COAD-READ, 221 samples
k=7 consensus clusters
for miRNA-seq 5’-isomiR abundance data

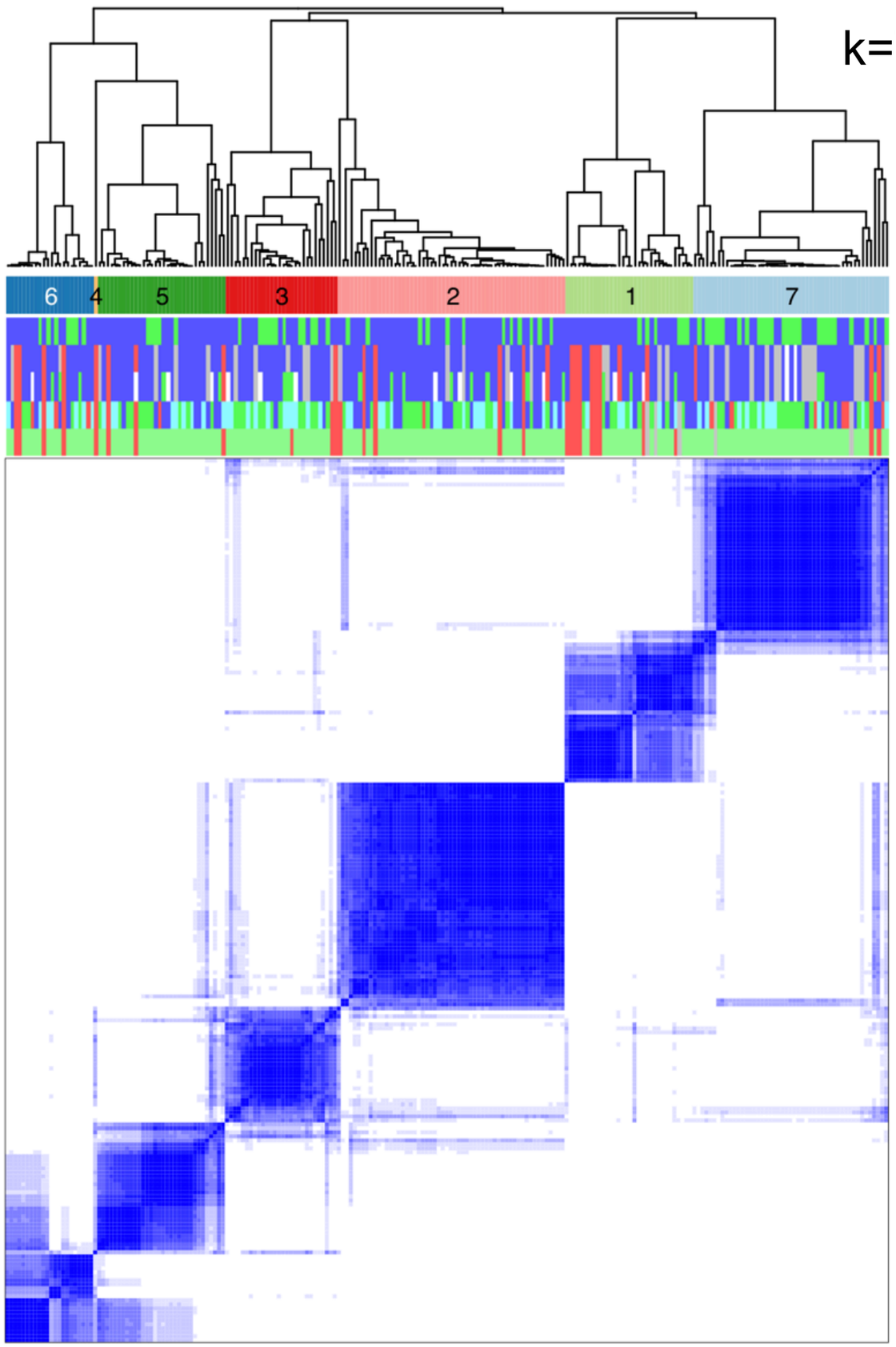
G. Robertson, Andy Chu, Samuel Wu, Elizabeth Chun, Andy Mungall, Inanc Birol

BC Cancer Agency Genome Sciences Centre

13 June 2011, 10h30

1. 5’-isomiR abundance data give well-separated consensus clusters.
2. Clusters appear to correlate poorly to covariate information.
3. For genes in Wnt signaling pathway, RPKM can vary between clusters.

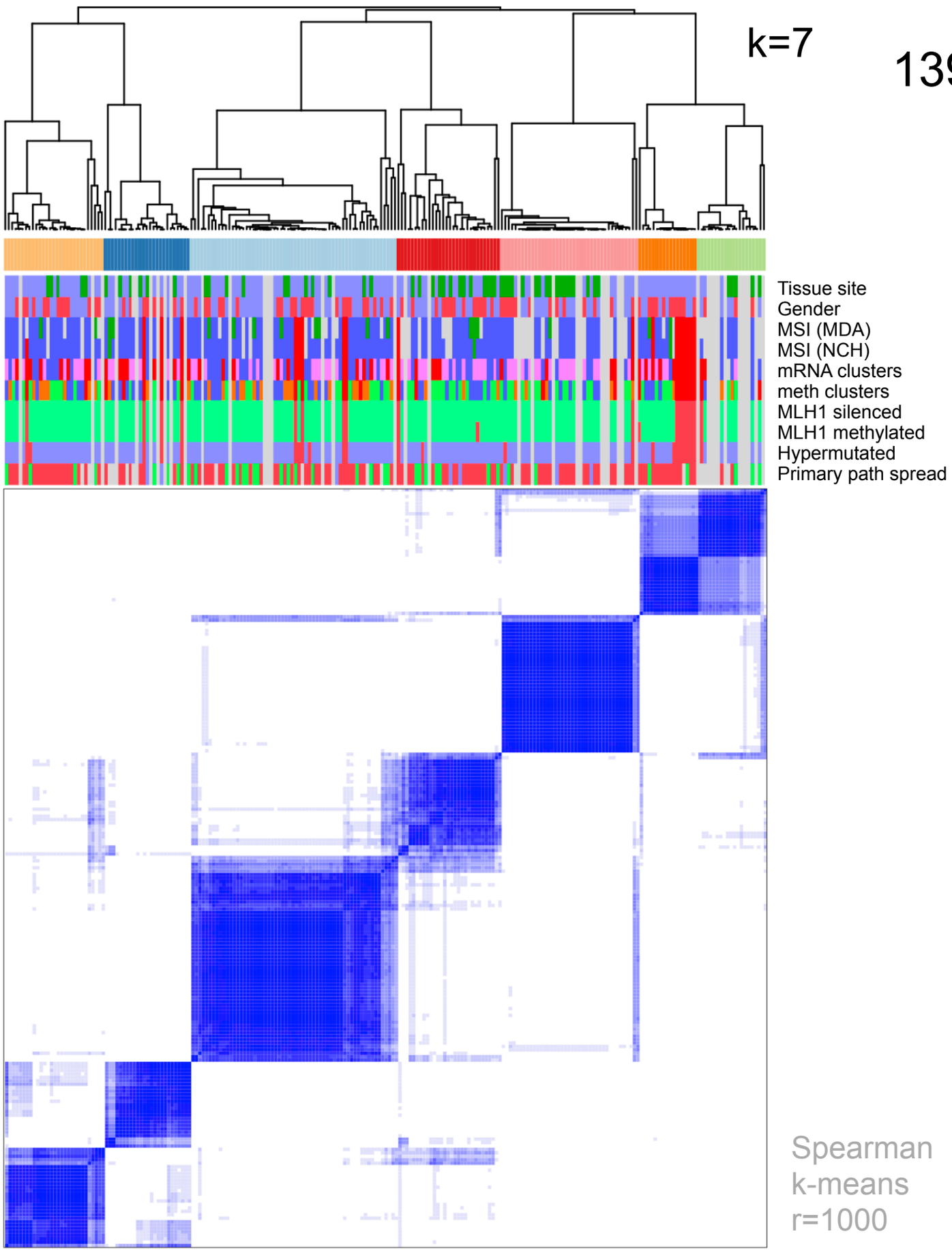
COAD-READ miRNA-seq
5943 unthresholded 5’-isomiR data
221 samples



