20	0.76	3.86	0.000	0.000	0.000	5275	tags=10%, list=1%, signal=1%		
3 REACTOME_PROCESSING_OF_INTRONLESS_PRE_MRNAS	Details... 20	0.80	3.80	0.000	0.000	0.000	5455	tags=10%, list=1%, signal=1%		
4 REACTOME_DNA_STRAND_ELONGATION	Details... 24	0.92	3.28	0.000	0.000	0.000	2523	tags=10%, list=1%, signal=1%		
5 REACTOME_MRNA_DECAY_BY_3'_5' EXORIBONUCLEASE	Details... 18	0.91	3.20	0.000	0.000	0.000	5983	tags=10%, list=1%, signal=1%		
6 REACTOME_LAGGING_STRAAND_SYNTHESIS	Details... 20	0.89	3.15	0.000	0.000	0.000	2923	tags=10%, list=1%, signal=1%		
7 REACTOME_TRANSITION_SYNTHESIS_BY_POLI	Details... 20	0.80	3.10	0.000	0.000	0.000	3461	tags=10%, list=1%, signal=1%		
8 REACTOME_BETA_GATCIN_PHOSPHORYLATION CASCADE	Details... 17	0.85	3.06	0.000	0.000	0.000	4564	tags=10%, list=1%, signal=1%		
9 REACTOME_KRPP_KHSPB BINDS AND DESTABILIZES_MRNA	Details... 15	0.91	3.06	0.000	0.000	0.000	4845	tags=10%, list=1%, signal=1%		
10 REACTOME_RIBOTIDE_ATPASE_CYCLE	Details... 22	0.86	2.94	0.000	0.000	0.000	5055	tags=10%, list=1%, signal=1%		
11 REACTOME_PROGRESSIVE_SYNTHESIS_ON_C_STRAAND_OF_THE_TELOMERE	Details... 18	0.94	2.85	0.000	0.000	0.000	3255	tags=10%, list=1%, signal=1%		
12 REACTOME_PROTEIN_HYDROXYLATION	Details... 18	0.89	2.80	0.000	0.000	0.000	3497	tags=10%, list=1%, signal=1%		
13 REACTOME_TETRATRANTOL_4TP_2TPK BINDS AND DESTABILIZES_MRNA	Details... 17	0.71	2.65	0.000	0.000	0.000	4845	tags=10%, list=1%, signal=1%		
14 REACTOME_GMP_FADING_DNA_PRIMER_SYNTHESIS_AND_LIGATION_IN_GLIOMA	Details... 23	0.80	2.64	0.000	0.000	0.000	5421	tags=10%, list=1%, signal=1%		
15 REACTOME_BUTYRATE_RESPONSE_FACTOR_1_RF1 BINDS AND DESTABILIZES_MRNA	Details... 17	0.86	2.61	0.000	0.000	0.000	4848	tags=10%, list=1%, signal=1%		

