

# Glioblastoma Multi-omics Phenotyping and Subtype Mechanism Study

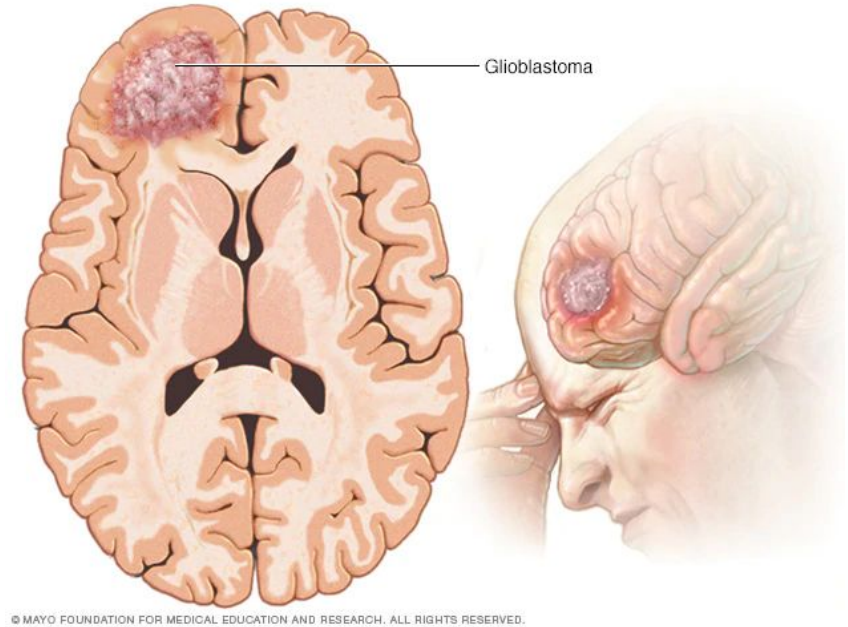
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# Introduction

**Glioblastoma:** aggressive cancer in the brain / spinal cord, originated from astrocytes that support nerve cells, difficult to treat

## Main treatment options:

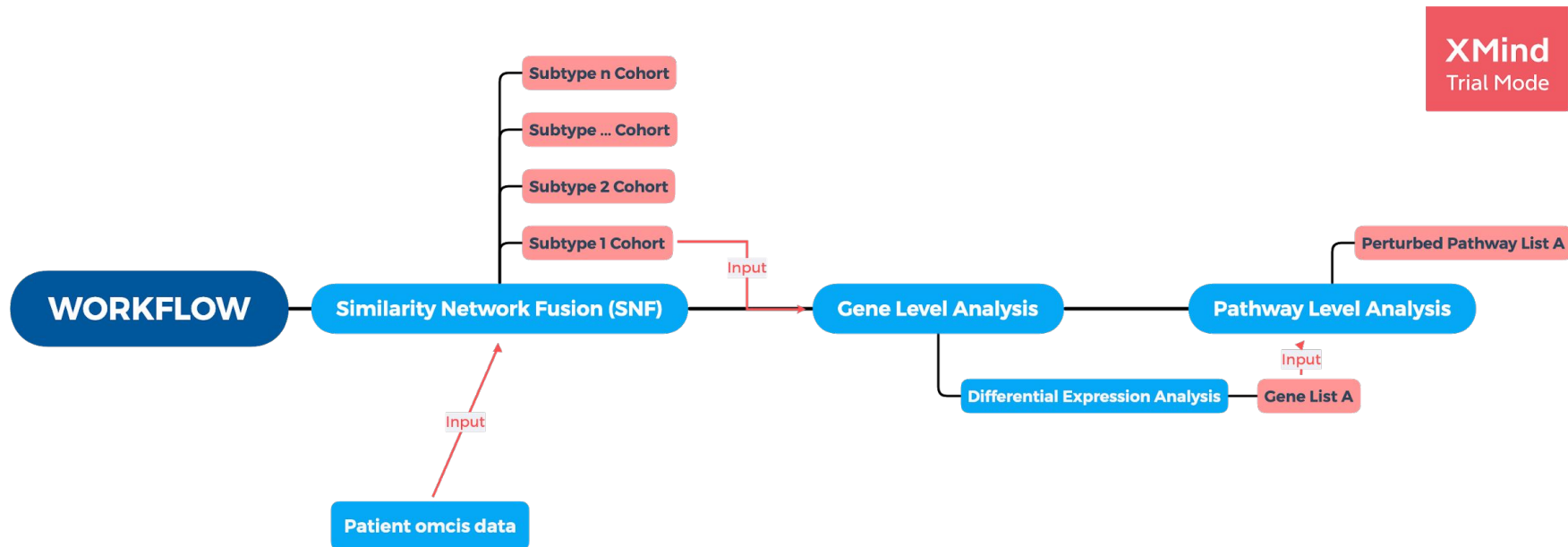
1. Surgery
2. Radiation therapy
3. Chemotherapy
4. Tumor treating fields (TTF) therapy
5. Targeted drug