COAD
READ

CIMP-H
CIMP-L
Cluster 3
Cluster 4

Spearman
k-means
r=1000



221 COAD-READ miRNA-seq
1393 thresholded 5’-isomiRs (t=0.1)

282 miRNA basenames (hsa-mir-10a)
416 MIMATs (MIMAT0000062)

crc_groupings_v2.txt
194 samples

Spearman
k-means
r=1000

221 COAD-READ miRNA-seq
1393 5’-isomiRs (t=0.1)

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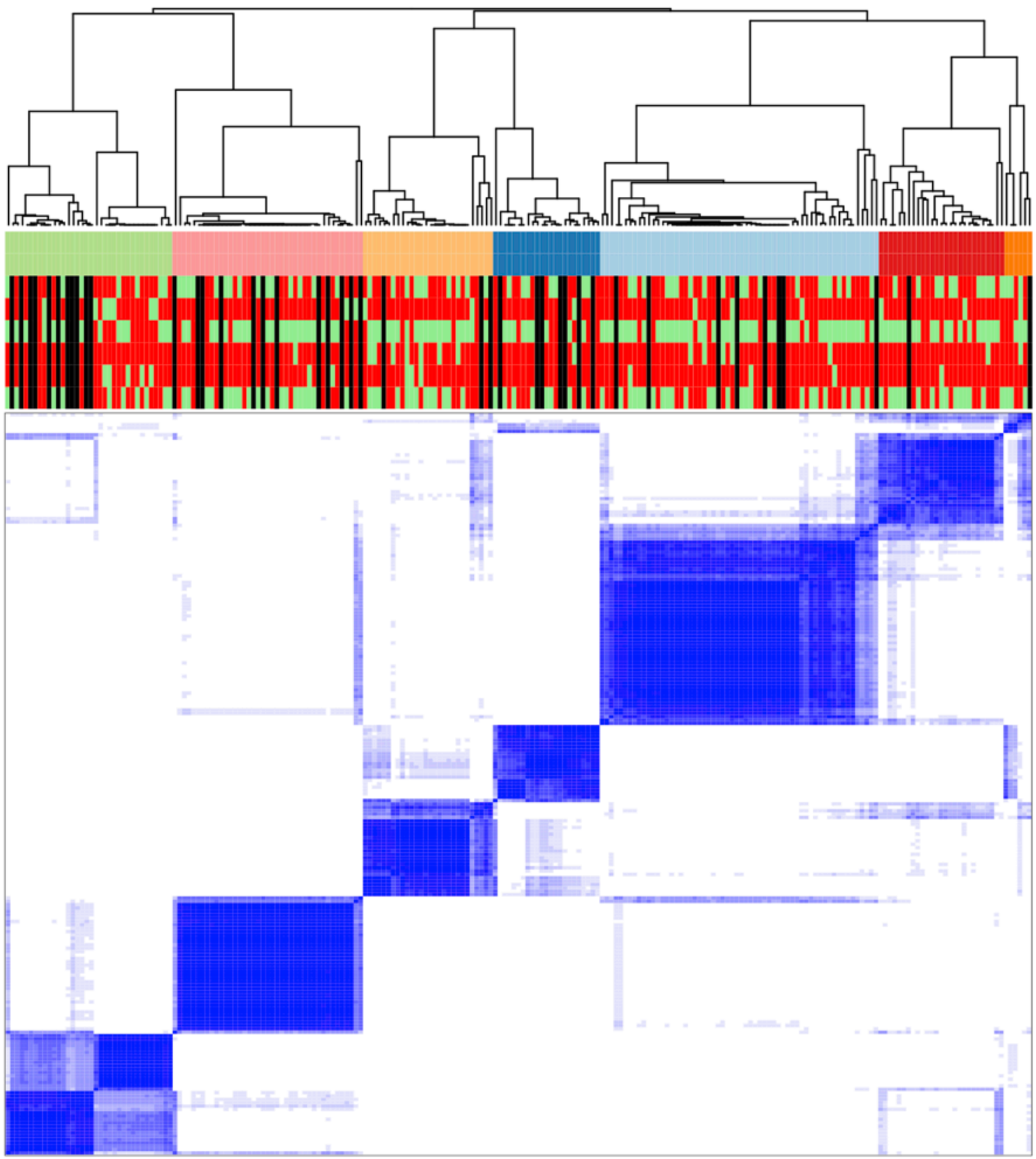