	GS follow link to MsigDB									
	GS DETAILS	SIZE	ES	NES	NOM P-VAL	FDR P-VAL	RANK	MAX	LEADING EDGE	
1 REACTOME_CLASS_I_PEROXISOMAL_MEMBRANE_PROTEIN_IMPORT	Details... 19	-0.82	-1.67	0.000	0.000	0.003	1387	tags=11%, list=2%, signal=1%		
2 REACTOME_SYNTHESIS_OF_BILE_ACIDS_AND_BILE_SALTS_VIA_7ALPHA_HYDROXYCHOLESTEROL	Details... 22	-0.71	-1.45	0.002	0.003	0.645	6392	tags=36%, list=11%, signal=41%		
3 REACTOME_FATTY_ACIDS	Details... 22	-0.70	-1.44	0.001	0.003	0.692	12807	tags=73%, list=23%, signal=2%		
4 REACTOME_SYNTHESIS_OF_BILE_ACIDS_AND_BILE_SALTS	Details... 32	-0.67	-1.43	0.001	0.004	0.783	6390	tags=29%, list=11%, signal=15%		
5 REACTOME_NEGATIVE_REGULATION_OF_FLTs	Details... 15	-0.71	-1.42	0.011	0.005	0.855	5376	tags=13%, list=10%, signal=15%		
6 REACTOME_Olfactory_SIGNALING_PATHWAY	Details... 72	-0.64	-1.41	0.000	0.005	0.862	14240	tags=74%, list=25%, signal=1%		
7 REACTOME_EICOSANOIDS	Details... 17	-0.66	-1.34	0.022	0.017	0.998	12807	tags=71%, list=23%, signal=9%		
8 REACTOME_WNT_LIGAND_BIOGENESIS_AND_TRAFFICKING	Details... 25	-0.64	-1.34	0.019	0.019	0.999	10990	tags=46%, list=19%, signal=9%		
9 REACTOME_PHOSPHOLIPASE_C_MEDIATED CASCADE_FGFR1	Details... 16	-0.67	-1.34	0.028	0.019	0.999	15220	tags=29%, list=27%, signal=10%		
10 REACTOME_SHC_MEDIATED CASCADE_FGFR1	Details... 20	-0.66	-1.34	0.022	0.020	1.000	15220	tags=46%, list=27%, signal=8%		
11 REACTOME_FGFR1_LIGAND_BINDING_AND_ACTIVATION	Details... 17	-0.67	-1.33	0.022	0.020	1.000	15220	tags=76%, list=27%, signal=8%		
12 REACTOME_PL3K CASCADE_FGFR1	Details... 21	-0.65	-1.33	0.020	0.022	1.000	15220	tags=57%, list=27%, signal=8%		
13 REACTOME_FORMATION_OF_THE_CORNIFIED_ENVELOPE	Details... 90	-0.59	-1.33	0.000	0.022	1.000	16333	tags=59%, list=29%, signal=8%		
14 REACTOME_PEPTIDE_HORMONE_METABOLISM	Details... 61	-0.60	-1.32	0.000	0.023	1.000	15829	tags=46%, list=28%, signal=10%		
15 REACTOME_INTERLEUKIN_2_FAMILY_SIGNALING	Details... 36	-0.61	-1.31	0.006	0.027	1.000	9727	tags=31%, list=17%, signal=3%		

Min PHRED
= 35

GSEA — Modified Max Read Length

Kate

Stem cells

Enrichment in phenotype: 0 (3 samples)

- 291 / 807 gene sets are upregulated in phenotype **0**
- 279 gene sets are significant at FDR < 25%
- 51 gene sets are significantly enriched at nominal pvalue < 1%
- 51 gene sets are significantly enriched at nominal pvalue < 5%
- [Snapshot](#) of enrichment results
- Detailed [enrichment results in html](#) format
- Detailed [enrichment results in TSV](#) format (tab delimited text)
- [Guide to interpret results](#)

Enrichment in phenotype: 4 (5 samples)

- 516 / 807 gene sets are upregulated in phenotype **4**
- 100 gene sets are significantly enriched at FDR < 25%
- 16 gene sets are significantly enriched at nominal pvalue < 1%
- 49 gene sets are significantly enriched at nominal pvalue < 5%
- [Snapshot](#) of enrichment results
- Detailed [enrichment results in html](#) format
- Detailed [enrichment results in TSV](#) format (tab delimited text)
- [Guide to interpret results](#)

Enterocytes

Enrichment in phenotype: 0 (3 samples)

- 288 / 807 gene sets are upregulated in phenotype **0**
- 267 gene sets are significant at FDR < 25%
- 46 gene sets are significantly enriched at nominal pvalue < 1%
- 46 gene sets are significantly enriched at nominal pvalue < 5%
- [Snapshot](#) of enrichment results
- Detailed [enrichment results in html](#) format
- Detailed [enrichment results in TSV](#) format (tab delimited text)
- [Guide to interpret results](#)

Enrichment in phenotype: 4 (5 samples)

- 519 / 807 gene sets are upregulated in phenotype **4**
- 103 gene sets are significantly enriched at FDR < 25%
- 20 gene sets are significantly enriched at nominal pvalue < 1%
- 52 gene sets are significantly enriched at nominal pvalue < 5%
- [Snapshot](#) of enrichment results
- Detailed [enrichment results in html](#) format
- Detailed [enrichment results in TSV](#) format (tab delimited text)
- [Guide to interpret results](#)

Default Parameters

Max read length = 85

GSEA — Modified Max Read Length (cont.)

