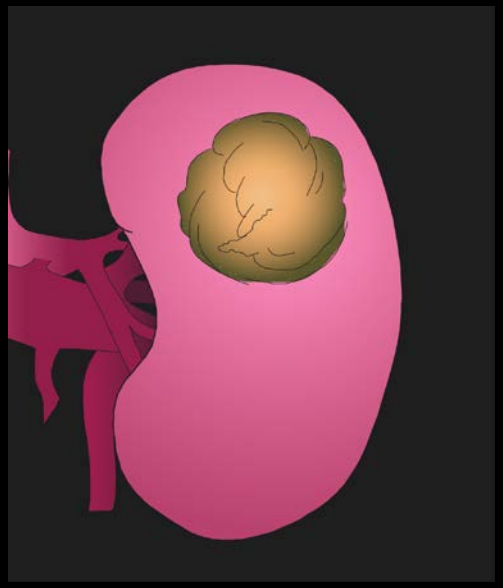
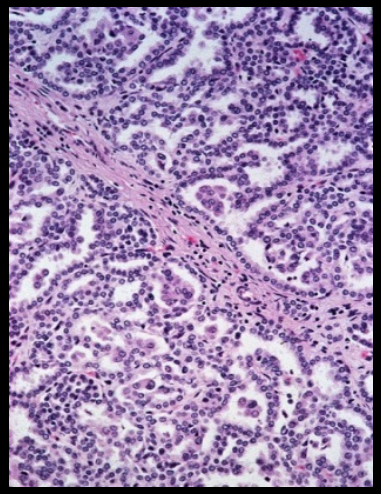
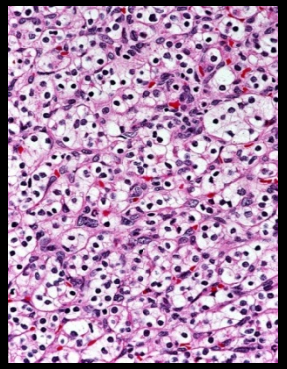
Comprehensive Molecular Characterization Papillary Renal Cell Carcinoma

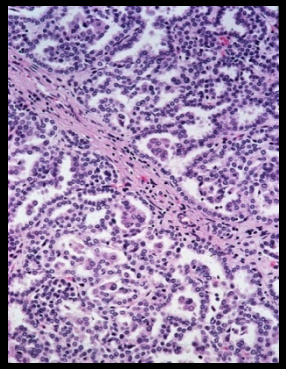
The Cancer Genome Atlas Research Network