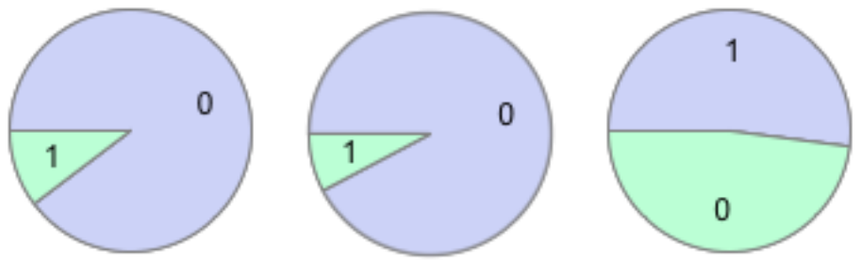
0	
1	

crc_groupings_v2.txt
194 samples

KRAS mut	
0	113
1	81

BRAF mut	
0	176
1	18

APC mut	
0	52
1	142



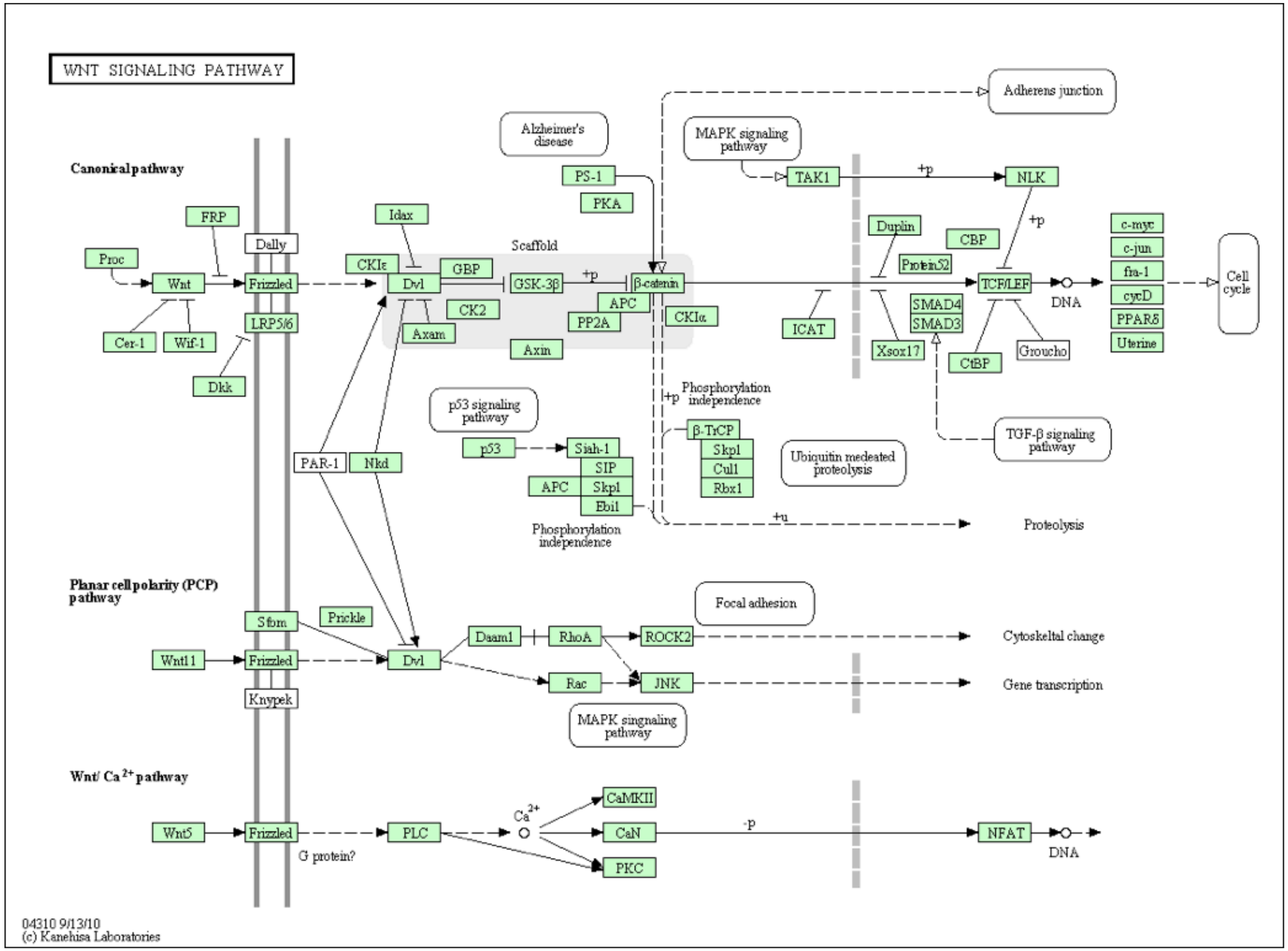
PIK3CA mut	
0	174
1	20

FBXW7 mut	
0	179
1	15

TP53 mut	
0	93
1	101

tracks: Samuel Wu

KEGG WNT signaling pathway



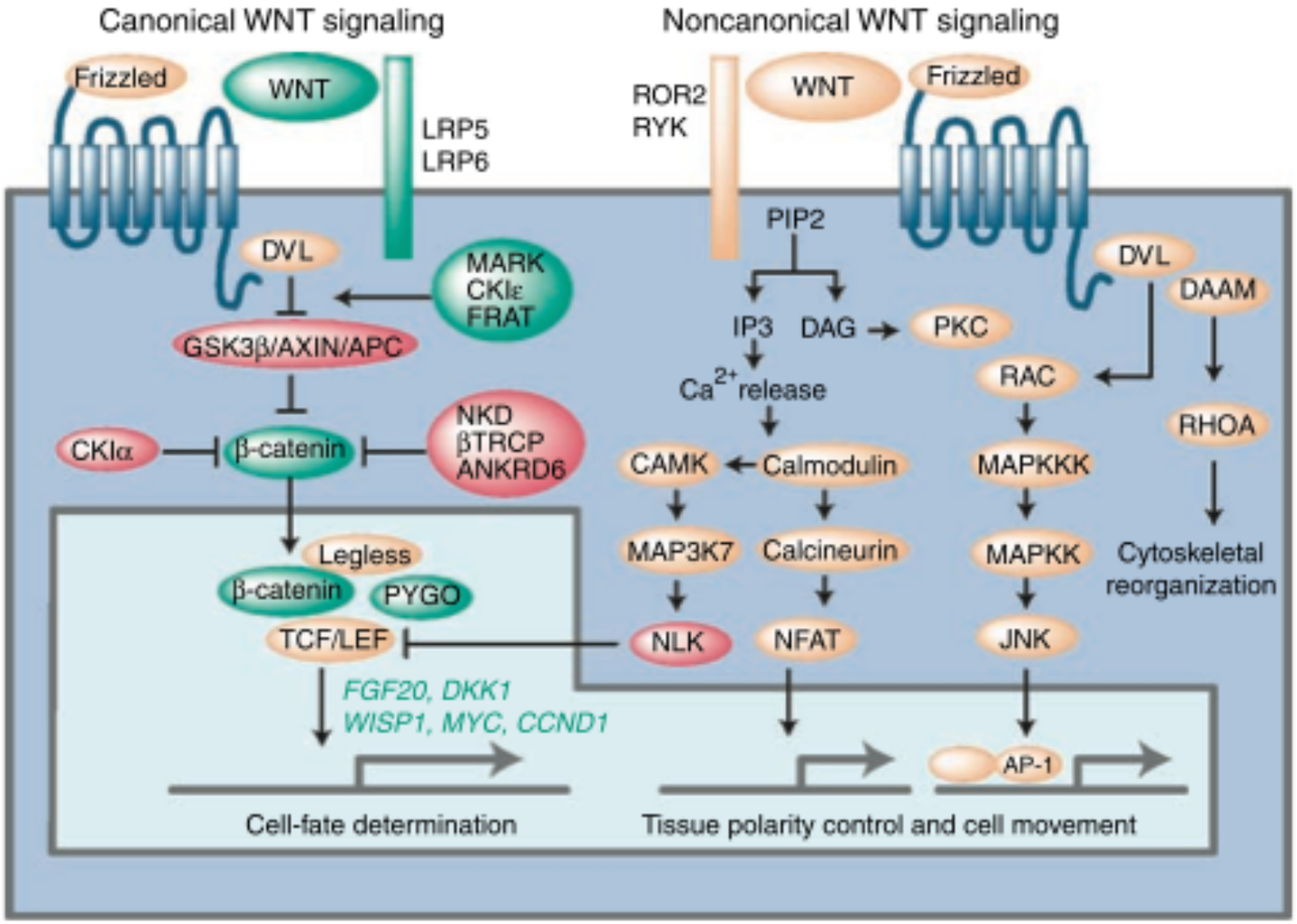
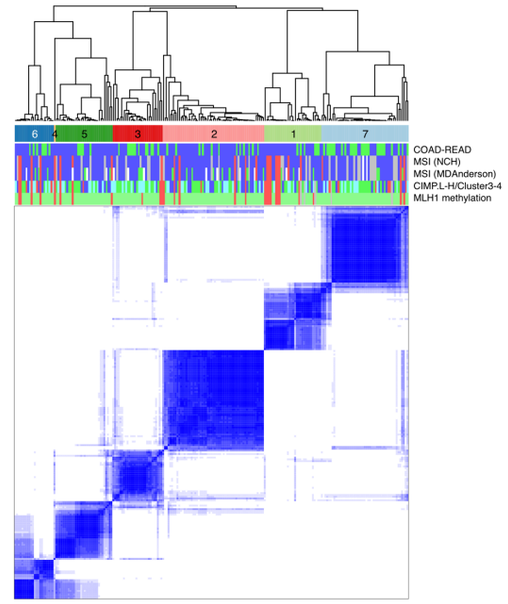