Kate

Default Parameters

	GS follow link to MSigDB									
	GS DETAILS	SIZE	E5	NES	NOM p-val	FDR p-val	FWER p-val	RANK AT MAX	LEADING EDGE	
1	REACTOME_POSTMITOTIC_NUCLEAR_PORE_COMPLEX_NPC_REFORMATION	Details... 22	0.90	3.37	0.000	0.000	3662	tags=>85%, ist=>85%		
2	REACTOME_BUTYRATE_RESPONSE_FACTOR_1_BRF1_BINDS_AND_DESTABILIZES_MRNA	Details... 17	0.85	3.32	0.000	0.000	4850	tags=>76%, ist=>95%		
3	REACTOME_LAGGING_STRAND_SYNTHESIS	Details... 20	0.92	3.20	0.000	0.000	2729	tags=>85%, ist=>74%		
4	REACTOME_INITIATION_OF_NUCLEAR_ENVELOPE_NE_REFORMATION	Details... 17	0.79	3.22	0.000	0.000	3237	tags=>65%, ist=>95%		
5	REACTOME_TRANSCRIPTIONAL_REGULATION_BY_E2Fs	Details... 23	0.78	3.14	0.000	0.000	4862	tags=>76%, ist=>95%		
6	REACTOME_MRNA_DECAY_BY_TD_S_EXORIBONUCLEASE	Details... 16	0.90	3.05	0.000	0.000	5309	tags=>85%, ist=>95%		
7	REACTOME_PCR_DEPENDENT_LONG_PATCH_BASE_EXCISION_REPAIR	Details... 21	0.87	3.00	0.000	0.000	2564	tags=>76%, ist=>85%		
8	REACTOME_KSRP_KHSP BINDS AND DESTABILIZES_MRNA	Details... 15	0.91	3.01	0.000	0.000	4830	tags=>85%, ist=>95%		
9	REACTOME_INHIBITION_OF_THE_PROTEOLYTIC_ACTIVITY_OF_APc_C_REQUIRED_FOR_THE_ONSET_OF_ANAPHASE_BY_MITOTIC_SPINDLE_CHECKPOINT_COMPONENTS	Details... 21	0.80	2.94	0.000	0.000	4420	tags=>76%, ist=>95%		
10	REACTOME_PHOSPHORYLATION_OF_THE_APc_C	Details... 20	0.78	2.93	0.000	0.000	5023	tags=>85%, ist=>95%		
11	REACTOME_PROCESSING_OF_ATRONESS_PRE_MRNAS	Details... 20	0.90	2.89	0.000	0.000	5568	tags=>100%, ist=>10%		
12	REACTOME_FGF12_ALTERNATIVE_SPLICING	Details... 17	0.78	2.77	0.000	0.000	4217	tags=>71%, ist=>76%		
13	REACTOME_FORMATION_OF_SENESCENCE_ASSOCIATED_HETEROCHROMATIN_FOCI_SAHE	Details... 16	0.86	2.62	0.000	0.000	3319	tags=>85%, ist=>95%		
14	REACTOME_PROCESSIVE_SYNTHESIS_ON_THE_LAGGING_STRAND	Details... 15	0.91	2.57	0.000	0.000	2729	tags=>87%, ist=>74%		
15	REACTOME_PROCESSIVE_SYNTHESIS_ON_THE_C_STRAND_OF_THE_TELOMERE	Details... 18	0.83	2.57	0.000	0.000	2471	tags=>65%, ist=>45%		
		Details... 20	0.71	2.52	0.000	0.000	3828	tags=>65%, ist=>70%		