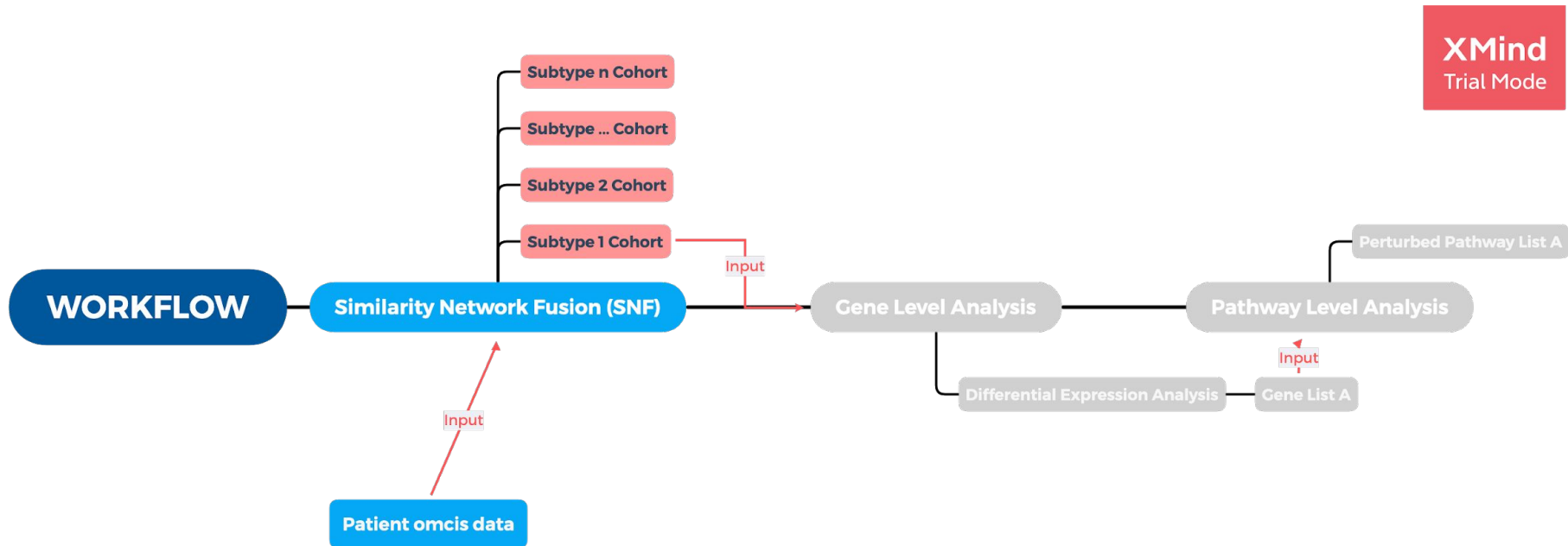
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<https://www.mayoclinic.org/diseases-conditions/glioblastoma/cdc-20350148>