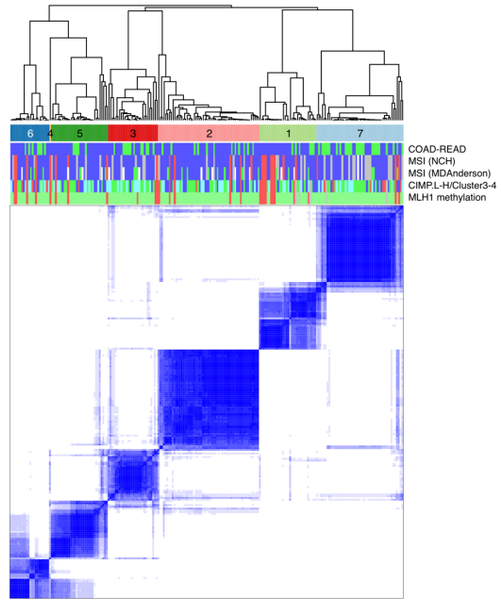
Fig.1. Landscape of WNT signaling cascades. WNT signals are transduced to the **canonical** pathway for cell fate determination, and to the **noncanonical** pathway for control of **cell movement and tissue polarity**. **Canonical** WNT signals are transduced through Frizzled family receptors and LRP5/LRP6 coreceptor to the h-catenin signaling cascade. **Noncanonical** WNT signals are transduced through Frizzled family receptors and ROR2/RYK coreceptors to the DVL-dependent (Rho family GTPases and JNK) or the Ca2+-dependent (NLK and NFAT) signaling cascades. Microtubule affinity ^ regulating kinase (MARK ; PAR-1) family kinases, CKIq, and FRAT are **positive** regulators of the **canonical** WNT pathway, whereas APC, AXIN1, AXIN2, CKIa, NKD1, NKD2, hTRCP1, hTRCP2, ANKRD6, NLK, and PPARg are **negative** regulators. FGF20, DKK1, WISP1, MYC, CCND1, and Glucagon (GCG) are **target** genes of the **canonical** WNT signaling pathway. WNT signals are context-dependently transduced to both pathways based on the expression profile of WNT, SFRP, WIF, DKK, Frizzled receptors, coreceptors, and the activity of intracellular WNTsignaling regulators. Katoh and Kato, Clin Cancer Res 2007, 13:4042.