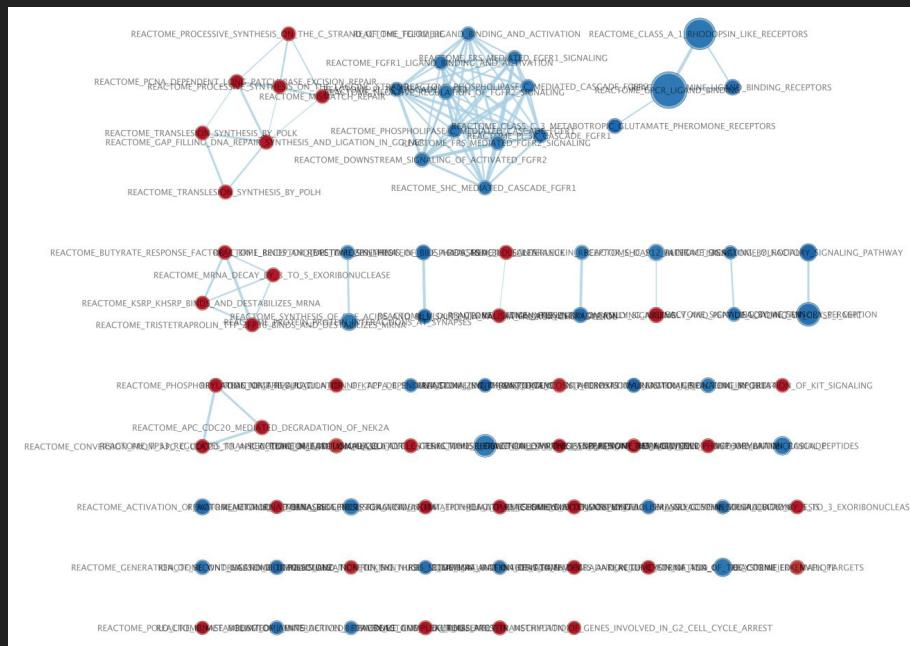
	GS follow link to MSigDB									
	GS DETAILS	SIZE	E5	NES	NOM p-val	FDR p-val	FWER p-val	RANK AT MAX	LEADING EDGE	
1	REACTOME_CLASS_I_PEROXISOMAL_MEMBRANE_PROTEIN_IMPORT	Details... 19	0.81	1.66	0.000	0.000	2009	tags=>11%, ist=>11%		
2	REACTOME_SYNTHESIS_OF_BILE_ACIDS_AND_BILE_SALTS_VIA_TALPHA_HYDROXYCHOLESTEROL	Details... 22	0.76	1.45	0.000	0.002	5980	tags=>11%, ist=>11%		
3	REACTOME_NEGATIVE_REGULATION_OF_FLT3	Details... 15	0.75	1.43	0.012	0.004	5779	tags=>12%, ist=>10%		
4	REACTOME_FATTY_ACIDS	Details... 22	0.68	1.42	0.025	0.004	7386	tags=>73%, ist=>10%		
5	REACTOME_SYNTHESIS_OF_BILE_ACIDS_AND_BILE_SALTS	Details... 22	0.69	1.41	0.003	0.005	8554	tags=>73%, ist=>10%		
6	REACTOME_Olfactory_SIGNALING_PATHWAY	Details... 72	0.68	1.40	0.000	0.007	917	tags=>26%, ist=>24%		
7	REACTOME_WNT_LIGAND_BIOGENESIS_AND_TRAFFICKING	Details... 25	0.64	1.34	0.017	0.019	14037	tags=>56%, ist=>26%		
8	REACTOME_Eicosanoids	Details... 17	0.65	1.32	0.038	0.024	13495	tags=>44%, ist=>24%		
9	REACTOME_AMINE_LIGAND_BINDING_RECEPORS	Details... 43	0.61	1.32	0.004	0.025	18466	tags=>74%, ist=>35%		
10	REACTOME_PHOSPHOPOLYPEPTIDE_C_MEDIATED CASCADE_FGFR1	Details... 16	0.65	1.32	0.038	0.026	16394	tags=>75%, ist=>25%		
11	REACTOME_FORMATION_OF_THE_COIFFED_ENVELOPE	Details... 90	0.58	1.32	0.000	0.026	17021	tags=>60%, ist=>30%		
12	REACTOME_SHC_MEDIATED CASCADE_FGFR1	Details... 20	0.64	1.32	0.035	0.027	16394	tags=>49%, ist=>29%		
13	REACTOME_NEUROKININS_AND_NEUROGLIGINS	Details... 31	0.61	1.31	0.017	0.029	19296	tags=>39%, ist=>17%		
14	REACTOME_FGFR1_LIGAND_BINDING_AND_ACTIVATION	Details... 17	0.65	1.31	0.030	0.030	16394	tags=>76%, ist=>26%		
15	REACTOME_PI_3K CASCADE_FGFR1	Details... 21	0.63	1.30	0.042	0.033	16394	tags=>57%, ist=>29%		