# Workflow



# Multi-omics Integration and Subtyping

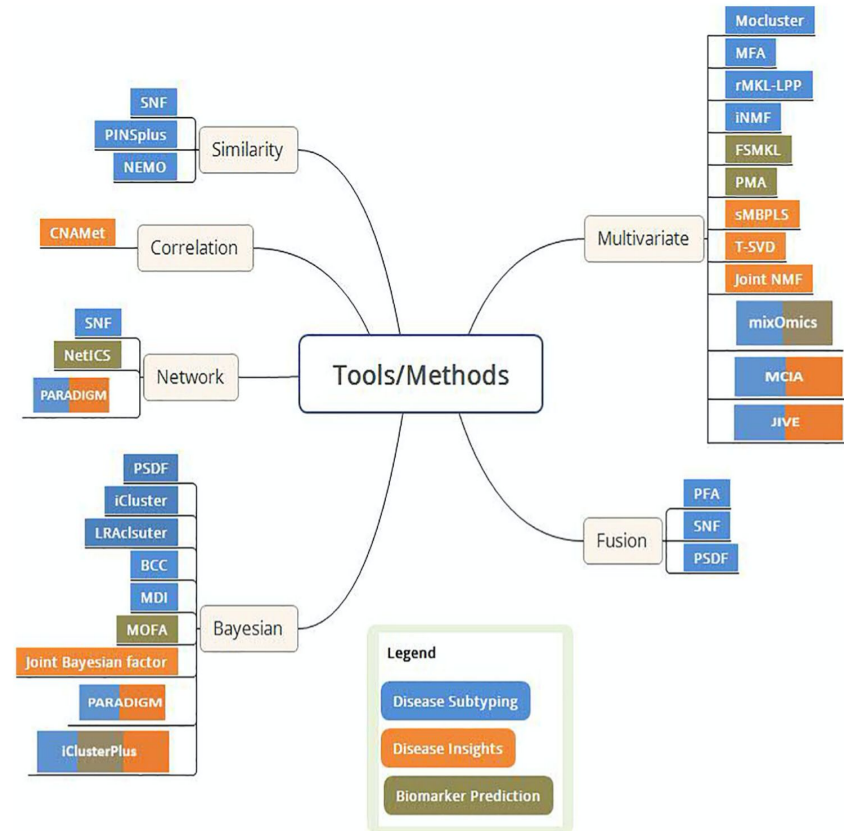


# Data & Preprocessing

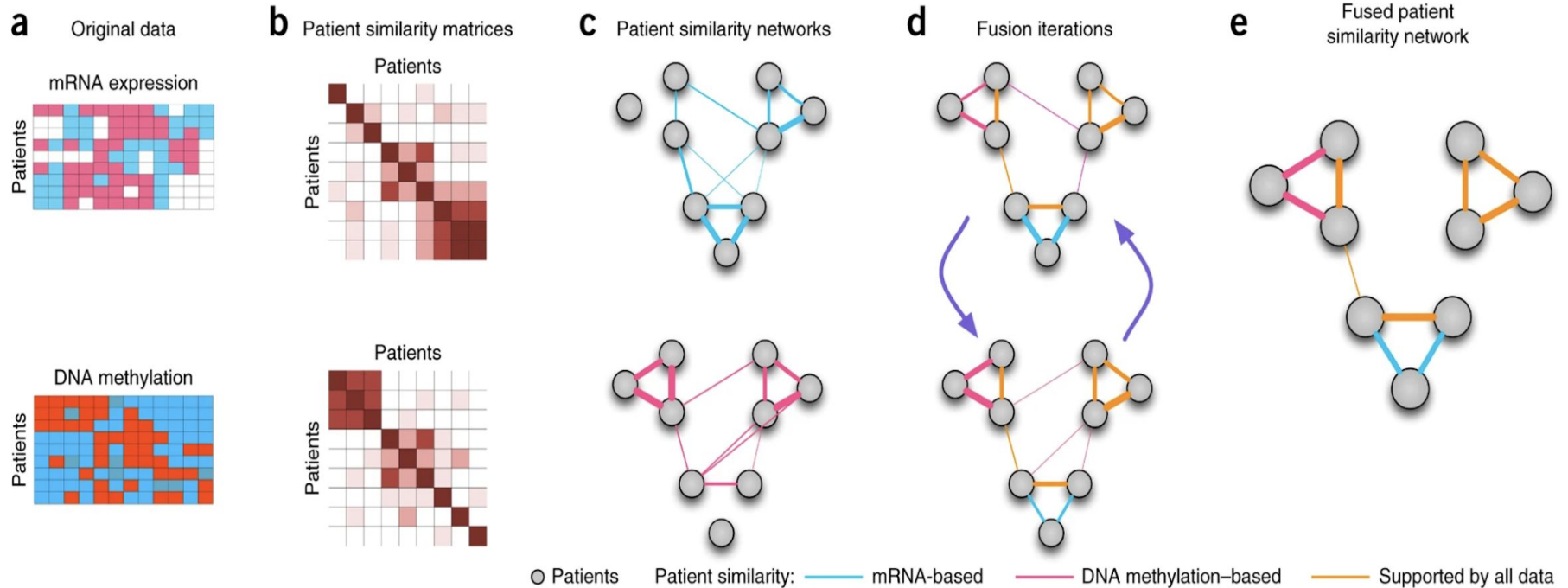
- Omics
  - Copy Number Variation
  - Methylation
  - Gene Expression
- Preprocessing
  - Intersection of omics
  - Missing data imputation (means)
  - 347 patients

# Multi-omics Integration

- Why multi-omics?
  - Data Availability
  - Understanding Interplay of molecules
- Which method?
  - Similarity Network Fusion (SNF)
  - PINSPlus



# Similarity Network Fusion (SNF)



# Subtyping -- Decide Cluster Numbers

- According to TCGA, 4 clusters
  - SNF results comparing with ground truth

```
[ (0, 0.3858267716535433),  
  (0, 0.2204724409448819),  
  (3, 0.6078431372549019),  
  (2, 0.5802469135802469) ]
```