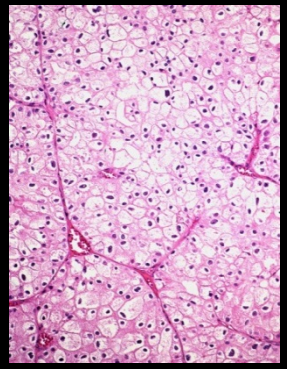


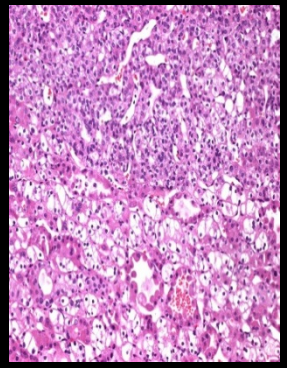


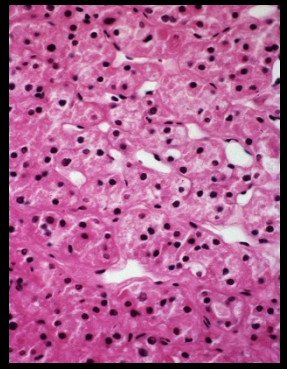
Renal Cell Carcinoma

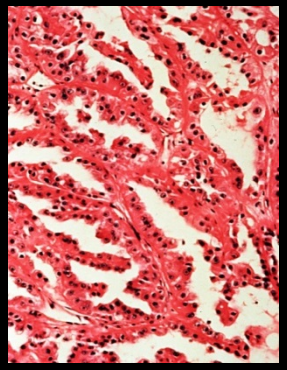

Clear Cell

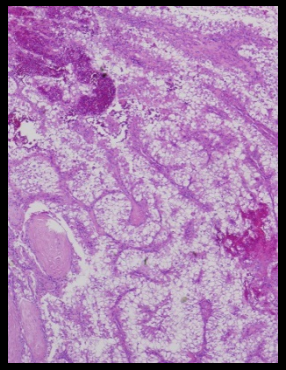

Papillary Type 1

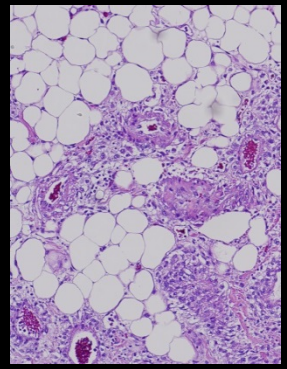

Chromophobe

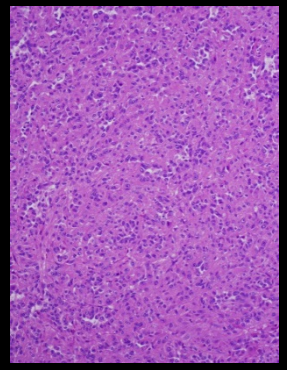

Hybrid

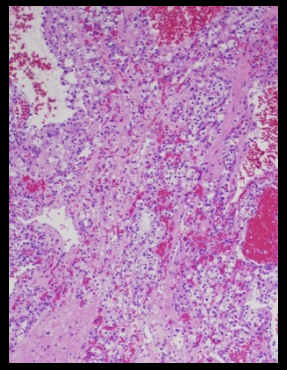

Oncocytoma


Papillary Type 2

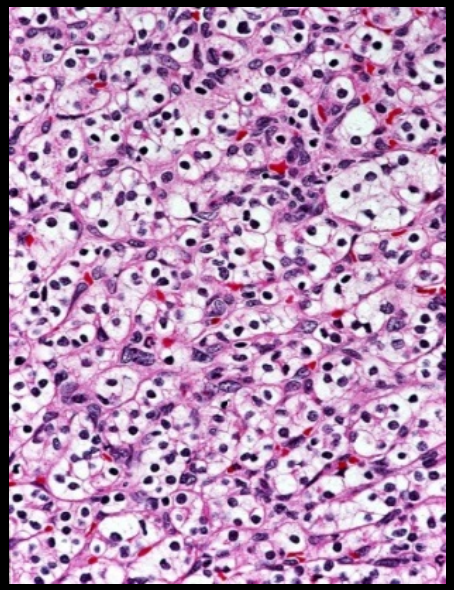

TFE3


Angiomyolipoma

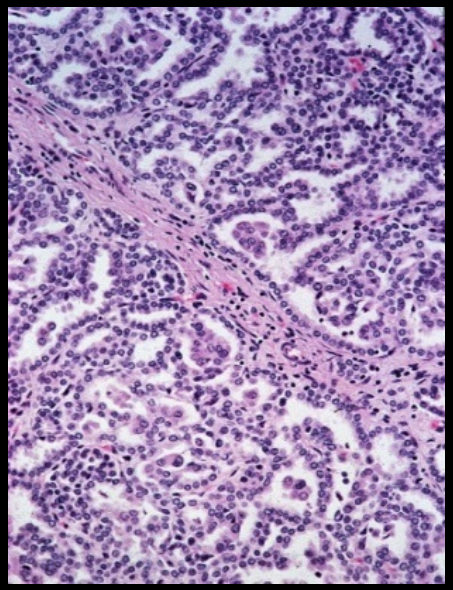

Oncocytic


Clear/Chromophobe

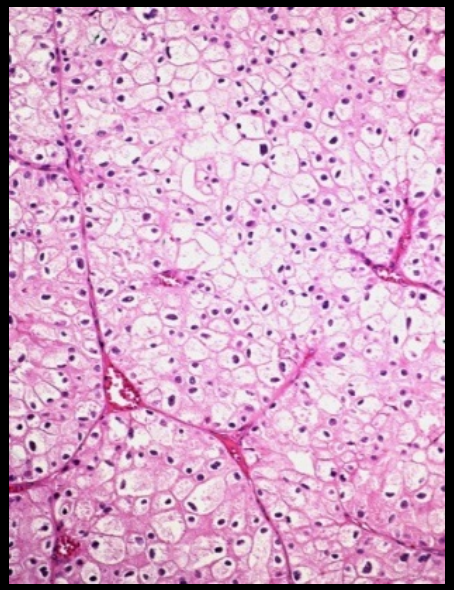
Renal Cell Carcinoma



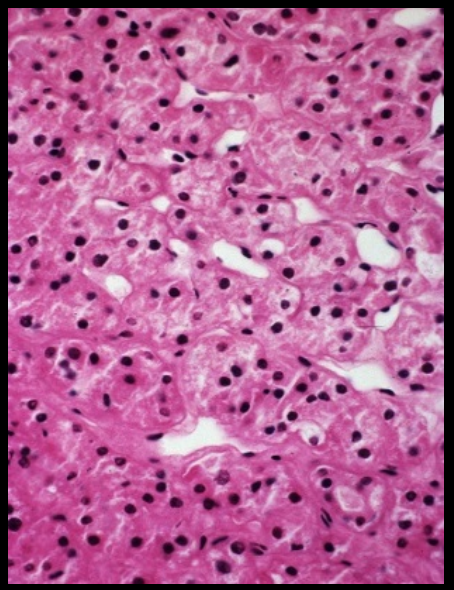
Clear Cell
75%



Papillary
15%

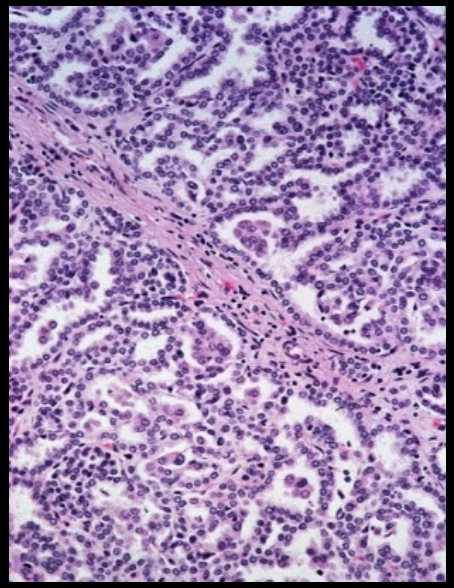


Chromophobe
5%

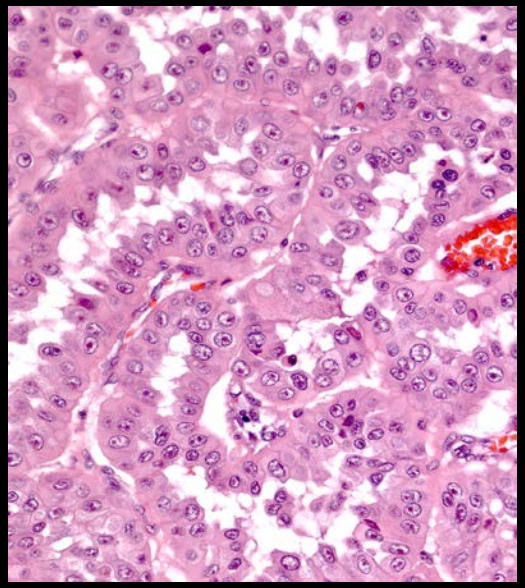


Oncocytoma
3%

Papillary Renal Cell Carcinoma

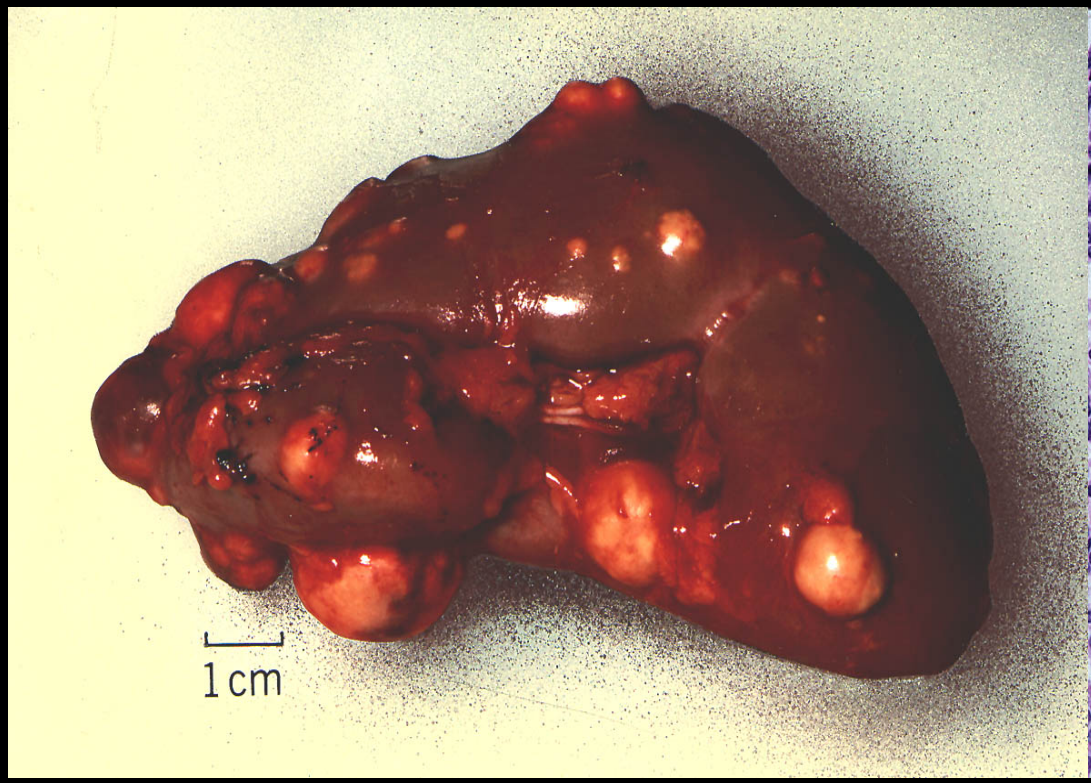


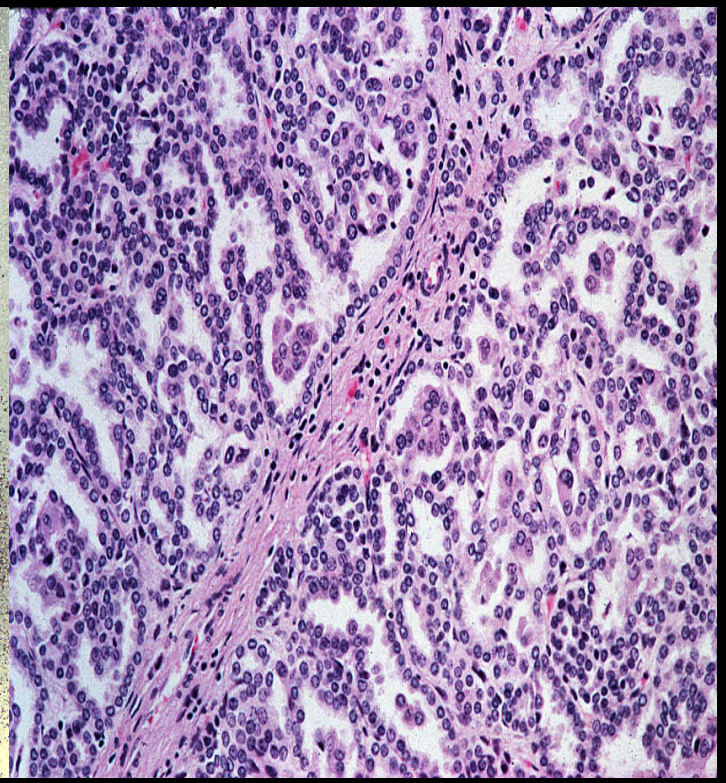
Type 1 Papillary



Type 2 Papillary

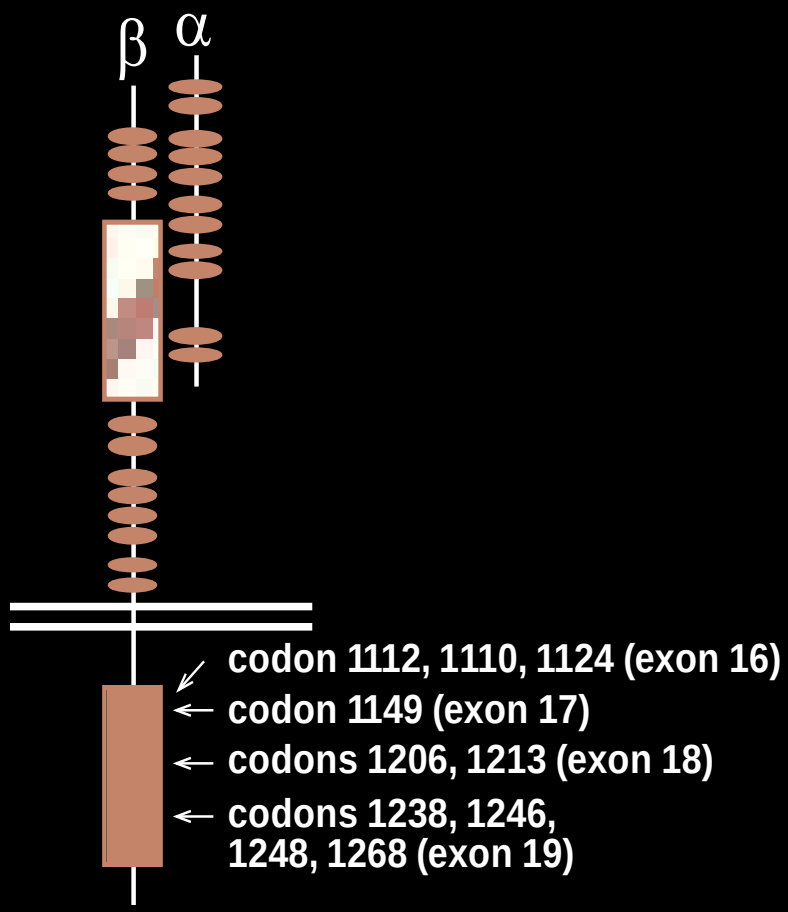
Hereditary Papillary Renal Carcinoma
Type 1 Papillary Renal Carcinoma



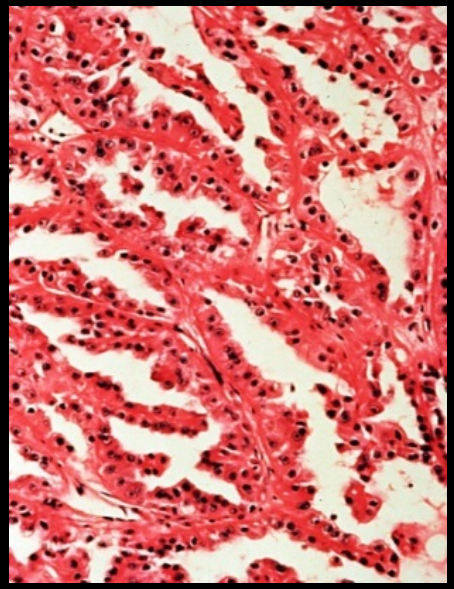


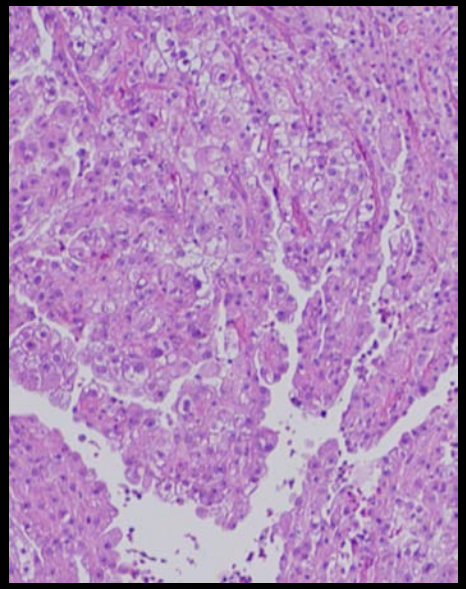
Type 1 Papillary RCC

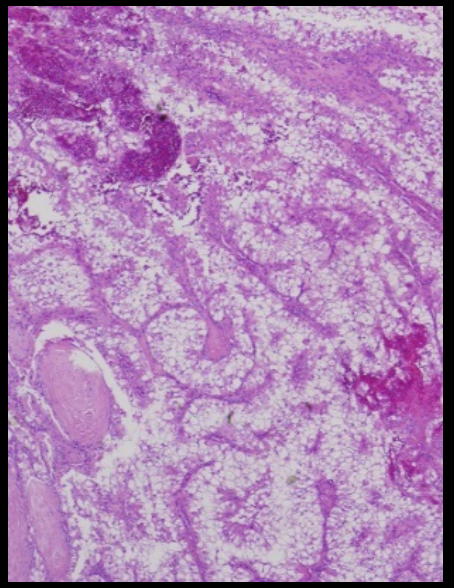
HPRC: *MET* Mutations



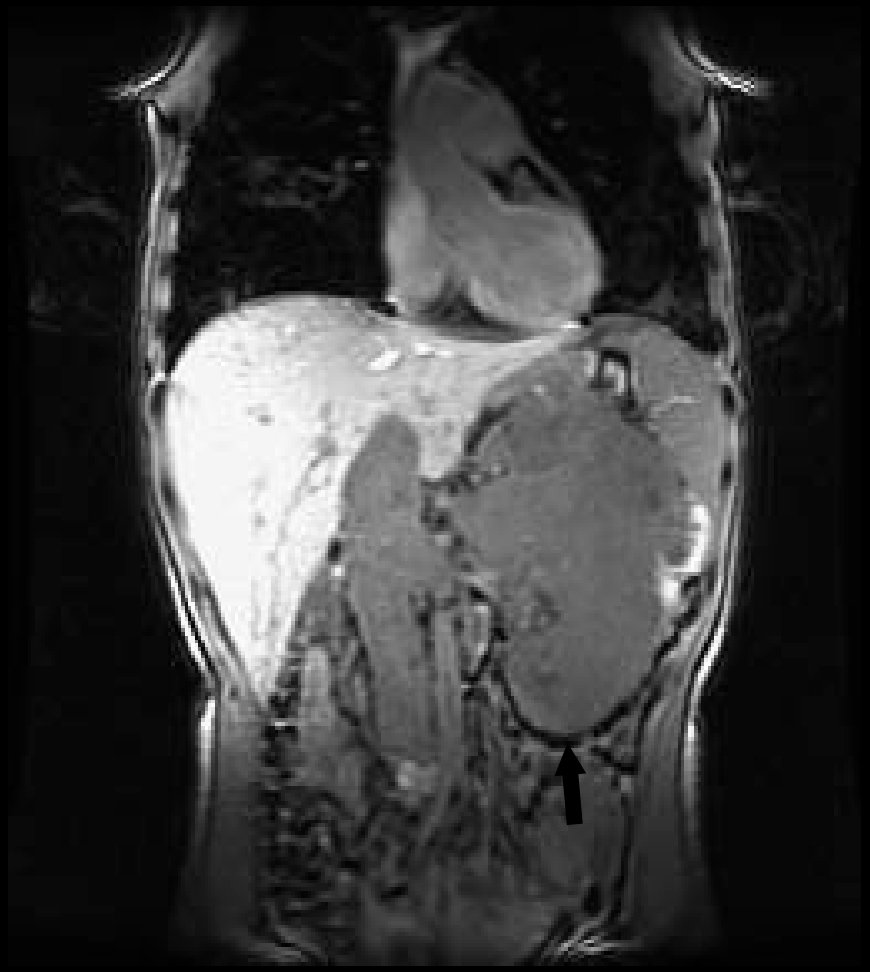
Type 2 Papillary RCC is Heterogeneous

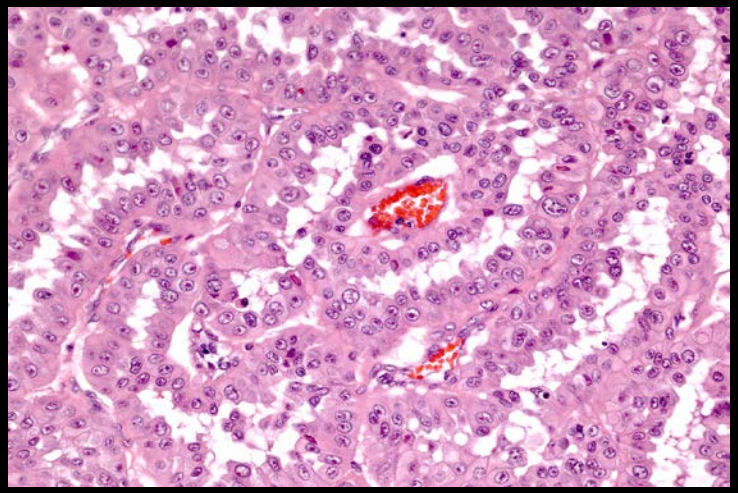






Hereditary Leiomyomatosis Renal Cell Carcinoma (HLRCC)