Max Read Length: 85

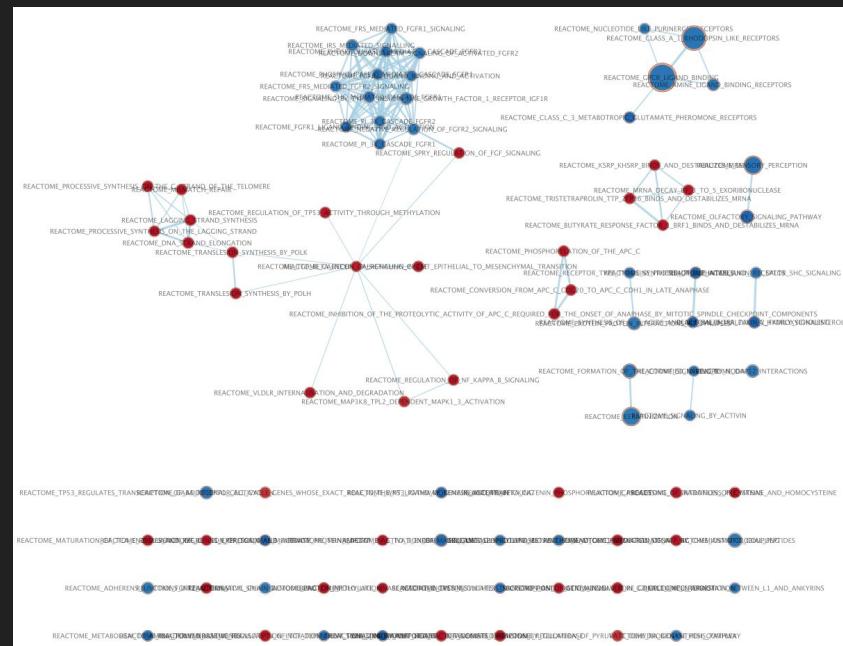
	GS follow link to MSigDB									
	GS DETAILS	SIZE	E5	NES	NOM p-val	FDR p-val	FWER p-val	RANK AT MAX	LEADING EDGE	
1	REACTOME_POSTMITOTIC_NUCLEAR_PORE_COMPLEX_NPC_REFORMATION	Details... 22	0.89	3.59	0.000	0.000	3777	tags=>82%, ist=>85%		
2	REACTOME_PCR_DEPENDENT_LONG_PATCH_BASE_EXCISION_REPAIR	Details... 21	0.87	3.42	0.000	0.000	2560	tags=>75%, ist=>80%		
3	REACTOME_PHORBOL_GTPASE_CYCLE	Details... 22	0.85	3.25	0.000	0.000	5305	tags=>82%, ist=>90%		
4	REACTOME_INHIBITION_OF_THE_PROTEOLYTIC_ACTIVITY_OF_APc_C_REQUIRED_FOR_THE_ONSET_OF_ANAPHASE_BY_MITOTIC_SPINDLE_CHECKPOINT_COMPONENTS	Details... 21	0.80	3.09	0.000	0.000	5002	tags=>81%, ist=>90%		
5	REACTOME_PROCESSIVE_SYNTHESIS_ON_THE_LAGGING_STRAND	Details... 15	0.91	3.09	0.000	0.000	2767	tags=>87%, ist=>91%		
6	REACTOME_LAGGING_STRAND_SYNTHESIS	Details... 20	0.92	3.05	0.000	0.000	2767	tags=>90%, ist=>95%		
7	REACTOME_MISMATCH_REPAIR	Details... 15	0.87	2.91	0.000	0.000	2333	tags=>67%, ist=>70%		
8	REACTOME_FORMATION_OF_SENESCENCE_ASSOCIATED_HETEROCHROMATIN_FOCI_SAHE	Details... 16	0.86	2.89	0.000	0.000	3374	tags=>94%, ist=>95%		
9	REACTOME_KSRP_KHSP BINDS AND DESTABILIZES_MRNA	Details... 15	0.91	2.83	0.000	0.000	4863	tags=>85%, ist=>95%		
10	REACTOME_MRNA_DECAY_BY_3' TO 5' EXORIBONUCLEASE	Details... 16	0.90	2.83	0.000	0.000	5319	tags=>85%, ist=>95%		
11	REACTOME_PROTEIN_HYDROXYLATION	Details... 16	0.74	2.72	0.000	0.000	4749	tags=>65%, ist=>73%		
12	REACTOME_TRISITETRAPROLIN_TTP_ZFP96 BINDS AND DESTABILIZES_MRNA	Details... 17	0.71	2.68	0.000	0.000	4852	tags=>75%, ist=>77%		
13	REACTOME_INITIATION_OF_NUCLEAR_ENVELOPE_NE_REFORMATION	Details... 20	0.73	2.66	0.000	0.000	3243	tags=>65%, ist=>69%		
14	REACTOME_CONVERSION_FROM_APc_C_CDC20_TO_APc_C_CDH1_IN_LATE_ANAPHASE	Details... 20	0.71	2.54	0.000	0.000	5006	tags=>75%, ist=>82%		
15	REACTOME_TELOMERE_EXTENSION_BY_TELOMERASE	Details... 20	0.71	2.52	0.000	0.000	3828	tags=>65%, ist=>70%		