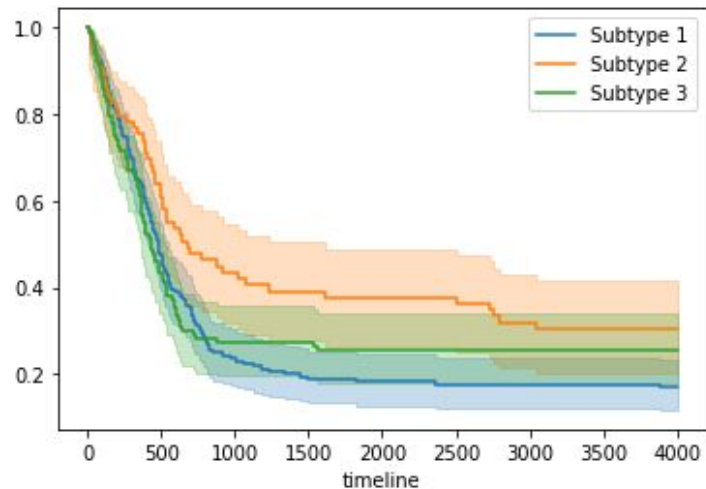
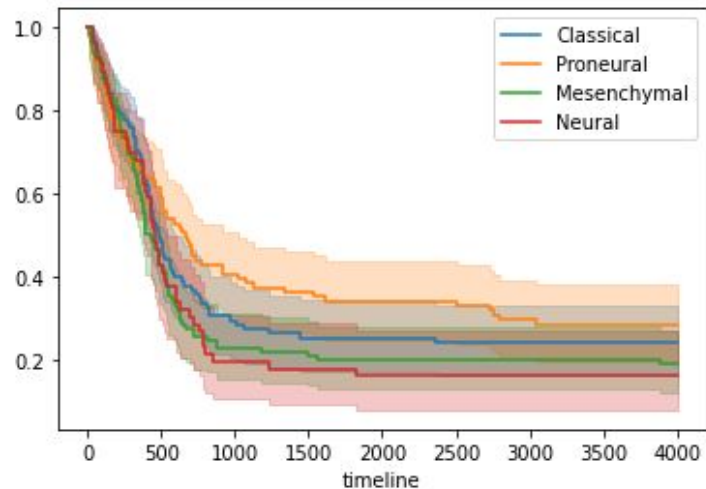
- Problem:
  - TCGA -- Consensus Clustering (hierarchical clustering)
  - Wang et. al revealed the existence of 3 subtypes
  - Clinical significance



# Survival Analysis

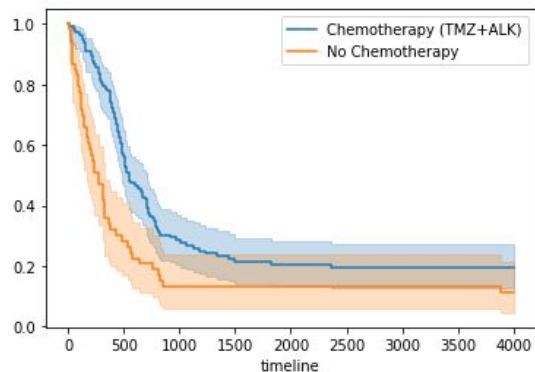
Comparison	Logrank test statistic	p-value
Classical - Proneural	0.791758	0.373569
Classical - Mesenchymal	1.321919	0.250248
Classical - Neural	1.147819	0.284006
Proneural - Mesenchymal	3.878100	0.048920
Proneural - Neural	3.738071	0.053186
Mesenchymal - Neural	0.004247	0.948038

Comparison	Logrank test statistic	p-value
Group 1 - Group 2	6.206181	0.012730
Group 2 - Group 3	2.637399	0.104374
Group 3 - Group 3	0.242386	0.622487