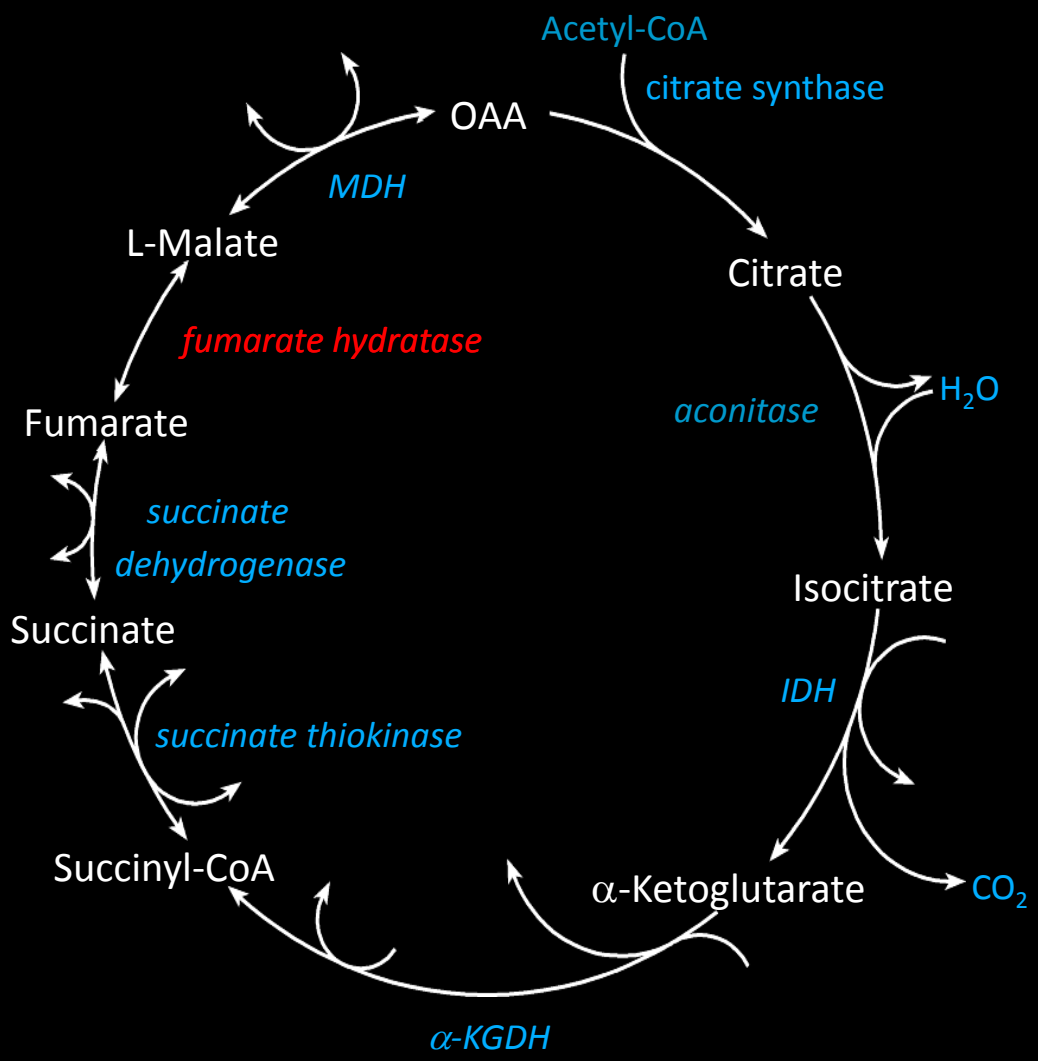




Type 2 Papillary RCC

Fumarate Hydratase (FH): HLRCC Gene

FH catalyzes the conversion of fumarate to malate

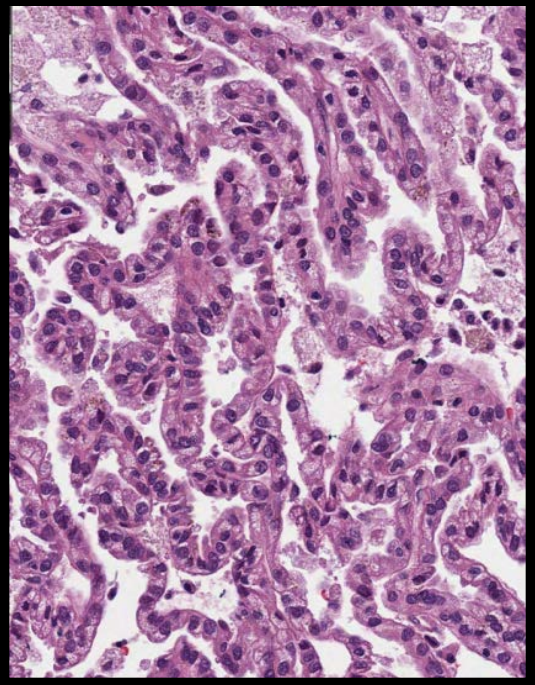


Tomlinson, et al. Nature Genetics:30 2002

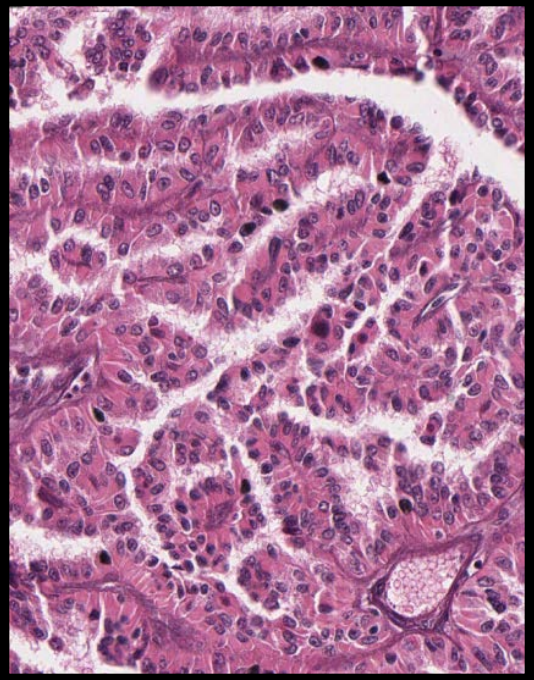
KIRP Analysis

Analysis Type	Method of Analysis	Samples Analysed
Copy Number Analysis	SNP6.0 Arrays	161 PRCC Samples 161 Normals
Somatic Mutation Analysis	Exome Sequencing	157 PRCC Samples 157 Normals
Methylation Analysis	Illumina BeadChip Assays	161 PRCC Samples 45 Normals
mRNA Expression Analysis	RNA-Seq	161 PRCC Samples 30 Normals
miRNA Expression Analysis	RNA-Seq	161 PRCC Samples 32 Normals
Protein Expression Analysis	Reverse phase protein array (RPPA)	125 PRCC Samples

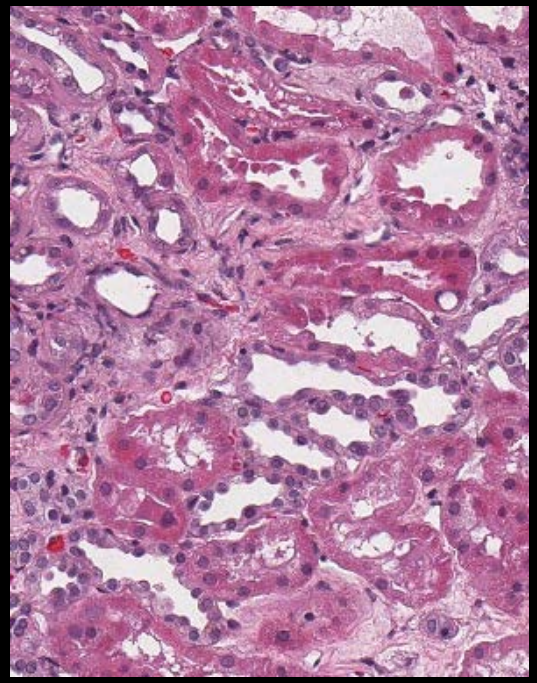
Pathology Analysis: N=161



TCGA-A4-7732
Type 1 PRCC
N=75



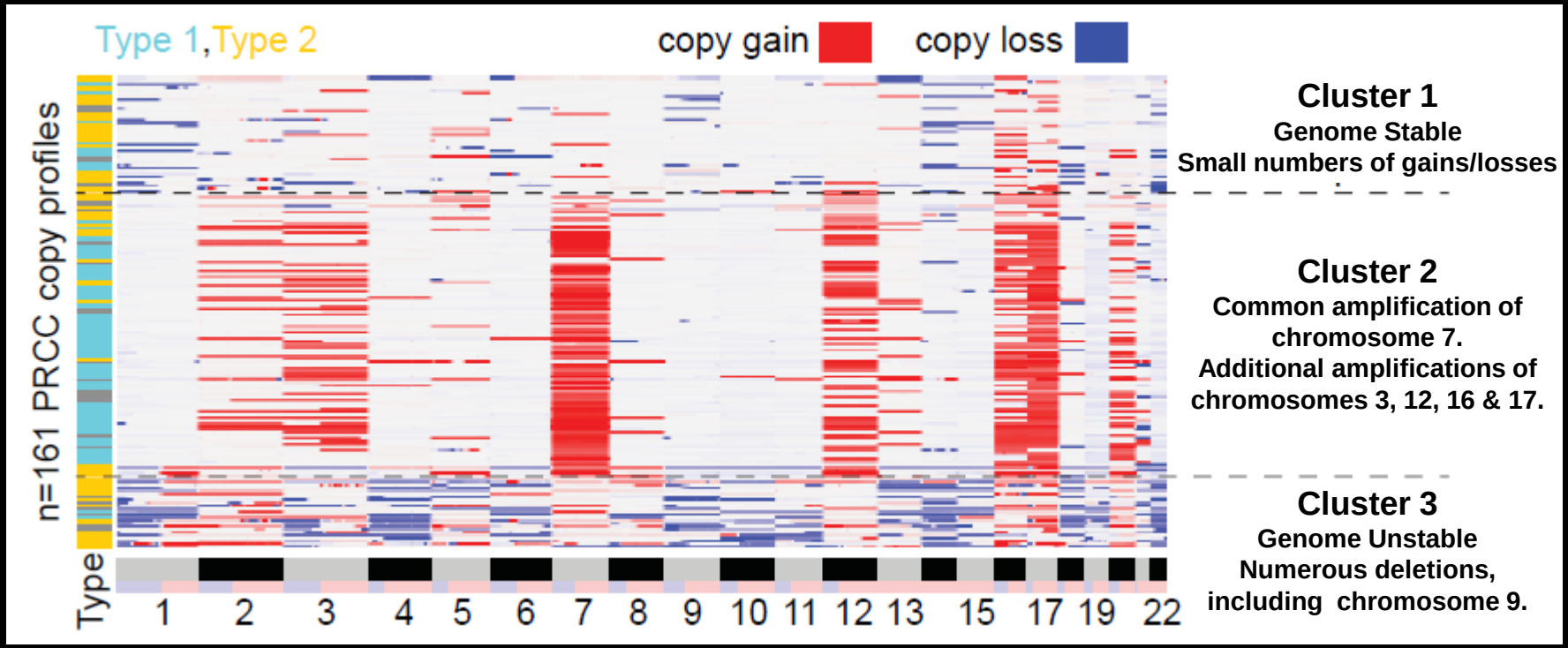
TCGA-BQ-5878
Type 2 PRCC
N=60



TCGA-BQ-5886
Unclassified PRCC
N=26

Chromosomal Copy Number Analysis

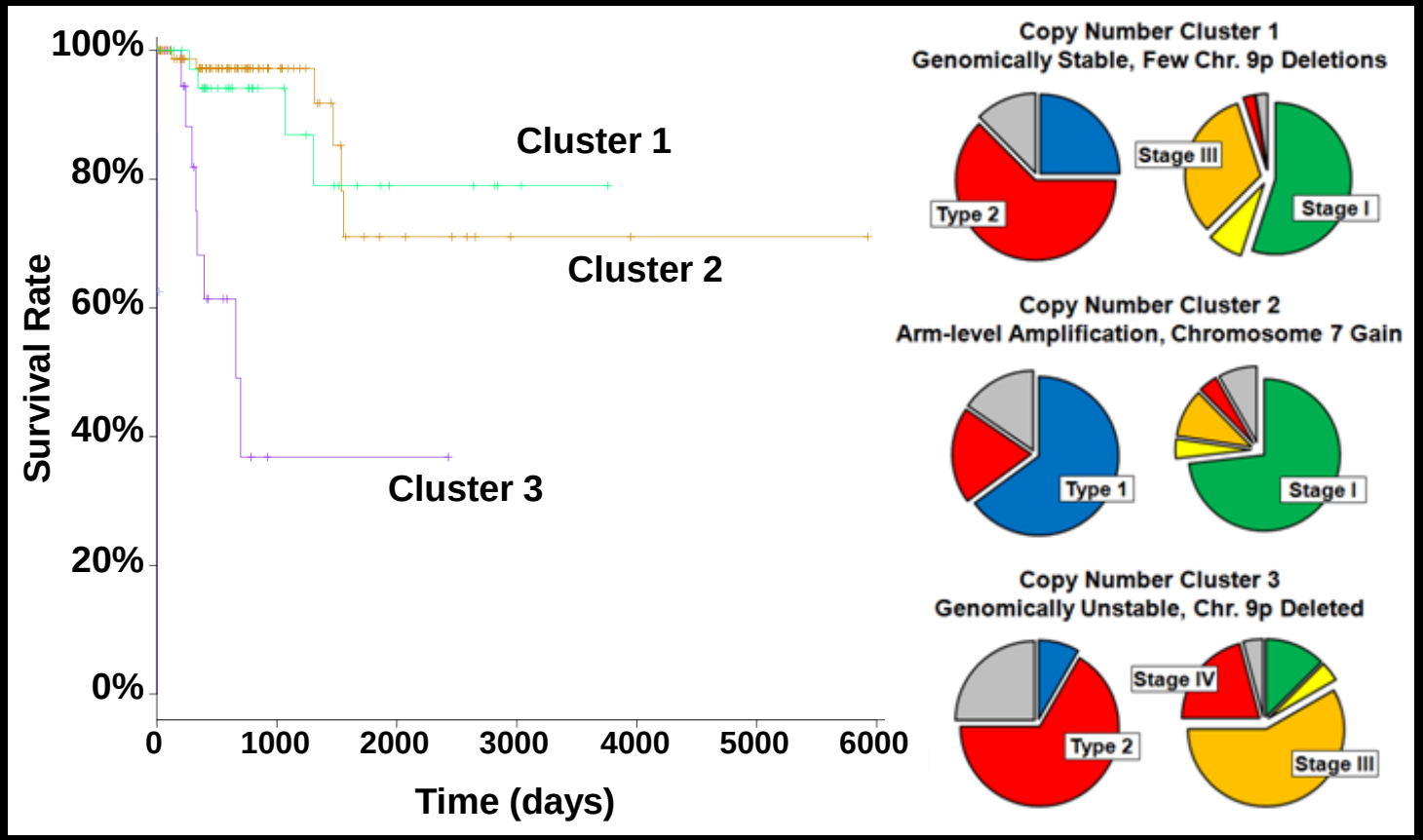
- Chromosomal level copy number analysis produced three distinct clusters
  1. Relative genomic stability
  2. Multiple chromosomal gain, notably chromosome 7
  3. Multiple deletions; including chromosome 9



- Cluster 2 predominantly Type 1 PRCC
- Cluster 1 and 3 predominantly Type 2 PRCC

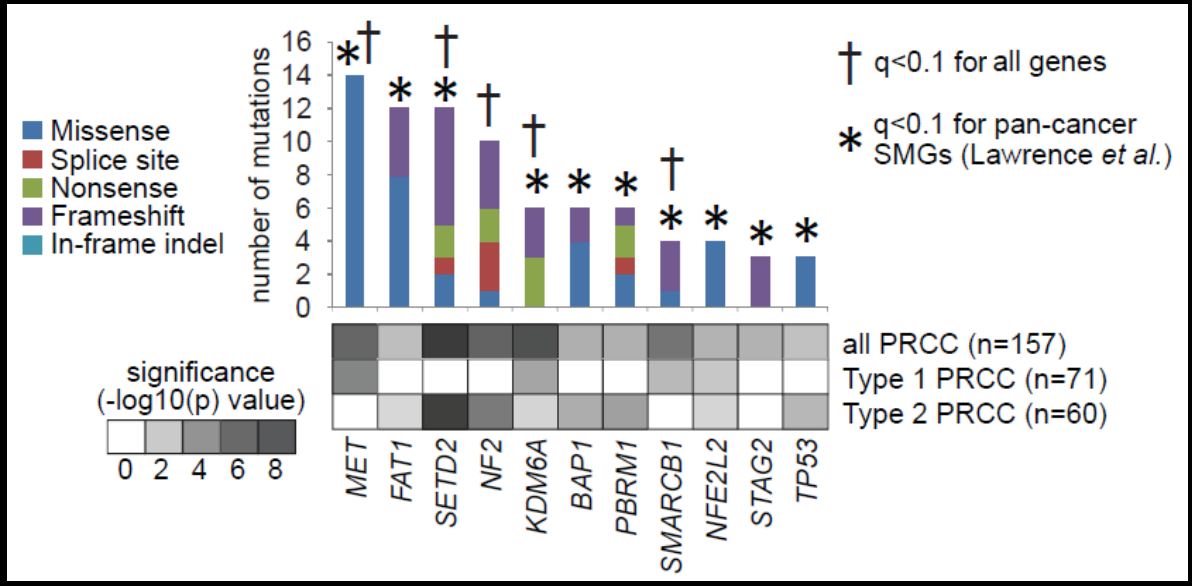
Chromosomal Copy Number Analysis

- Chromosomal level copy number analysis produced three distinct clusters
  1. Relative genomic stability
  2. Multiple chromosomal gain, notably chromosome 7
  3. Multiple deletions; including chromosome 9