	GS follow link to MSigDB									
	GS DETAILS	SIZE	E5	NES	NOM p-val	FDR p-val	FWER p-val	RANK AT MAX	LEADING EDGE	
1	REACTOME_CLASS_I_PEROXISOMAL_MEMBRANE_PROTEIN_IMPORT	Details... 19	0.80	1.67	0.000	0.000	2005	tags=>11%, ist=>10%		
2	REACTOME_SYNTHESIS_OF_BILE_ACIDS_AND_BILE_SALTS_VIA_TALPHA_HYDROXYCHOLESTEROL	Details... 22	0.75	1.42	0.001	0.005	4955	tags=>91%, ist=>11%		
3	REACTOME_FATTY_ACIDS	Details... 22	0.68	1.42	0.001	0.005	19408	tags=>79%, ist=>24%		
4	REACTOME_NEGATIVE_REGULATION_OF_FLT3	Details... 15	0.71	1.41	0.014	0.006	5777	tags=>65%, ist=>10%		
5	REACTOME_SYNTHESIS_OF_BILE_ACIDS_AND_BILE_SALTS	Details... 32	0.68	1.41	0.003	0.006	8372	tags=>29%, ist=>11%		
6	REACTOME_Olfactory_SIGNALING_PATHWAY	Details... 72	0.65	1.38	0.000	0.010	3737	tags=>69%, ist=>24%		
7	REACTOME_PI_3K CASCADE_FGFR	Details... 21	0.68	1.36	0.012	0.013	15122	tags=>31%, ist=>27%		
8	REACTOME_FGFR_LIGAND_BINDING_AND_ACTIVATION	Details... 17	0.65	1.36	0.009	0.015	15122	tags=>67%, ist=>10%		
9	REACTOME_SHC_MEDIATED CASCADE_FGFR1	Details... 20	0.65	1.36	0.012	0.015	9966	tags=>62%, ist=>20%		
10	REACTOME_PHOSPHOPOLYPEPTIDE_C_MEDIATED CASCADE_FGFR1	Details... 16	0.67	1.34	0.037	0.017	15122	tags=>75%, ist=>27%		
11	REACTOME_WNT_LIGAND_BIOGENESIS_AND_TRAFFICKING	Details... 25	0.65	1.34	0.017	0.019	14540	tags=>65%, ist=>20%		
12	REACTOME_PI_3K CASCADE_FGFR	Details... 22	0.65	1.32	0.029	0.025	15122	tags=>65%, ist=>27%		
13	REACTOME_Ecdysteroids	Details... 17	0.65	1.31	0.042	0.026	15408	tags=>71%, ist=>20%		
14	REACTOME_FIR_MEDIATED_FIGHTER_SIGNALING	Details... 22	0.65	1.31	0.019	0.026	15122	tags=>75%, ist=>20%		
15	REACTOME_AMINE_LIGAND_BINDING_RECEPORS	Details... 43	0.65	1.31	0.002	0.026	15883	tags=>71%, ist=>33%		