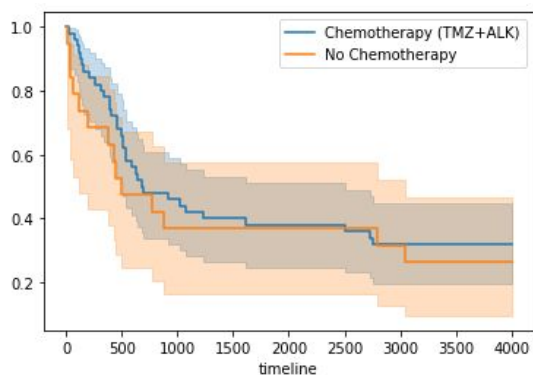


# Response to chemotherapy

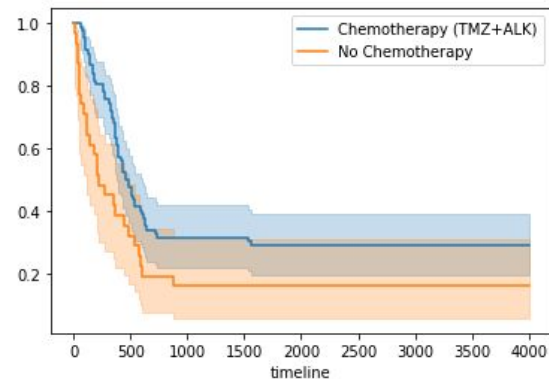
Subtype 1



Subtype 2



Subtype 3



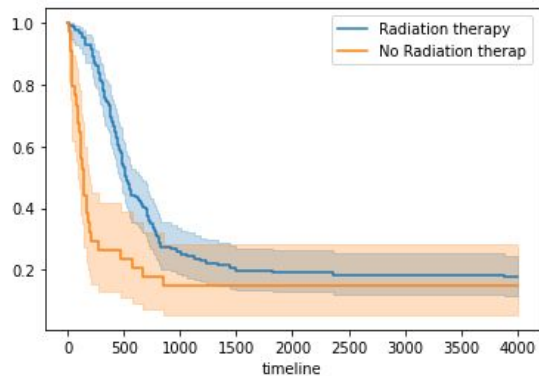
Logrank	p	-log2(p)
13.03	<0.005	11.67

Logrank	p	-log2(p)
0.54	0.46	1.11

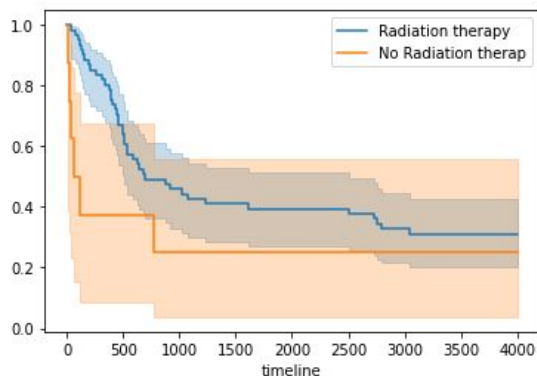
Logrank	p	-log2(p)
5.81	0.02	5.97

# Response to radiation therapy

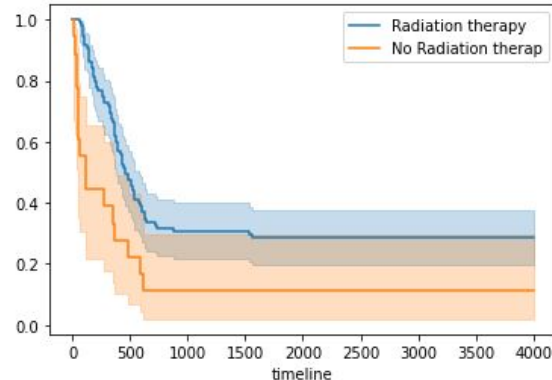
Subtype 1



Subtype 2



Subtype 3



Logrank	p	-log2(p)
12.82	<0.005	11.51

Logrank	p	-log2(p)
2.10	0.15	2.76

Logrank	p	-log2(p)
9.95	<0.005	9.28

# Clinical Feature Analysis

	CDE_DxAge	CDE_survival_time	days_to_birth	CDE_chemo_alk_days
Subtype 1	61.37	426.38	61.57	148.987
Subtype 2	52.54	633.61	53.64	143.187
Subtype 3	58.39	313.6	58.77	82.01