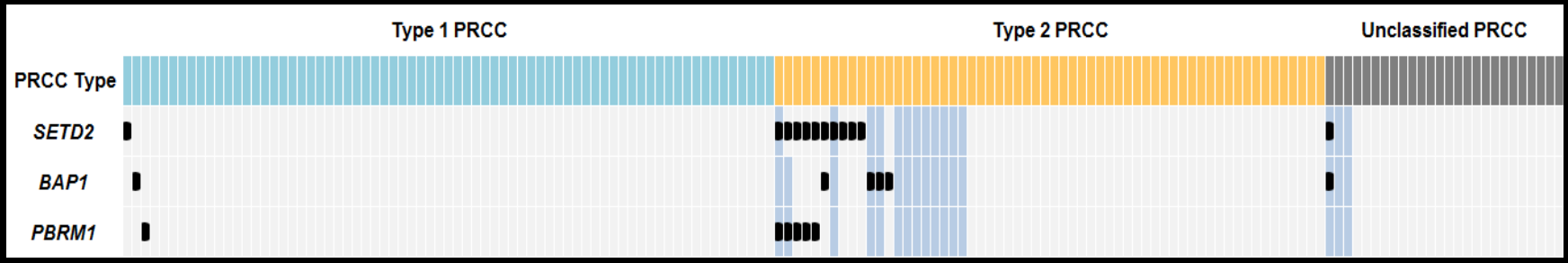


Somatic Exome Mutation Analysis

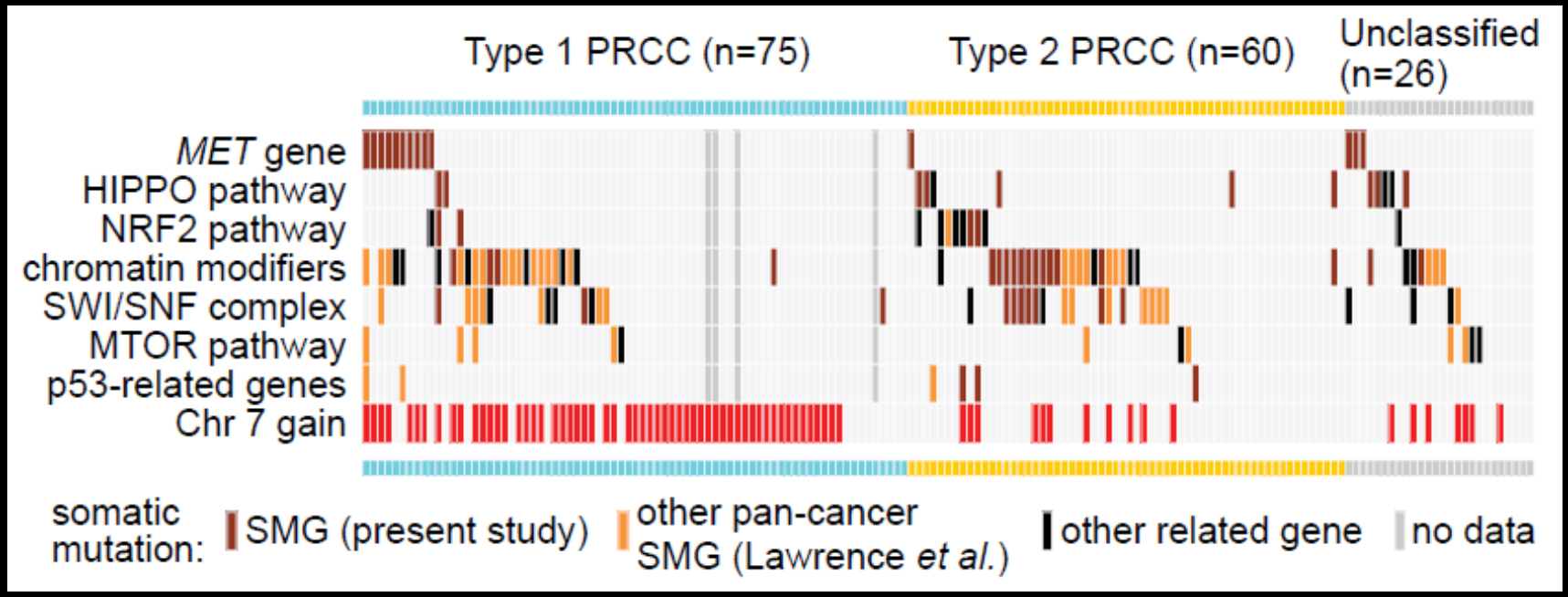
- Mutation analysis was performed using the MutSig 2.0CV with q-values <0.1
- In addition, analysis was performed to evaluate genes identified in PanCan21.



- Chromatin remodeling/modifier genes mutated in clear cell RCC were also mutated in PRCC
- Associated with Type 2 PRCC

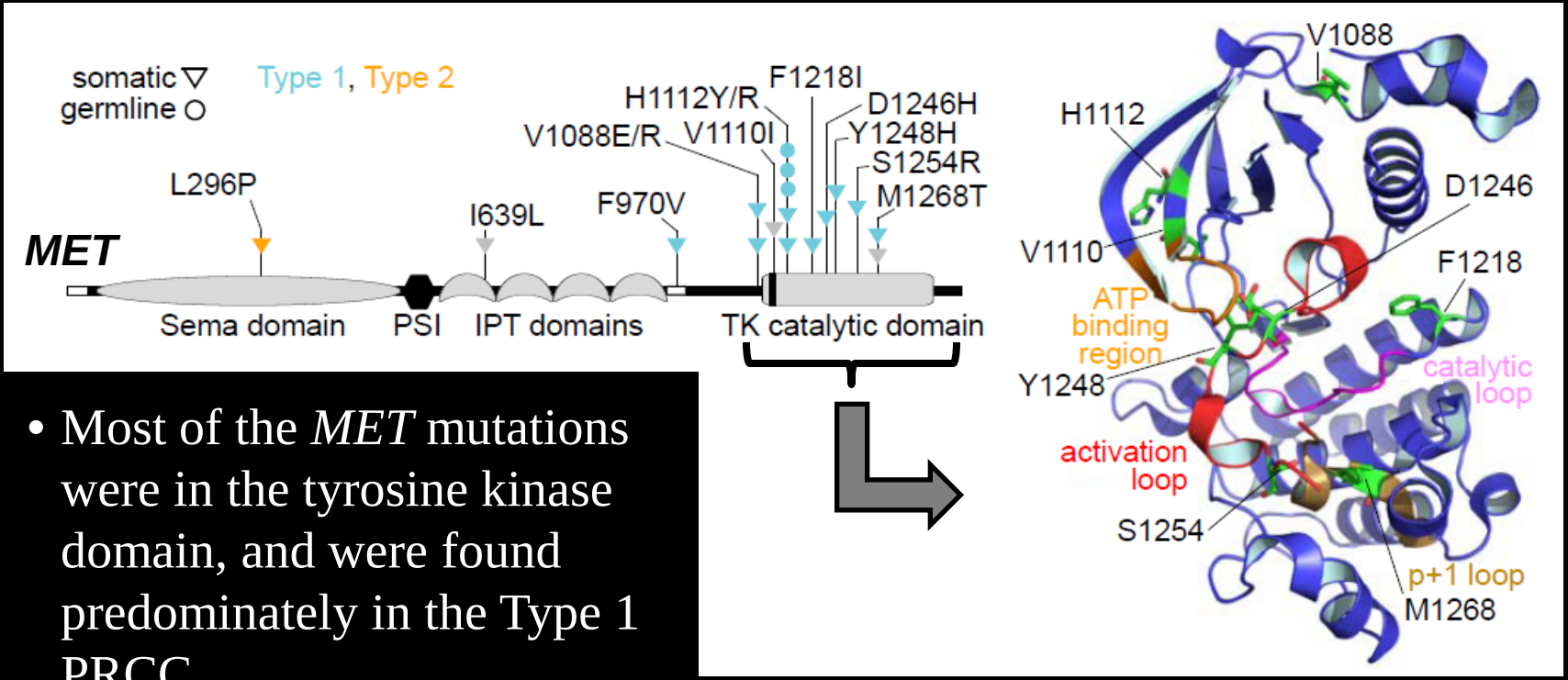


Pathway Mutation Analysis



- Several of the genes associated with PRCC exist as components of pathways or complexes, such as the Hippo pathway and several chromatin modifier pathways.
- Mutations of pathway genes were found in both Type 1 and Type 2 PRCC
  - SWI/SNF complex (20% and 27% respectively)
  - Chromatin modifier pathways (35% and 38% respectively)
  - Hippo signaling pathway (3% and 10% respectively)

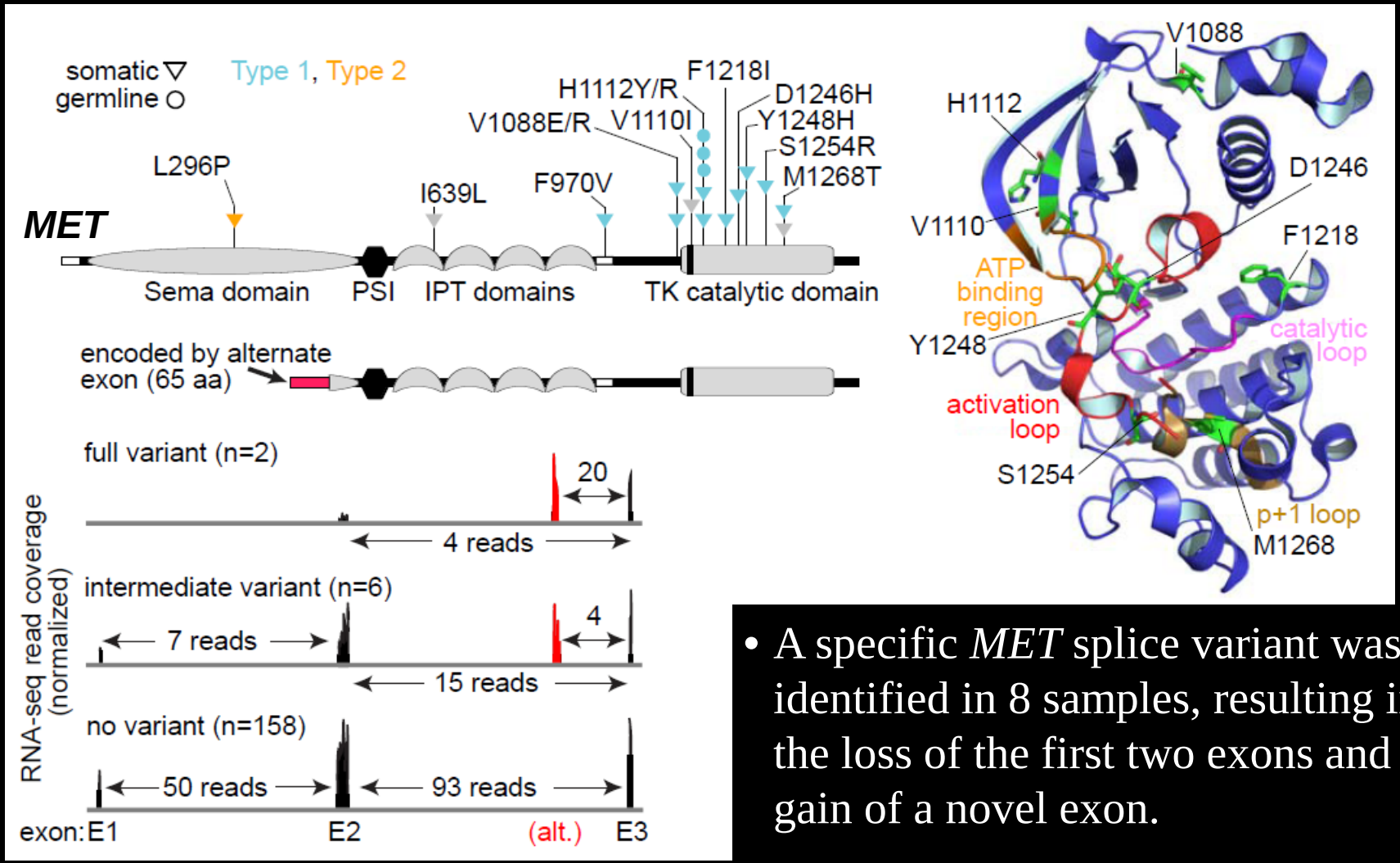
Type 1 PRCC Specific Alteration - *MET*



- Most of the *MET* mutations were in the tyrosine kinase domain, and were found predominately in the Type 1 PRCC.

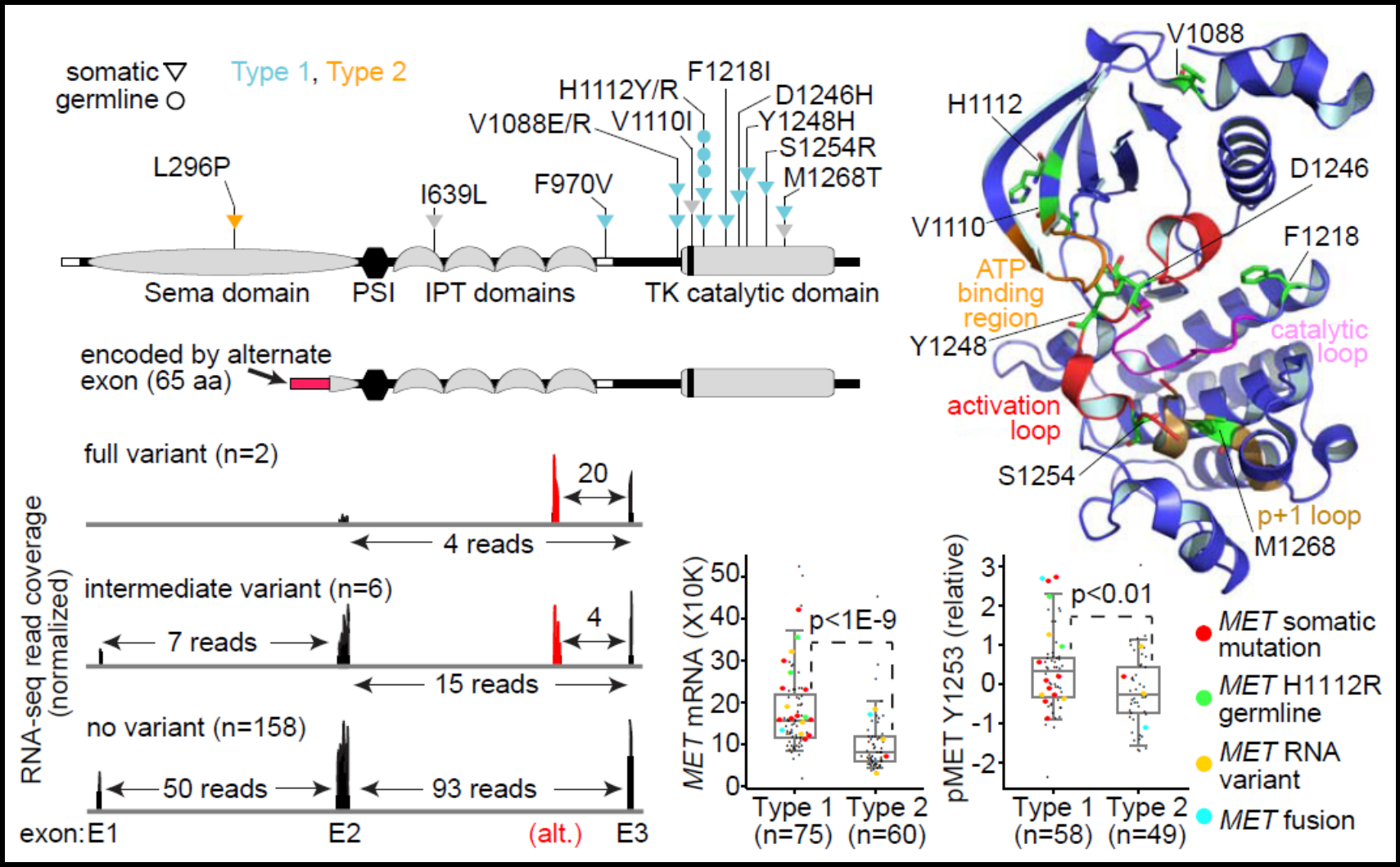
- 14 *MET* mutations were somatic; 3 were germline.