Enrichment Map — Modified Minimum PHRED Score

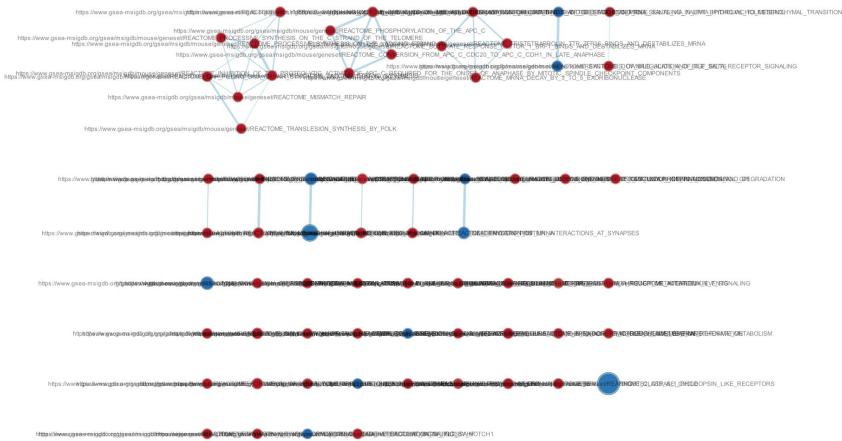


Default Parameters



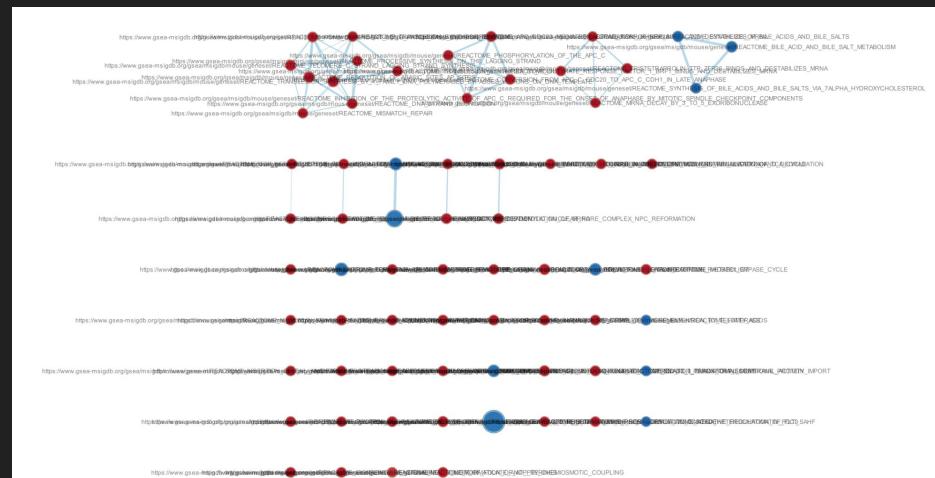
Min PHRED = 35

Enrichment Map — Modified Sliding Window



Default Parameters

Alex



Sliding Window = 2

Discussion - Biologic Relevance

Stem Enrichment

Bryan

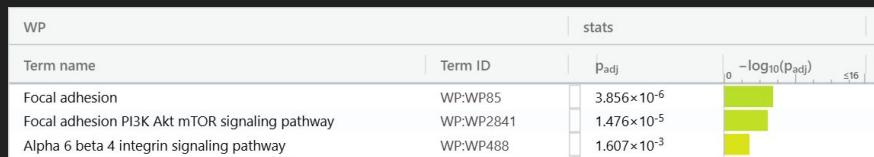
5	REACTOME_MRNA_CAPPING	Details	28	0.70	3.14	0.000	0.000	0.000	5126	tags=64%, list=9%, signal=71%
6	REACTOME_PHOSPHORYLATION_OF_THE_APC_C	Details	20	0.78	3.06	0.000	0.000	0.000	5023	tags=80%, list=9%, signal=88%
7	REACTOME_DNA_STRAND_ELONGATION	Details	24	0.91	3.00	0.000	0.000	0.000	2729	tags=88%, list=6%, signal=92%
8	REACTOME_BUTYRATE_RESPONSE_FACTOR_1_BRF1 BINDS AND DESTABILIZES mRNA	Details	17	0.85	2.96	0.000	0.000	0.000	4830	tags=76%, list=6%, signal=84%
9	REACTOME_PROCESSIVE_SYNTHESIS_ON_THE_LAGGING_STRAND	Details	15	0.91	2.91	0.000	0.000	0.000	2729	tags=97%, list=0%, signal=100%
10	REACTOME_PROCESSIVE_SYNTHESIS_ON_THE_C_STRAND_OF_THE_TELOMERE	Details	18	0.83	2.76	0.000	0.000	0.000	2471	tags=67%, list=4%, signal=70%

Stem cells are proliferative

Pond, K. W., et al. 2022

Enterocyte Enrichment

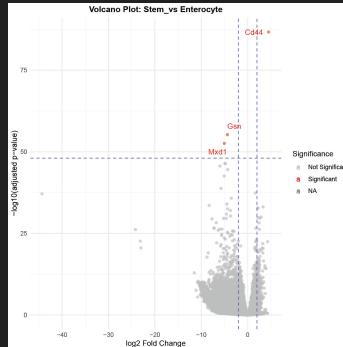
Kate



Focal Adhesions important for enterocytes

Ashton, G. H. et al. 2010

Alex



Mxd1 competitive antagonist with Myc.
Myc important for stem cells

Raffaeiner, P., et al. 2020.

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