WNTs: RPKM across 7 miRNA-seq-based clusters

crc_244_gene_rpkm.txt



SFRPs, WIF1 and DKKs: RPKM abundance across 7 clusters



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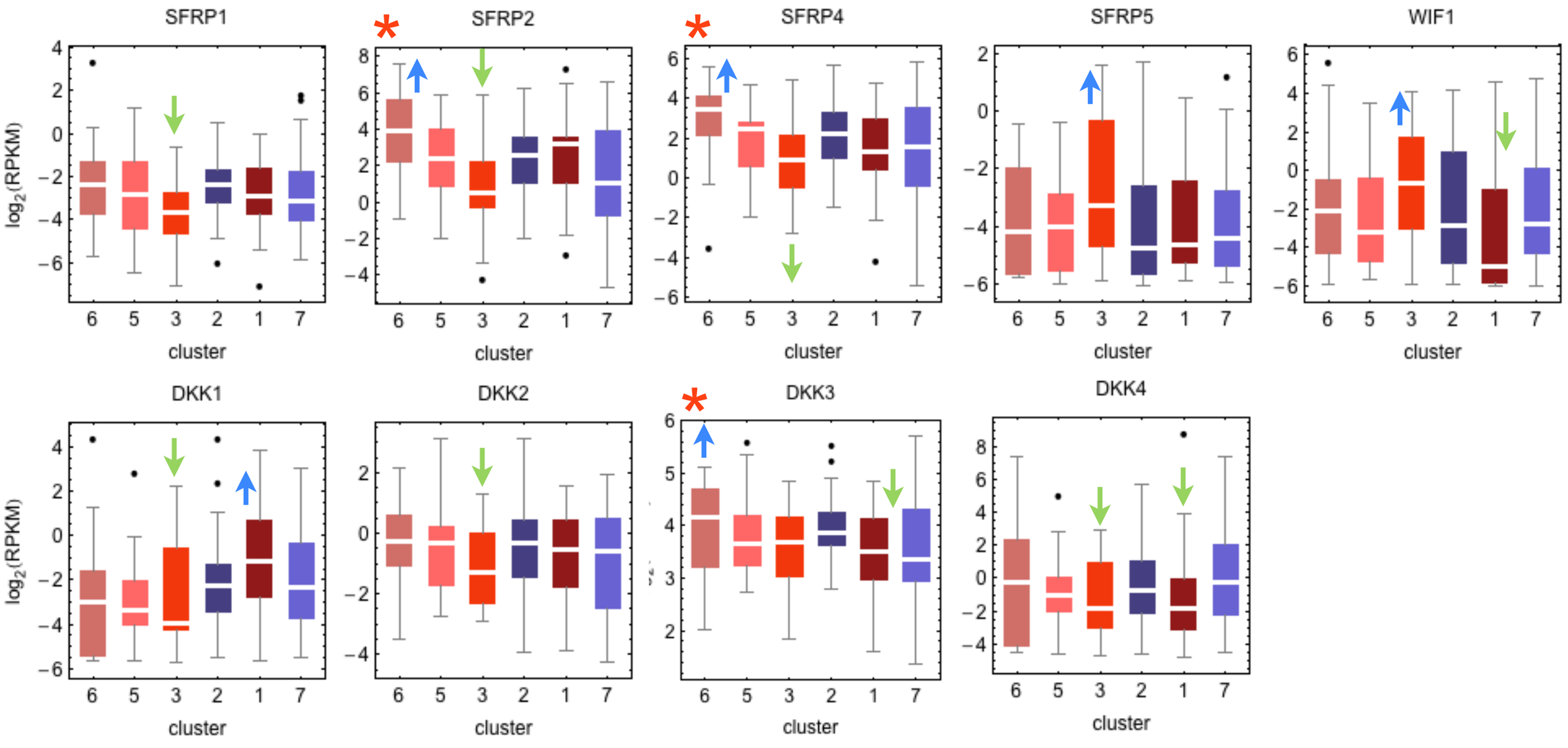
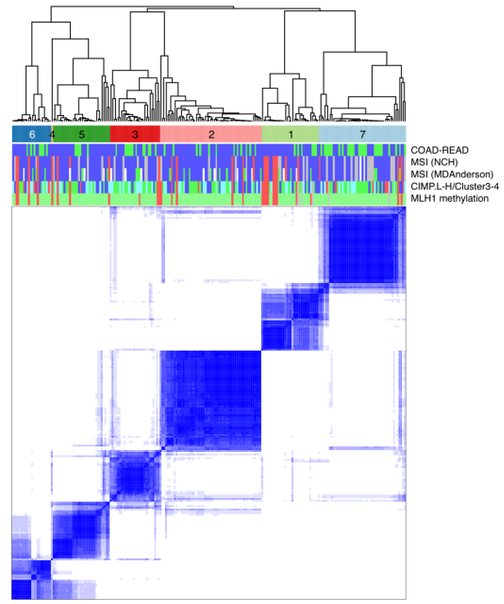
SFRP1 SFRP2 SFRP4 SFRP5



Frizzled 1 to 10: RPKM abundance across 7 clusters

```
grep "FZD" crc_244_gene_rpkm.txt | cut -f1
FZD1|8321
FZD2|2535
FZD3|7976
FZD4|8322
FZD5|7855
FZD6|8323
FZD7|8324
FZD8|8325
FZD9|8326
FZD10|11211
```



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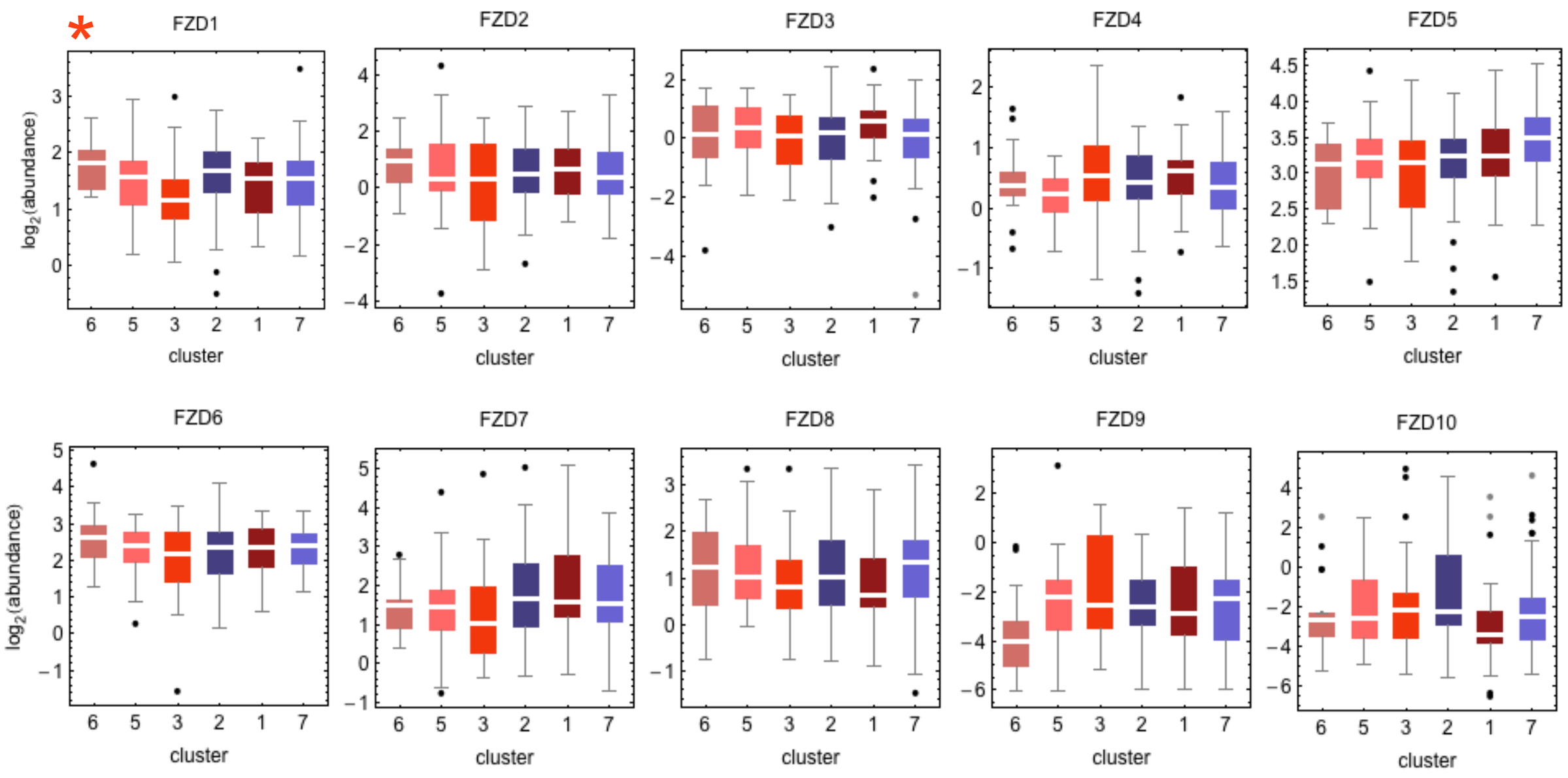