Anova  
Test

# Clinical Feature Analysis Summary

- Subtype 2 contains younger patients with favorable prognosis

# Clinical Feature Analysis

	NON G-CIMP	G-CIMP
G1	161	4
G2	42	24
G3	108	5

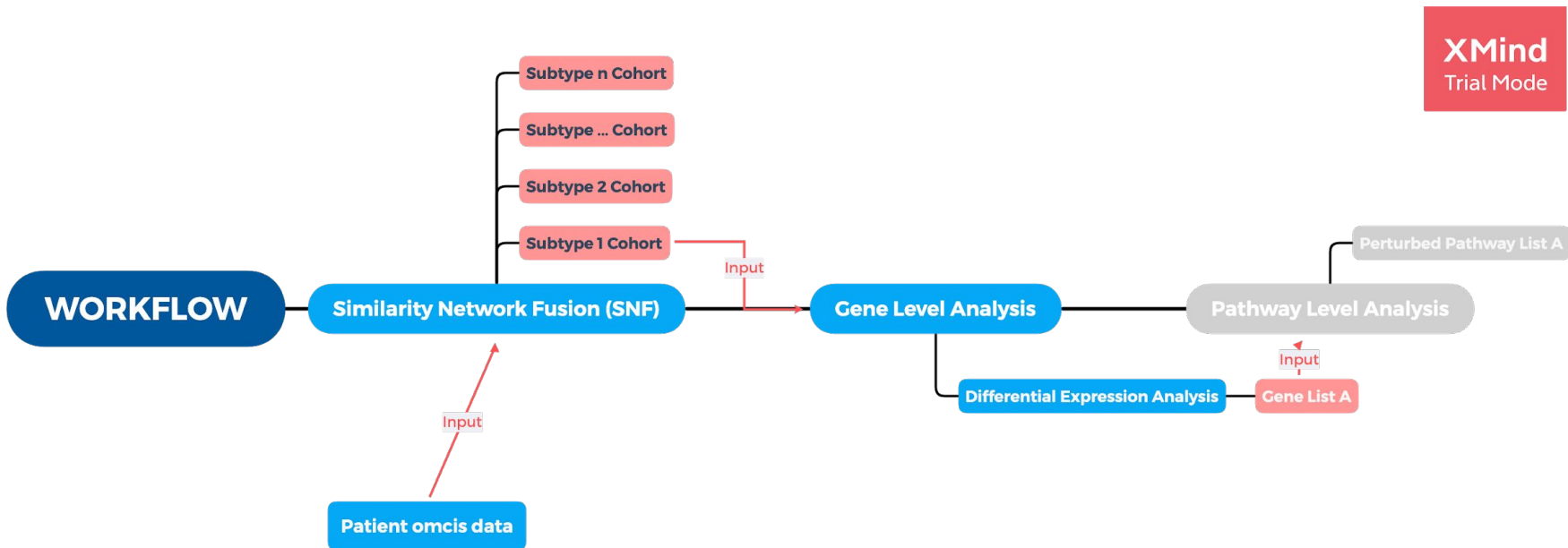
Chi-square Test

```
[('CDE_therapy', 0.0239712693970924),  
 ('CDE_vital_status', 0.04219955419397384),  
 ('G_CIMP_STATUS', 1.897978420038419e-15),  
 ('GeneExp_Subtype', 8.534673419184012e-22),  
 ('_PANCAN_CNA_PANCAN_K8', 2.1989441476890682e-14),  
 ('_PANCAN_DNAMethyl_GBM', 9.889506786244145e-30),  
 ('_PANCAN_DNAMethyl_PANCAN', 2.253317472999156e-15),  
 ('_PANCAN_mutation_PANCAN', 0.002746407829379185),  
 ('form_completion_date', 0.04856771110301229),  
 ('hormonal_therapy', 0.004905154740707577),  
 ('initial_pathologic_diagnosis_method', 0.04956822543700062),  
 ('tissue_source_site', 7.59186908651961e-05),  
 ('vital_status', 0.023497201679813208),
```

# Clinical Feature Analysis Summary

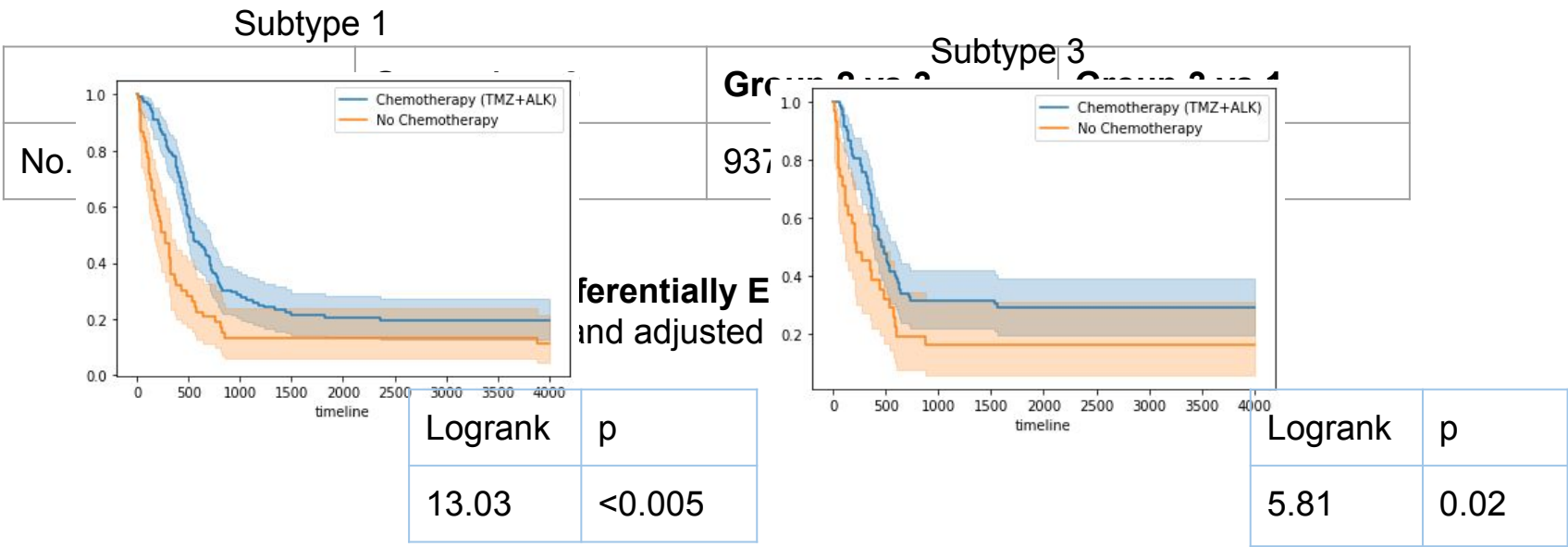
- Subtype 2 contains younger patients with favorable prognosis
- Baysan, et al also revealed a subset of GBM, G-CIMP tumors
- Lv, Shasha, et al revealed IDH1 subtype
  - [0.9030303030303031, 0.9130434782608695, 0.8938053097345132]