Type 1 PRCC Specific Alteration - *MET*

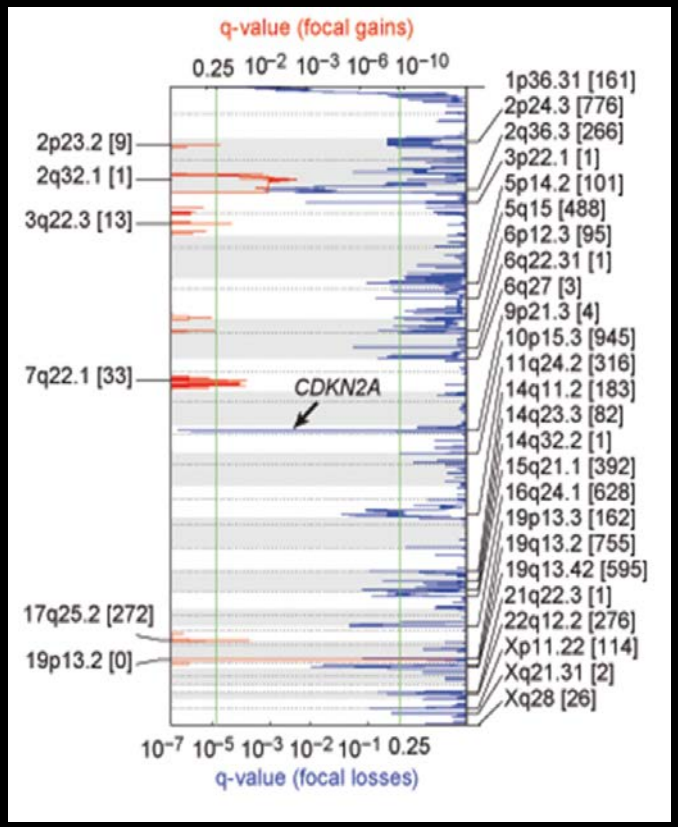


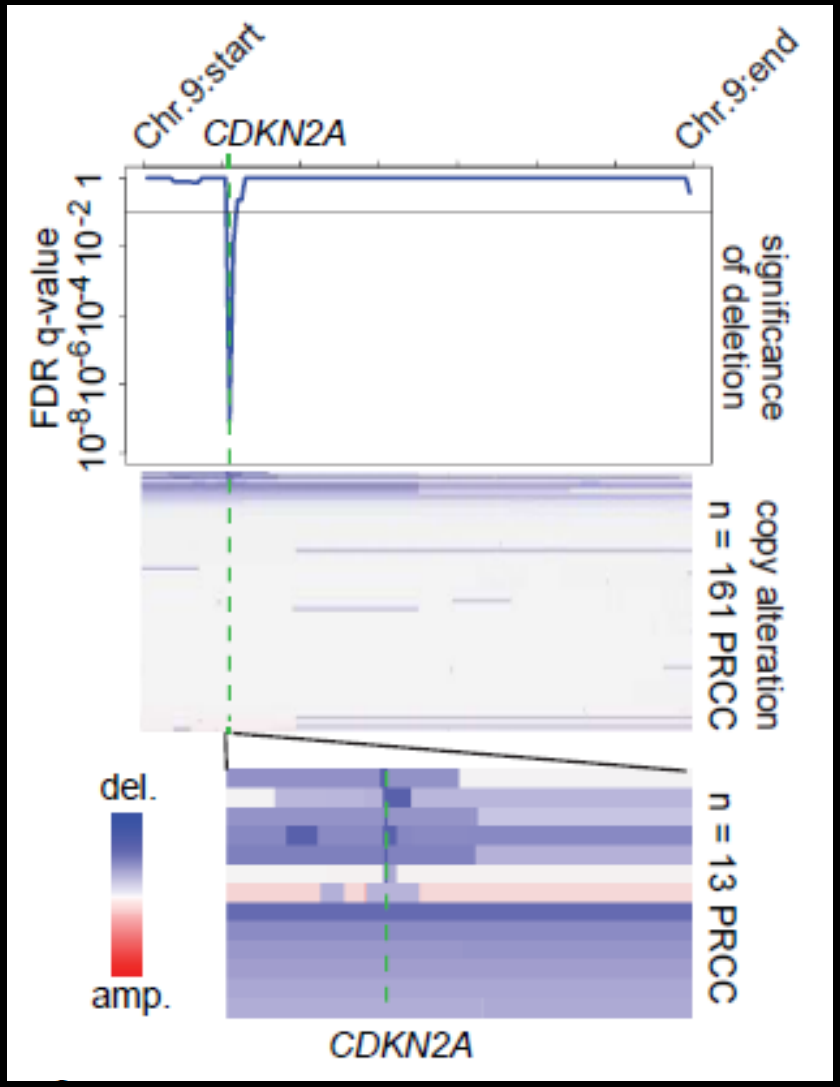
- A specific *MET* splice variant was identified in 8 samples, resulting in the loss of the first two exons and gain of a novel exon.

Type 1 PRCC Specific Alteration - *MET*



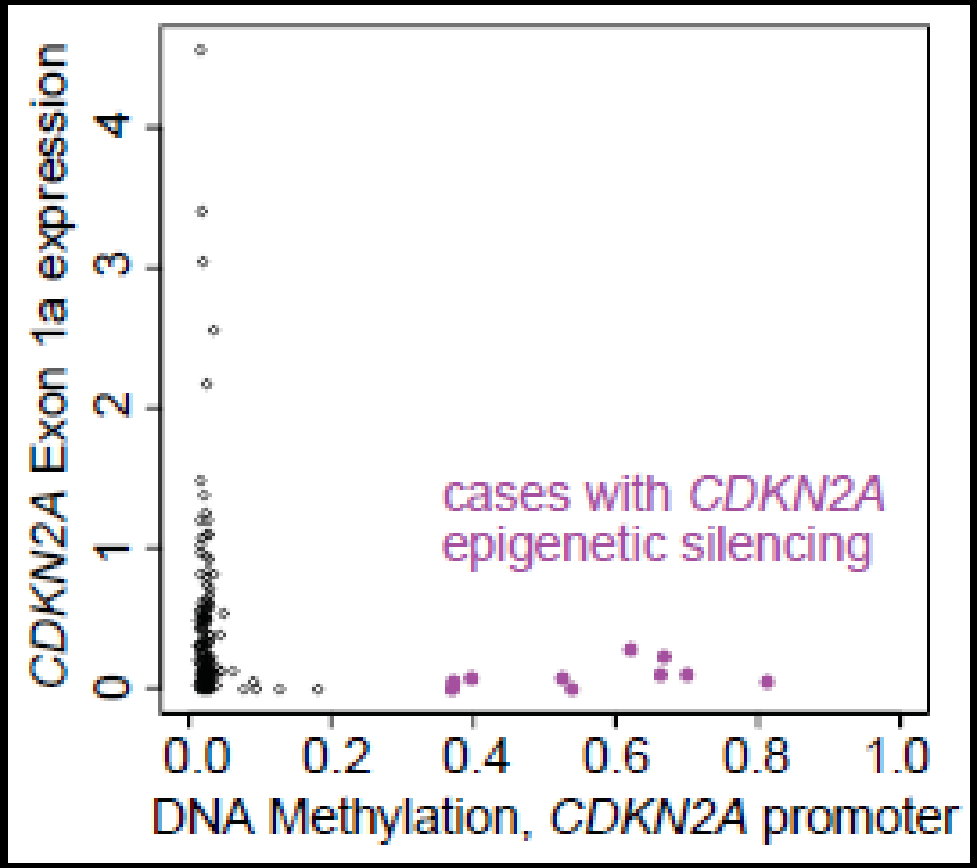
Type 2 PRCC Specific Alterations - *CDKN2A*





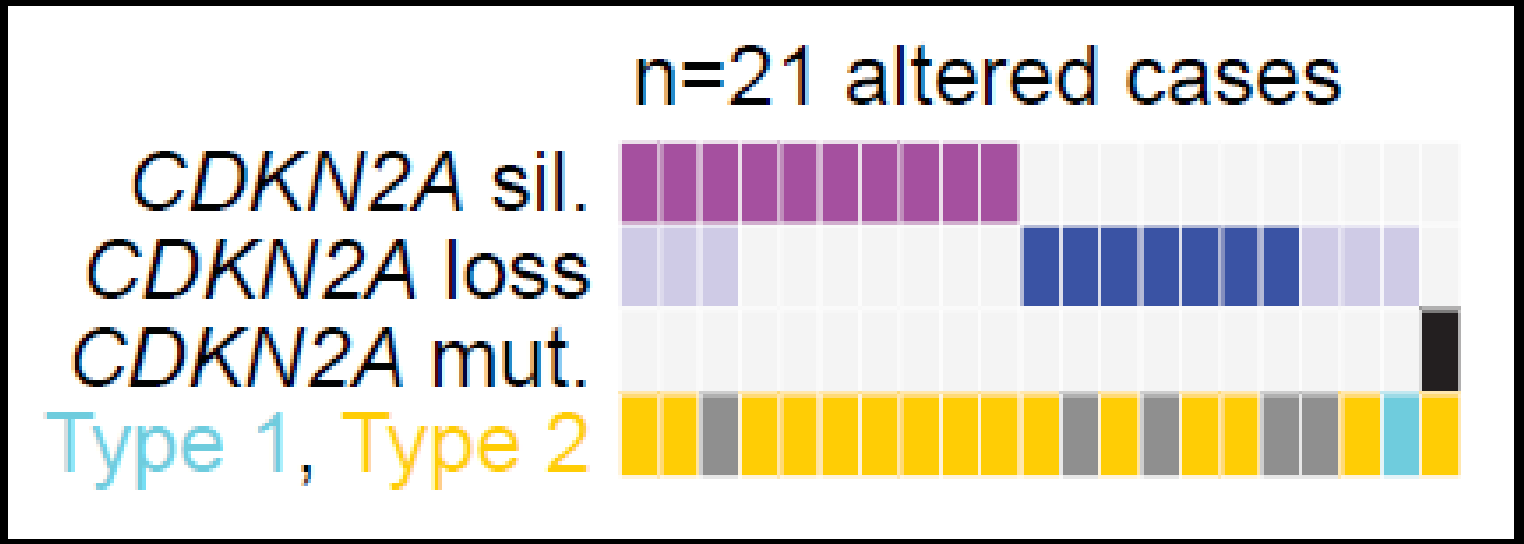
- GISTIC analysis revealed a deleted region of chromosome 9p containing the *CDKN2A* (p16) gene.

Type 2 PRCC Specific Alterations - *CDKN2A*



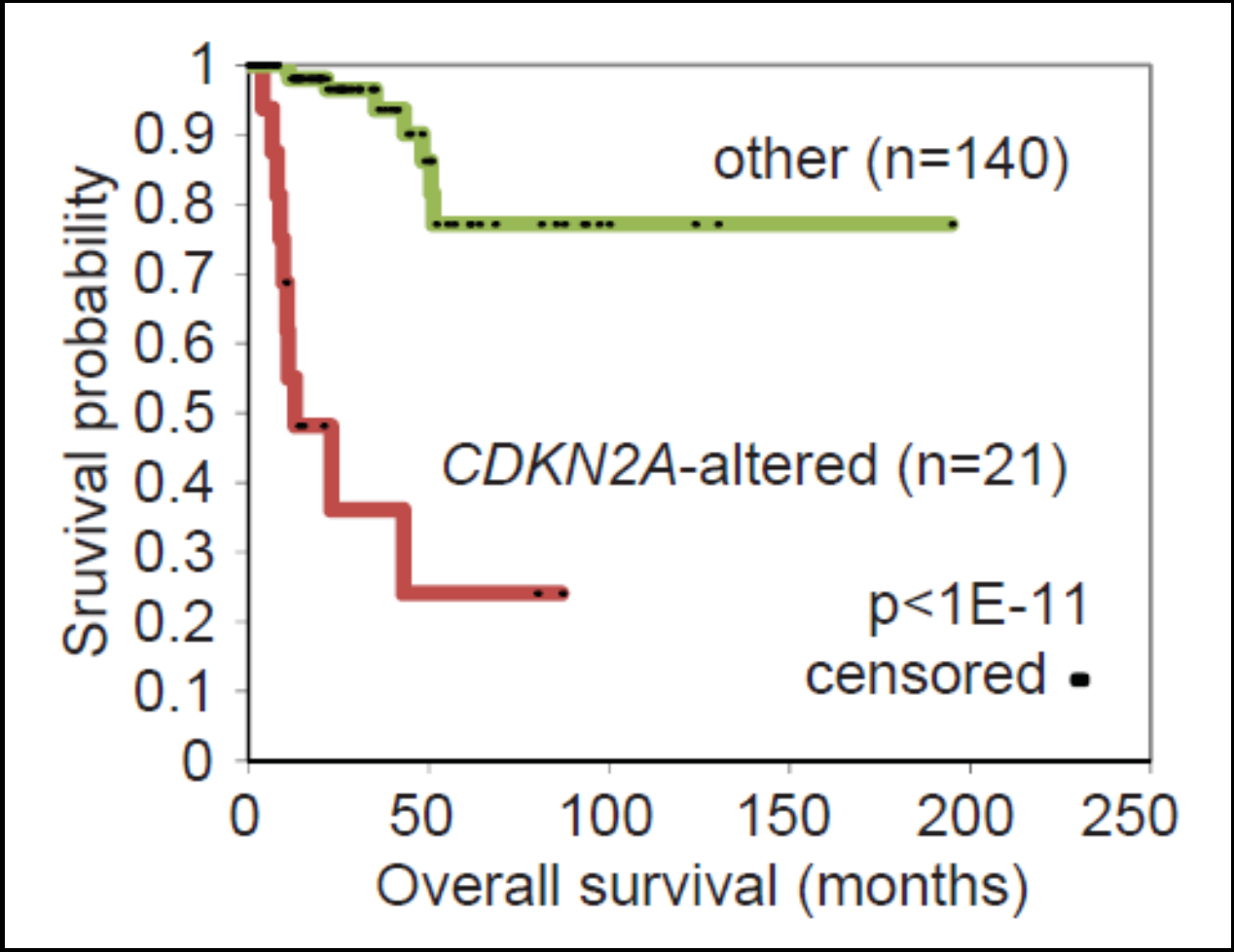
- *CDKN2A* promoter hypermethylation was identified in 10 tumors.
- Each correlated with low expression.