RPKM abundance across 7 clusters

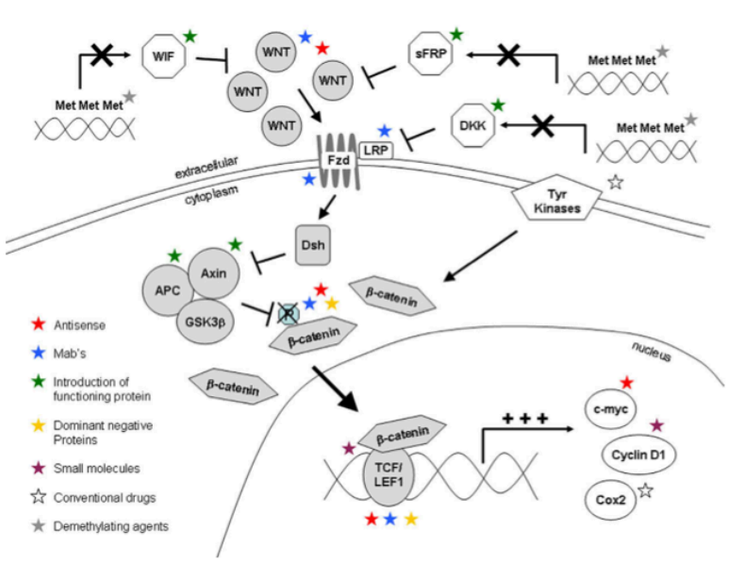
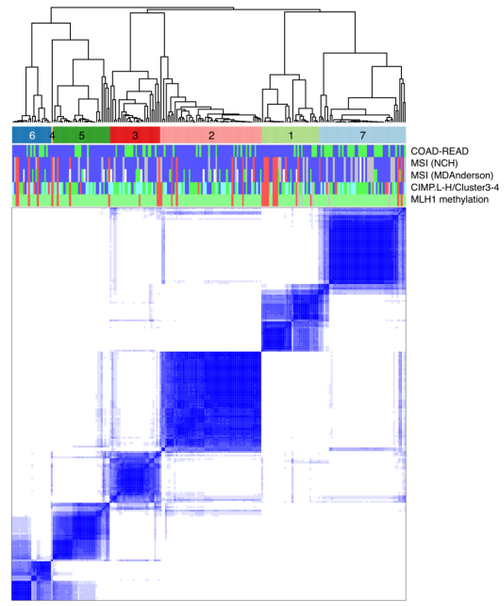
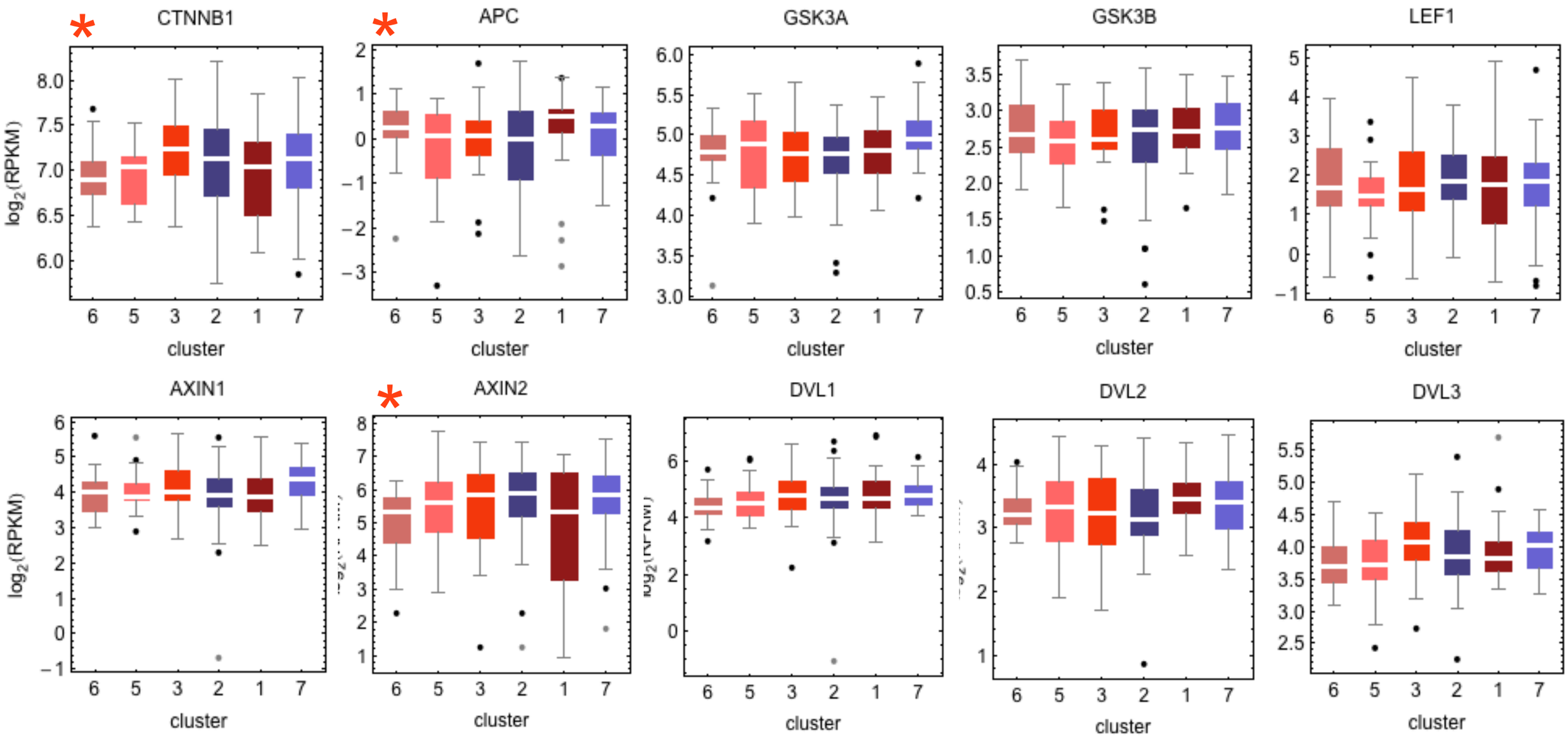


Fig. 1 – Active WNT/b-catenin signalling cascade: WNT proteins bind the receptor complex and activate DSH, which inhibits the **APC/Axin/GSK3b** complex, preventing phosphorylation of b-catenin, which accumulates in the cytoplasm, translocates into the nucleus and activates TCF/**LEF1** family transcription factors. Stars indicate possible intervention targets and approaches following shown colour scheme. **APC**: adenomatous polyposis coli; Cox2: cyclooxygenase 2; DKK: Dickkopf; **DSH**: Dishevelled; FZD: Frizzled; GSK3: glycogen synthase kinase 3; LEF1: Lymphoid enhancer-binding factor 1; LRP: low-density lipoprotein receptor-related protein, Mabs: Monoclonal antibodies, Met: hypermethylation; P: phosphorylation, sFRP: secreted frizzled related protein; TCF: T-cell factor; Tyr kinase: Tyrosine kinase; WIF1: WNT inhibitory factor 1. Gehrke et al, 2009. Eur J Cancer 25:2759-2767.