# Differential Expression Analysis





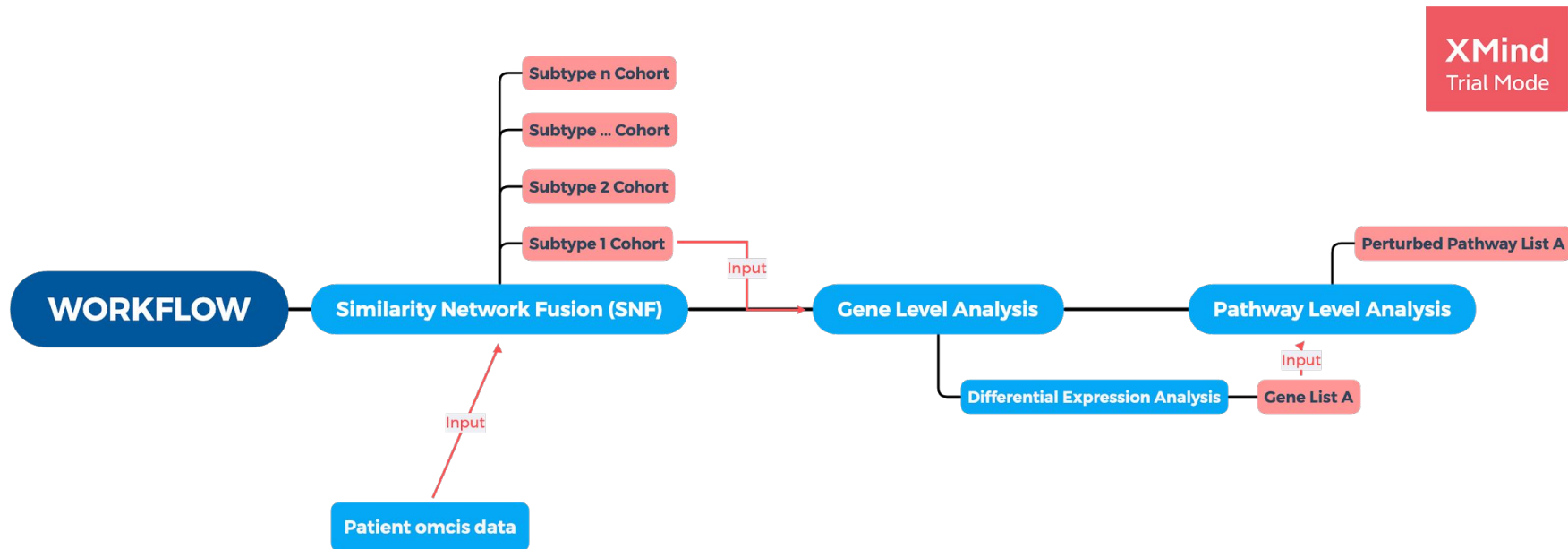
# Differential Expression Analysis



MGMT: 3 > 1      Promoter unmethylation and high expression levels of *O*6-methylguanine DNA methyltransferase (MGMT) have been recognized as indicators of the TMZ-resistant phenotype,

CTSD: 3 > 1      significant association with Cathepsin D (CTSD) overexpression, which is reported to prevent the effect of TMZ.