Type 2 PRCC Specific Alterations - *CDKN2A*



- *CDKN2A* gene alterations were found in 21 tumors
- 15 (71%) were Type 2 PRCC

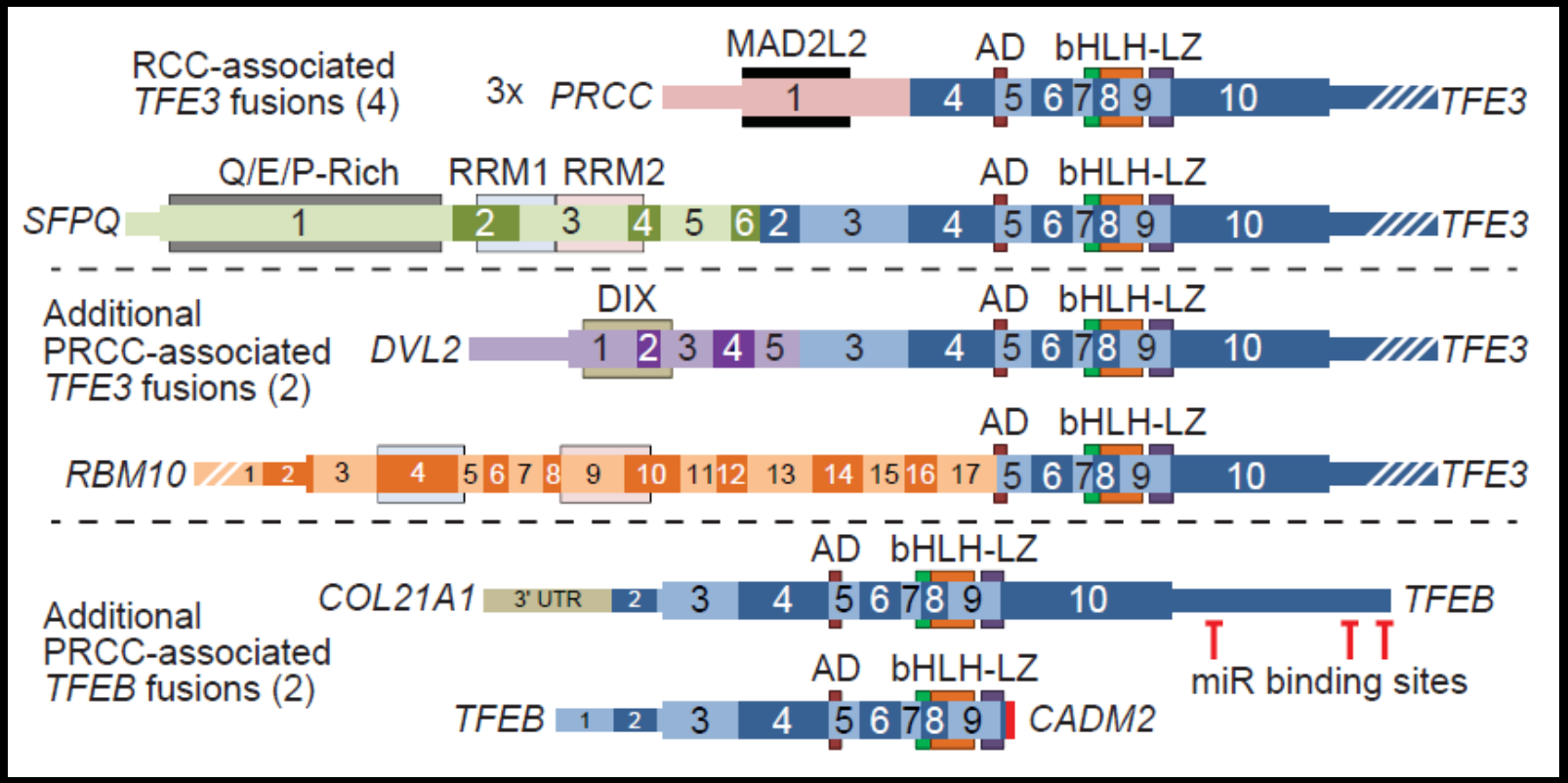
Type 2 PRCC Specific Alterations - *CDKN2A*



- Patients with *CDKN2A* alterations had poorer overall survival.

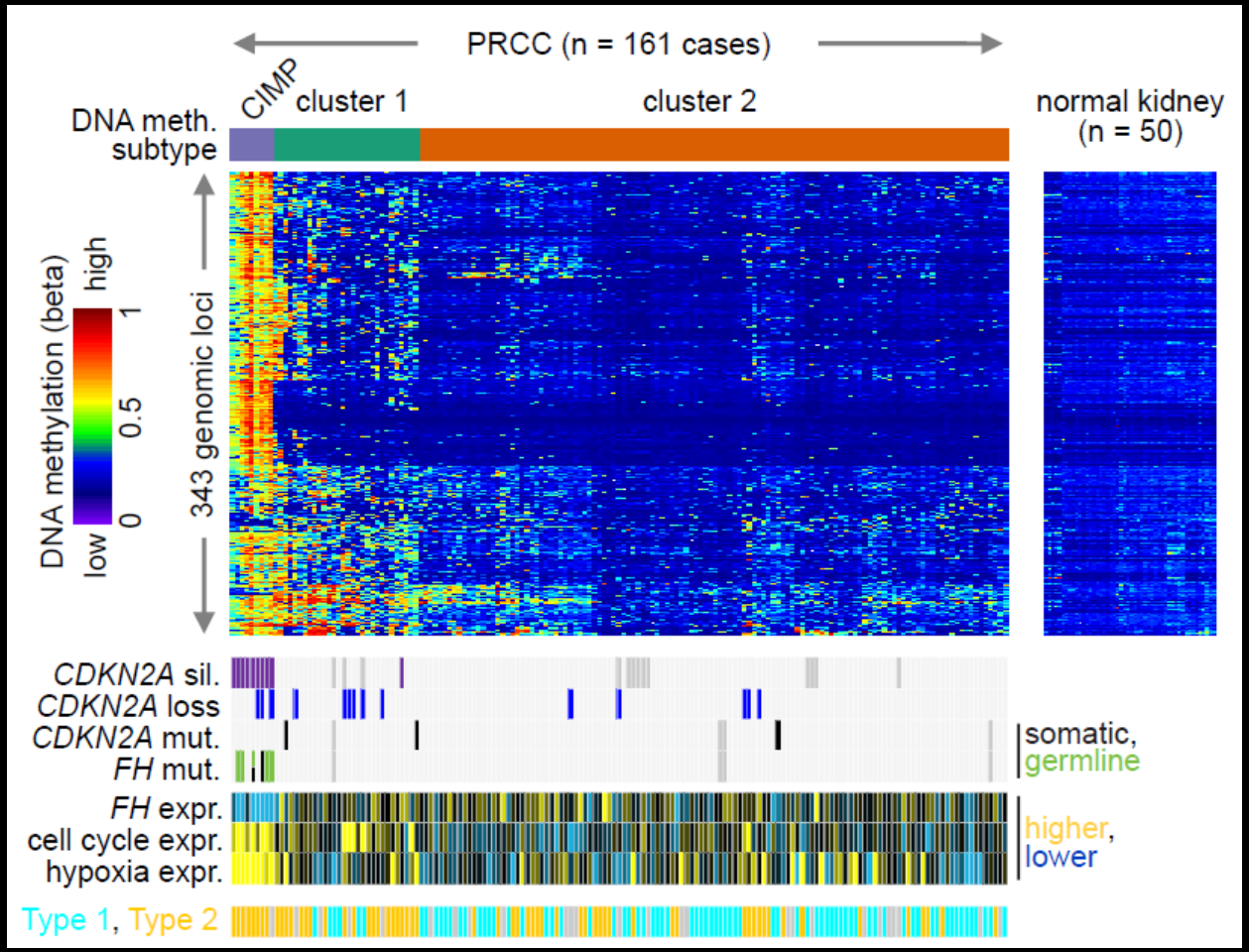
TFE3/TFEB Fusion PRCC

- TFE3/TFEB gene fusions were identified in 12% of Type 2 PRCC tumors, including patients in their 7th and 8th decade.
- The *TFE3* fusions included 4 with known fusion partners (*PRCC* and *SFPQ)* and 2 with novel fusion partners, *RBM10* and *DVL2*.
- The two *TFEB* fusions both involved novel fusion partners, *COL21A1* and *CADM2*.

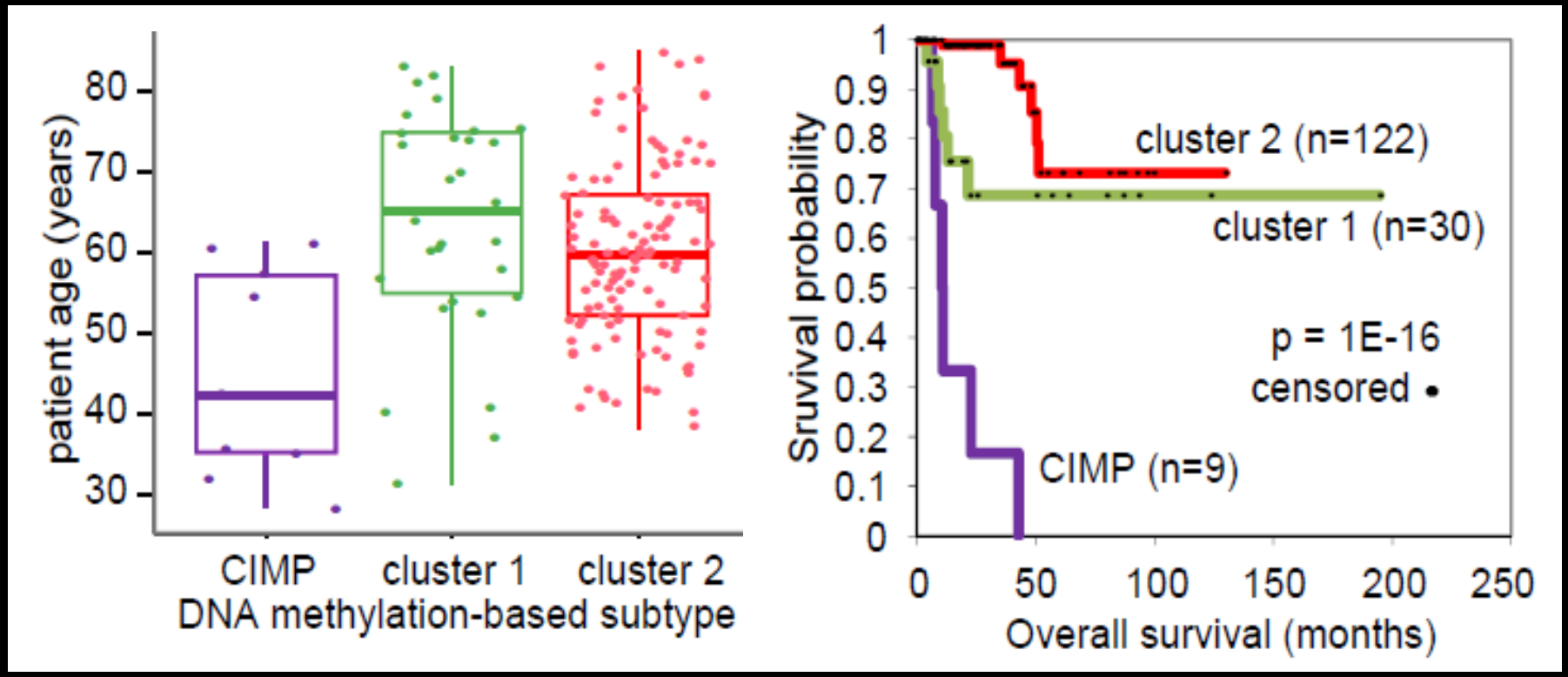


Methylation Analysis

- Assessment of the global methylation patterns separated samples into 3 clusters
- One of which demonstrated the CpG Island Methylator Phenotype (CIMP).
- Eight of 9 CIMP PRCC samples were Type 2 PRCC.
- CIMP phenotype strongly associated with somatic and germline FH mutation, low *FH* expression