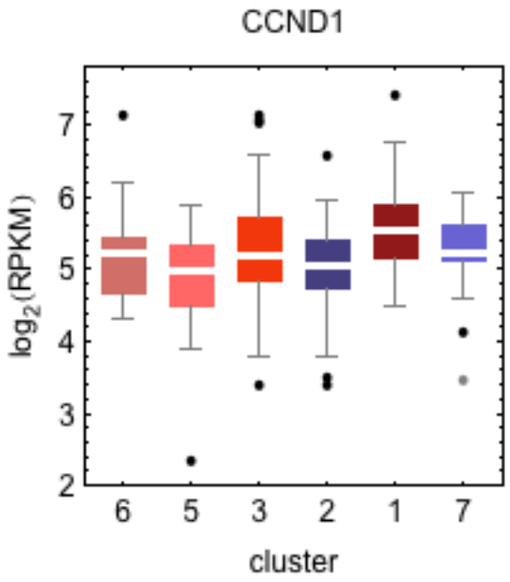


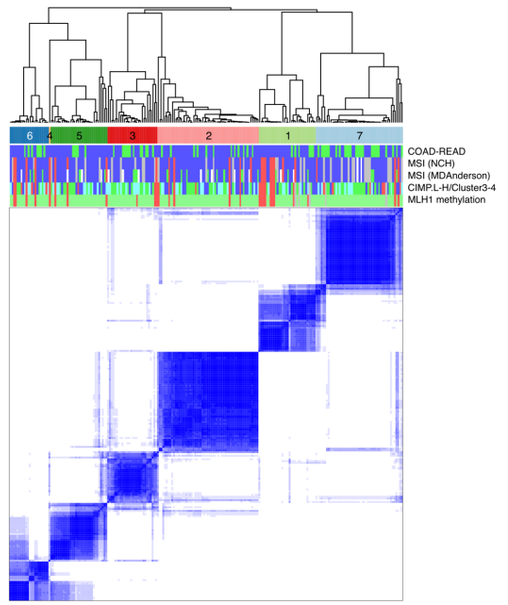
21 May 2011, 12h00

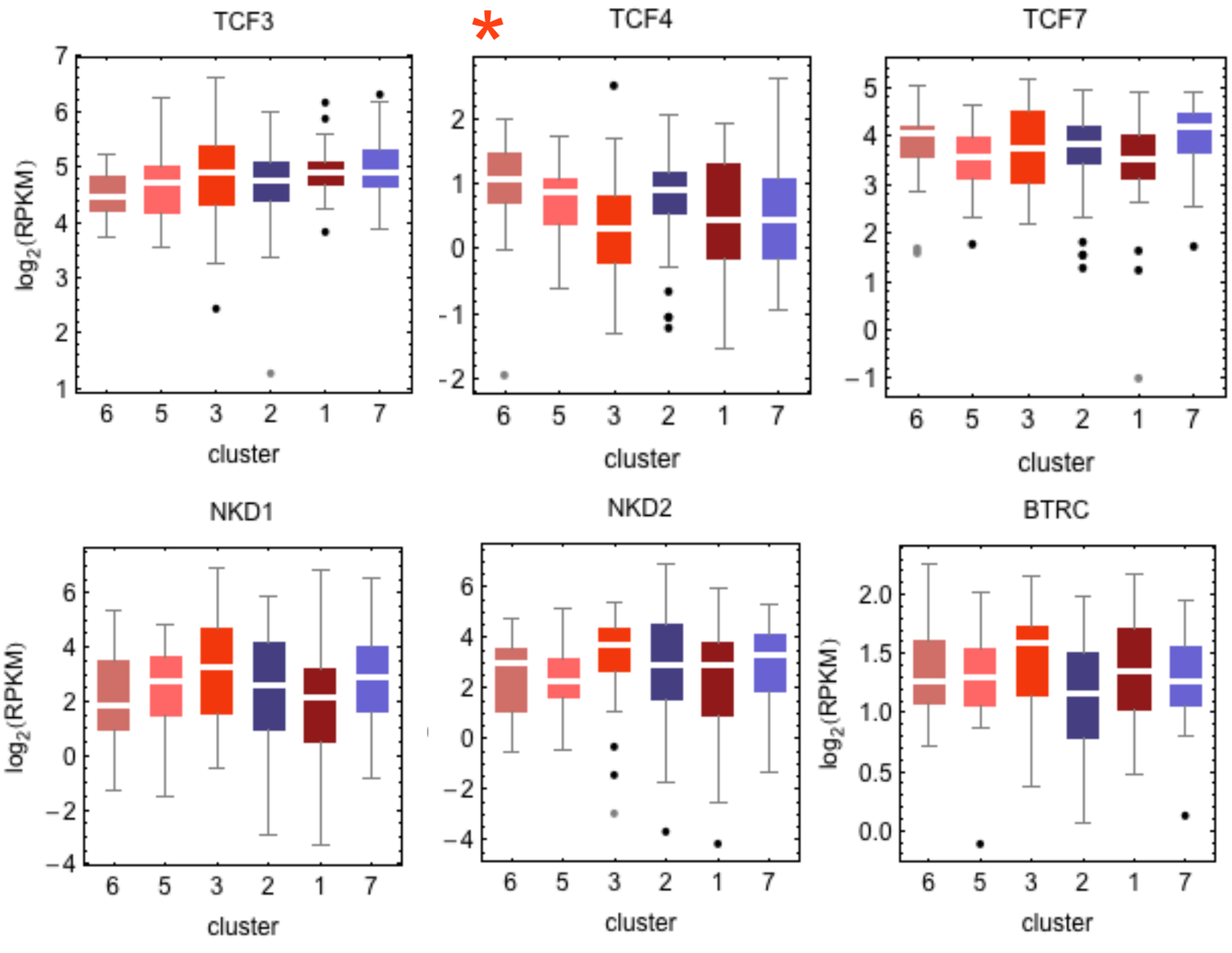


TCFs, NKDs: RPKM abundance across 7 clusters

grobertson:mRNA-seq grobertson$ grep "TCF" crc_244_gene_rpkm.txt |
TCF3|6929
TCF4|6925
TCF7|6932
TCF12|6938
TCF15|6939
TCF19|6941
TCF20|6942
TCF21|6943
TCF23|150921
TCF25|22980
TCF7L1|83439
TCF7L2|6934
TCFL5|10732