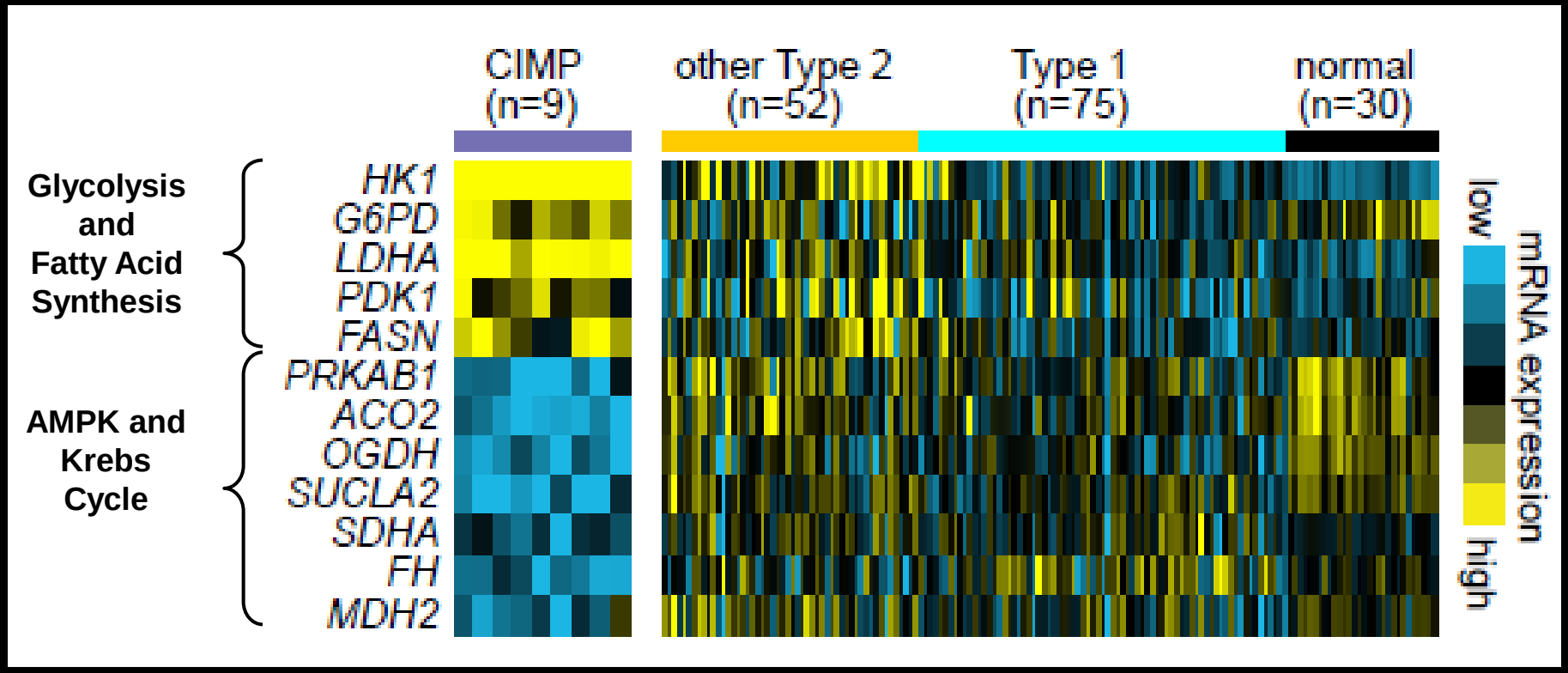
CIMP PRCC Phenotype



Early Onset

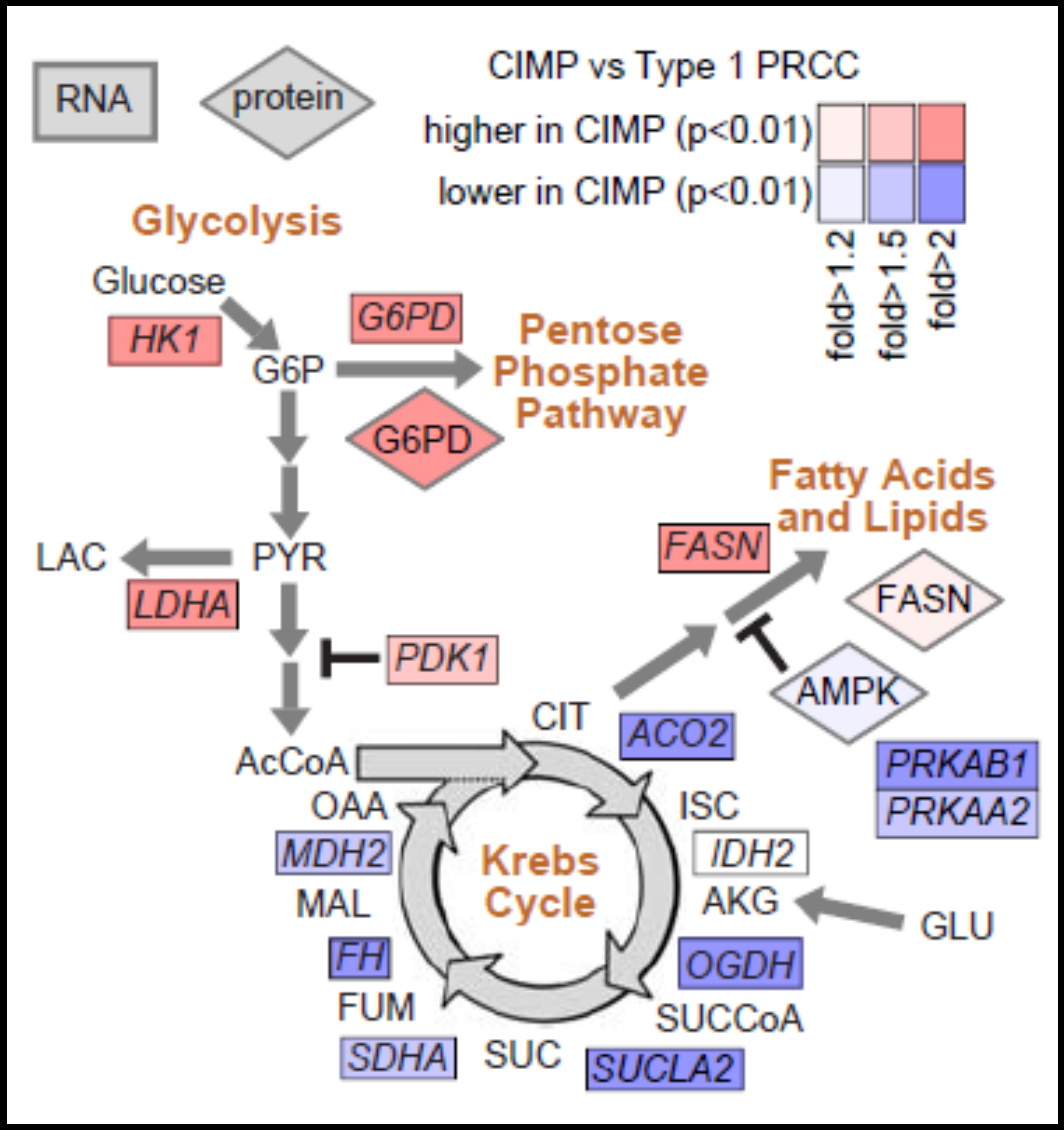
Low Survival

CIMP PRCC Phenotype

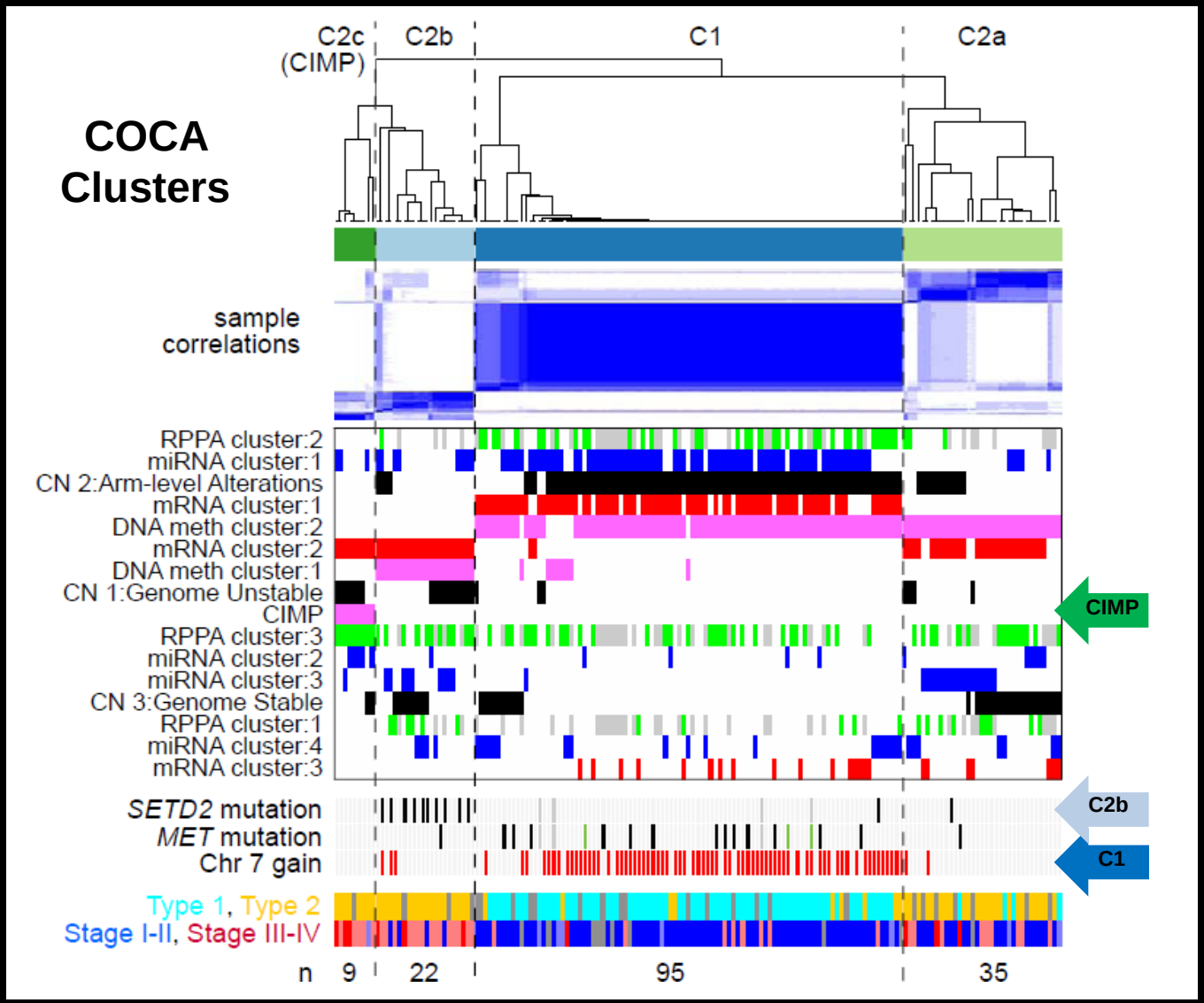


Increased Glycolysis, Fatty Acid Synthesis
Decreased TCA Cycle, Decreased AMPK

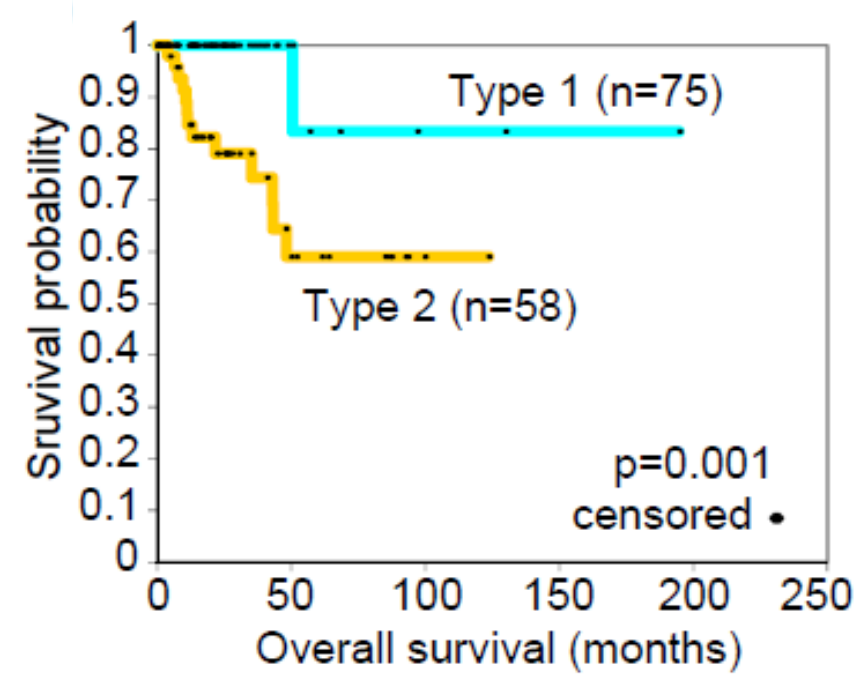
CIMP PRCC Phenotype

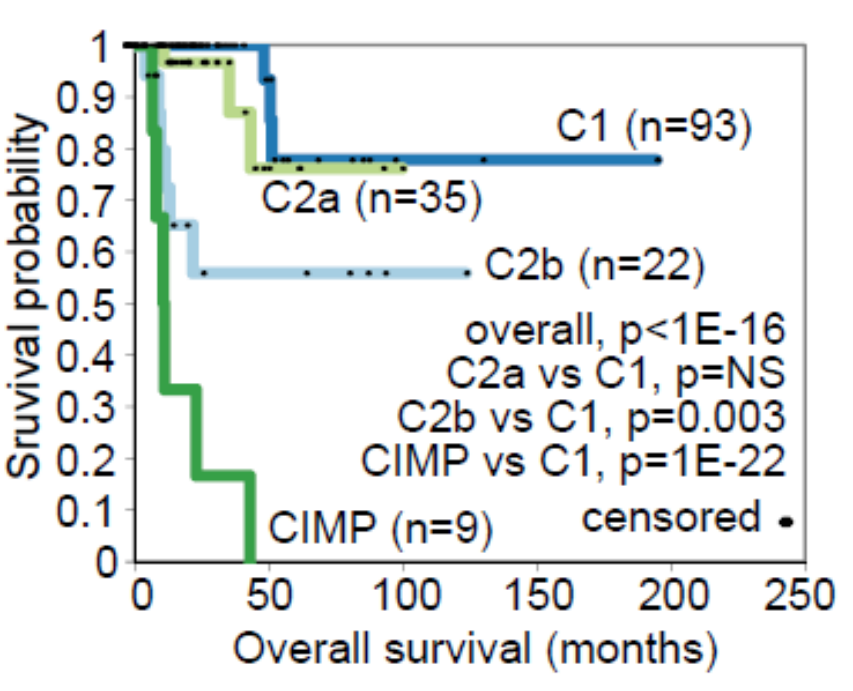


Cluster of Cluster Analysis (COCA)

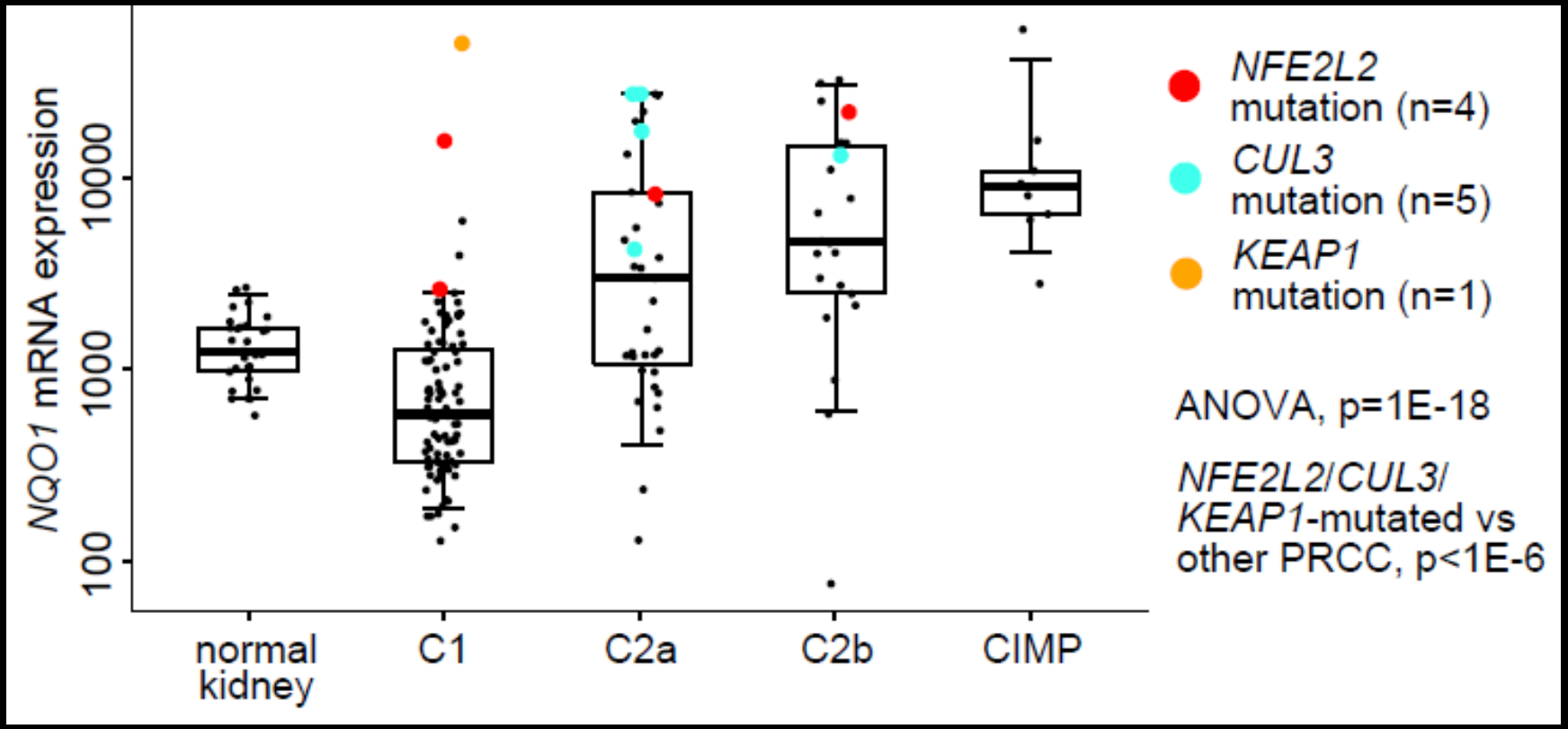


Cluster of Cluster Analysis (COCA)