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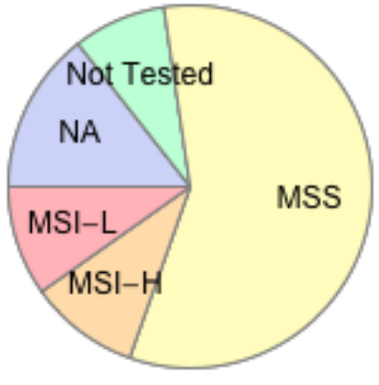
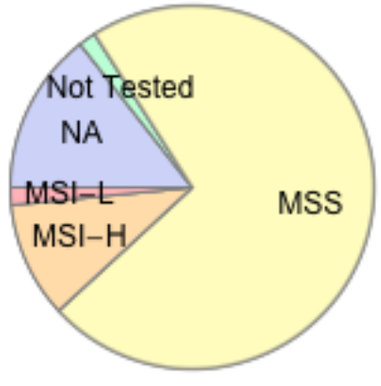
CRC groupings v2 (194 samples)

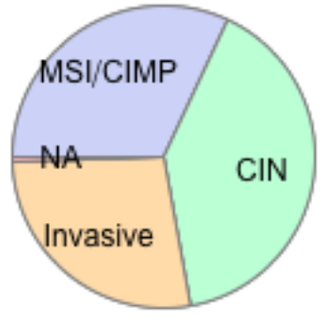
MSS, MSI, CIMP, CIN



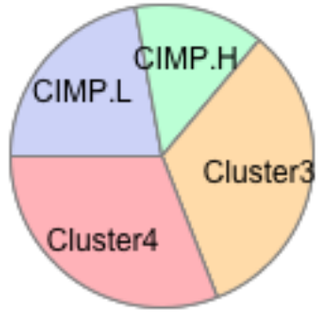
MSI (MDA)	
MSI-H	19
MSI-L	19
MSS	112
NA	28
Not Tested	16



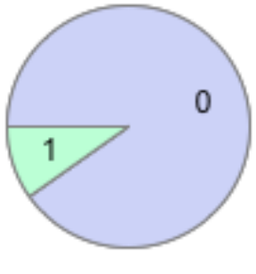
MSI (MDA)	
MSI-H	20
MSI-L	3
MSS	140
NA	28
Not Tested	3



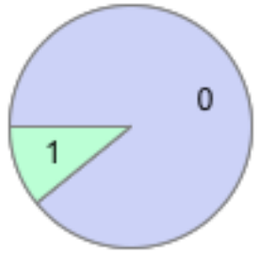
mRNA cluster	
CIN	78
Invasive	53
MSI/CIMP	62
NA	1



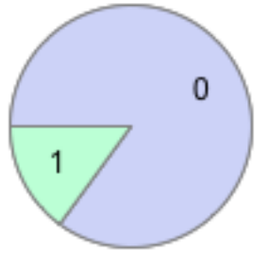
Meth cluster	
CIMP.H	27
CIMP.L	43
Cluster3	64
Cluster4	60



MLH1 sil	
0	175
1	19

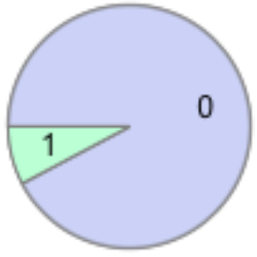


MLH1 hyper	
0	173
1	21

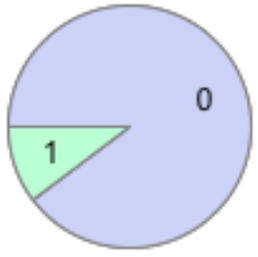


Hypermut	
0	165
1	29

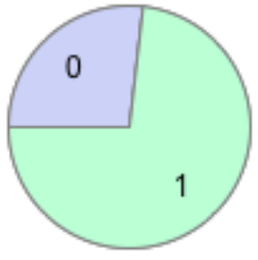
MLH1, hypermutation



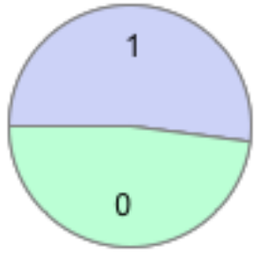
FBXW7 mut	
0	179
1	15



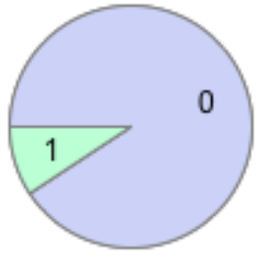
PIK3CA mut	
0	174
1	20



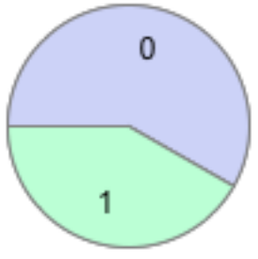
APC mut	
0	52
1	142



TP53 mut	
0	93
1	101

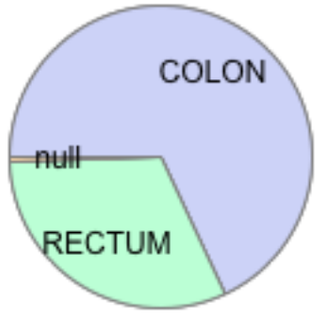


BRAF mut	
0	176
1	18

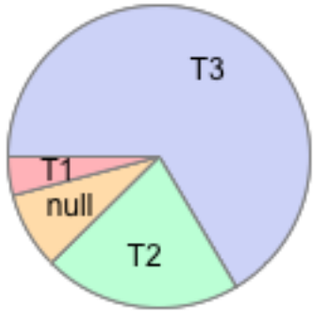


KRAS mut	
0	113
1	81

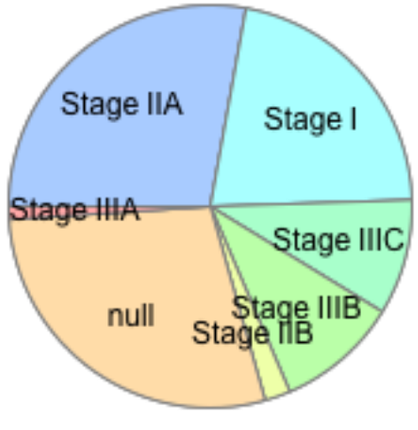
Key mutated genes



Tissue site	
COLON	132
null	1
RECTUM	61



Tumor spread	
null	16
T1	8
T2	41
T3	129



Tumor stage	
null	55
Stage I	42
Stage IIA	54
Stage IIB	4
Stage IIIA	2
Stage IIIB	19
Stage IIIC	18

Tumor spread and stage