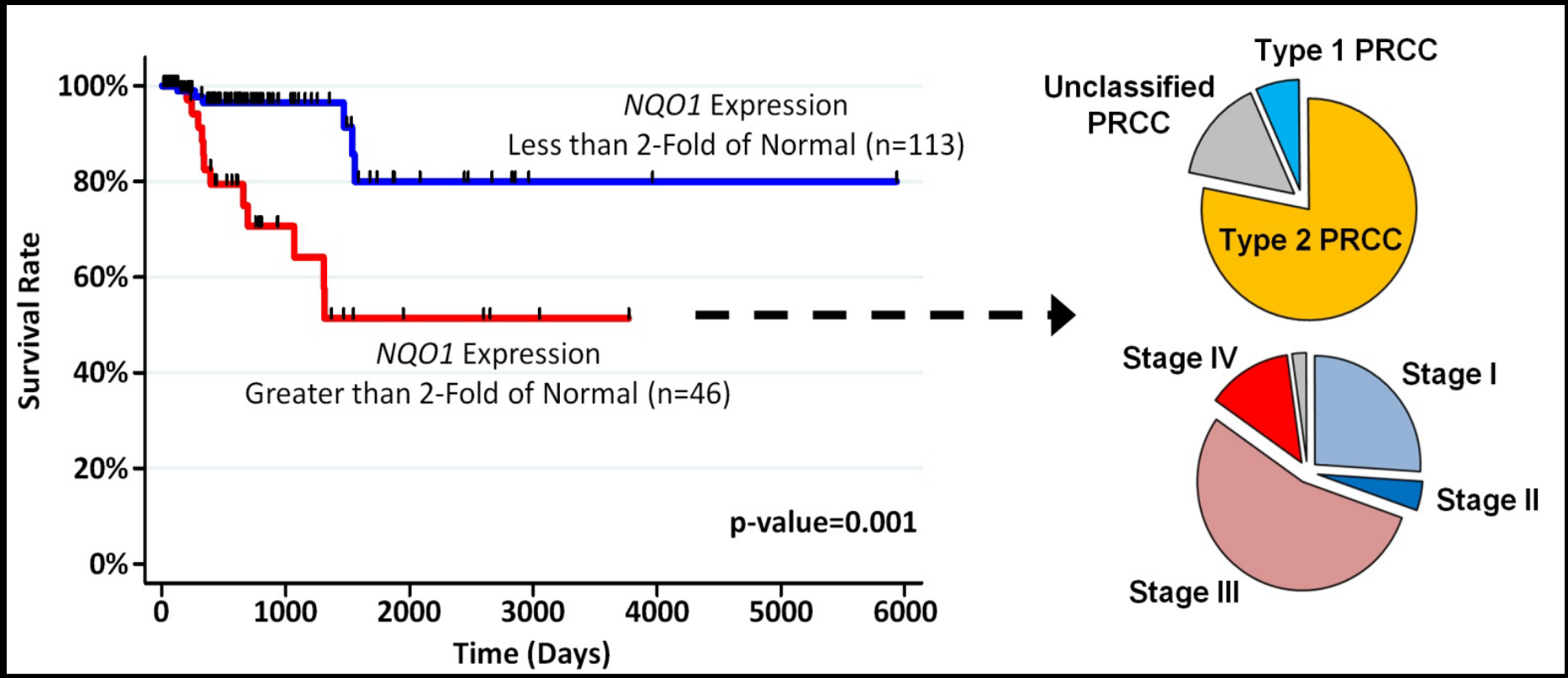




The NRF2 Pathway in Papillary Cancer



The NRF2 Pathway in Papillary Cancer



KIRP Conclusions

1. Type 1 PRCC and Type 2 PRCC are genomically distinctly different tumors with differing clinical outcomes.
2. Type 1 PRCC tumors are associated with *MET* mutations, *MET* splice variants and gain of chromosome 7.
3. Type 2 PRCC is made up of at least 3 distinct subtypes with differing survival.
4. *CDKN2A* alterations are associated with Type 2 PRCC and poor survival.

KIRP Conclusions

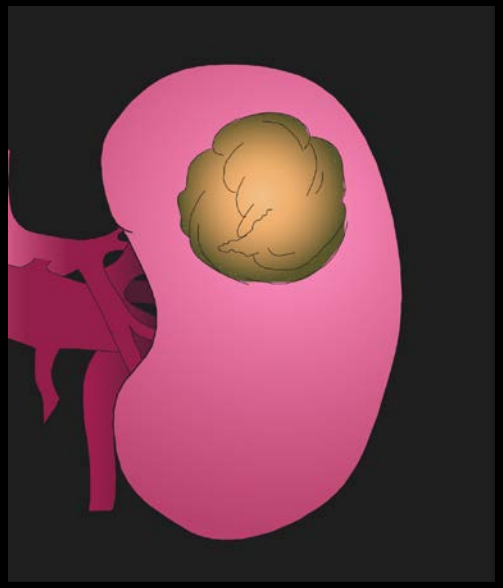
5. *TFE3* and *TFEB* gene fusions are found in 12% of Type 2 PRCC and can be found in older patients.

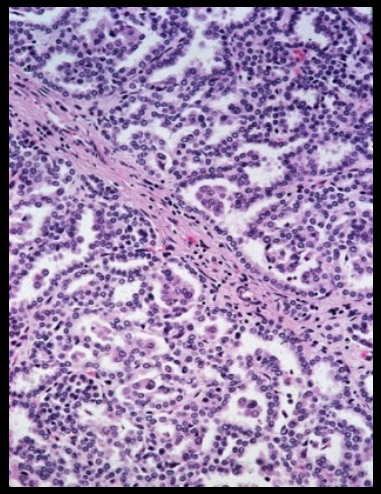
6. CIMP Type 2 PRCC tumors are early onset, poor survival tumors characterized by a metabolic shift to aerobic glycolysis and decreased oxidative phosphorylation.

7. The NRF2 pathway is up-regulated in Type 2 PRCC and is associated with high stage, low survival disease.

Comprehensive Molecular Characterization Papillary Renal Cell Carcinoma

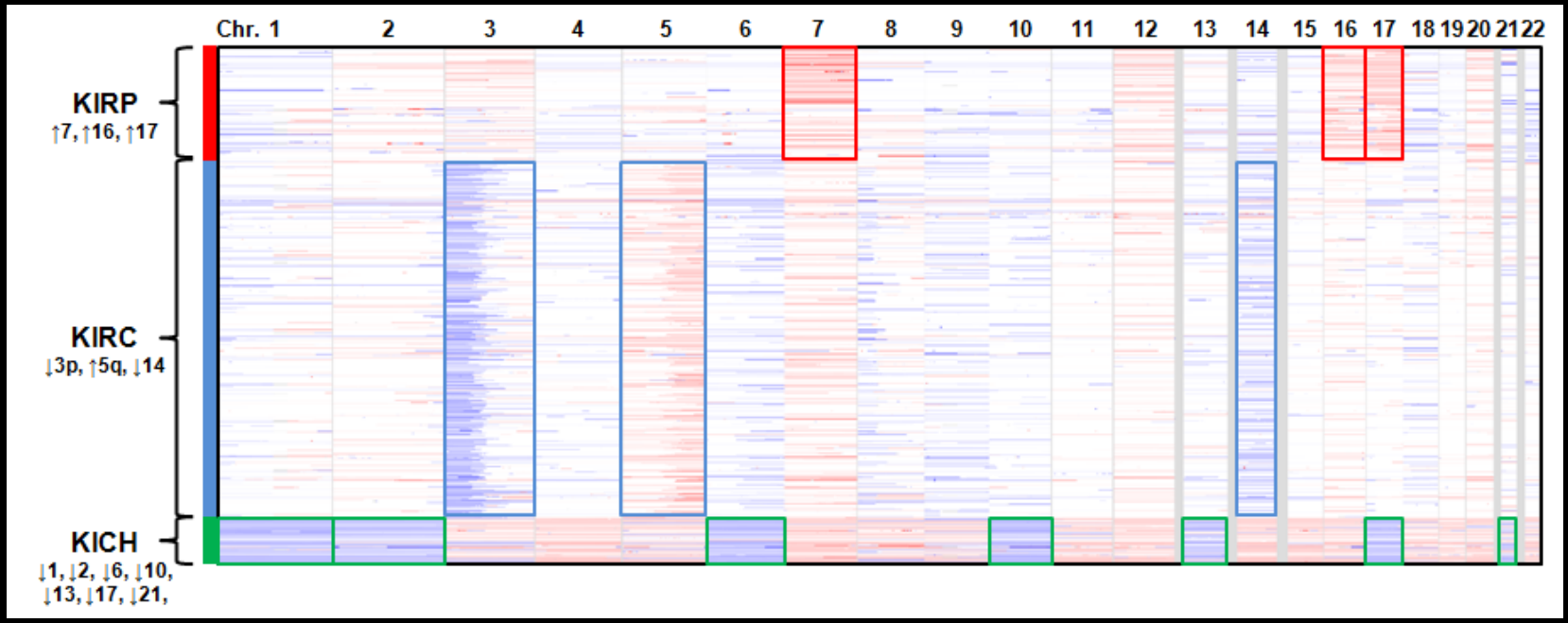
The Cancer Genome Atlas Research Network



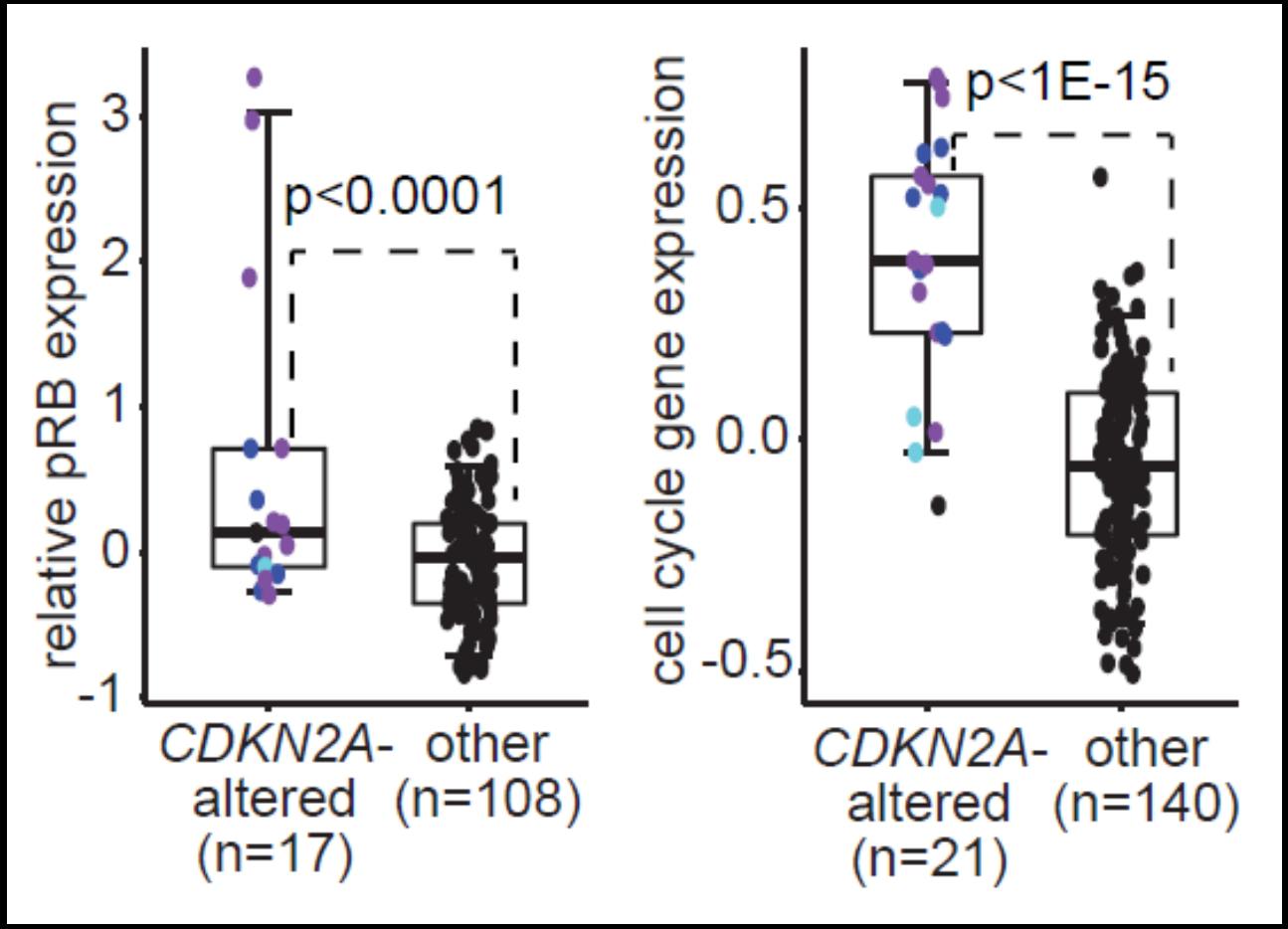


Comparative Copy Number Analysis

- KIRC: 3p loss
- KICH: multiple deletions
- KIRP: chromosome 7 increase



Type 2 PRCC Specific Alterations - *CDKN2A*



- Comparative analysis of tumors with & without *CDKN2A* alteration demonstrated significantly increased levels of pRB & cell cycle related genes.