# Gene Ontology/Pathway Enrichment Analysis

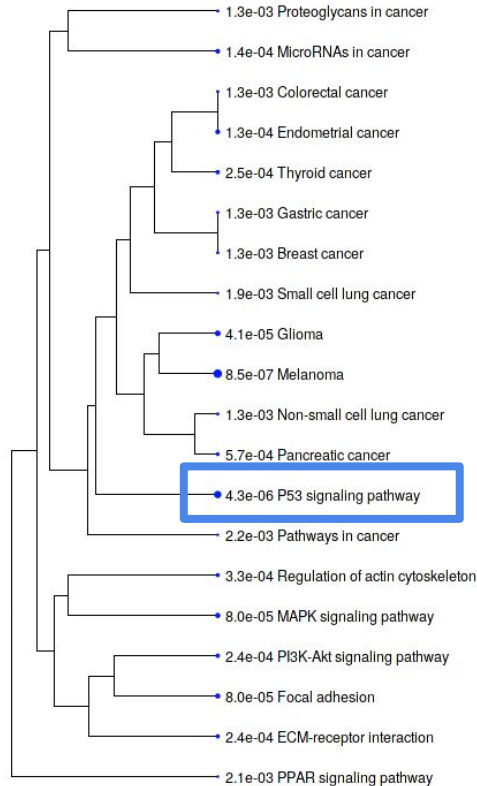


ShinyGO:  
<http://bioinformatics.sdstate.edu/go/>

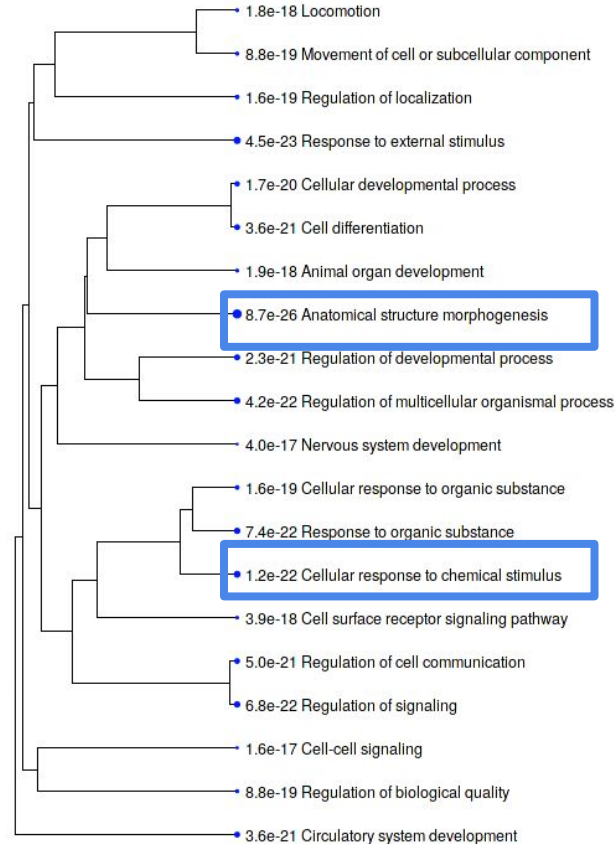
# Pathway Enrichment Analysis

Subtype 1 vs Subtype 2

## KEGG pathways



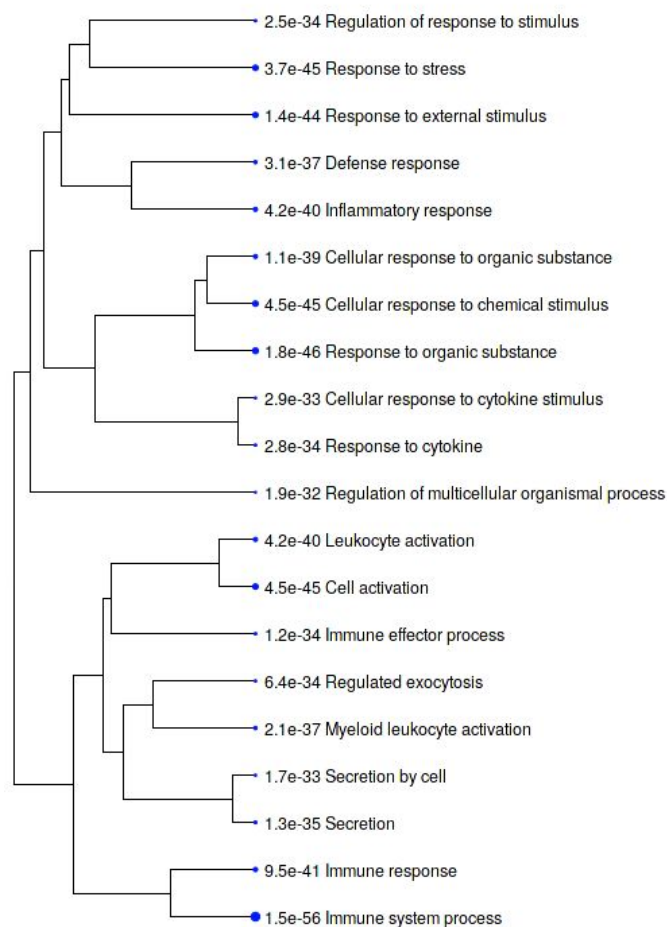
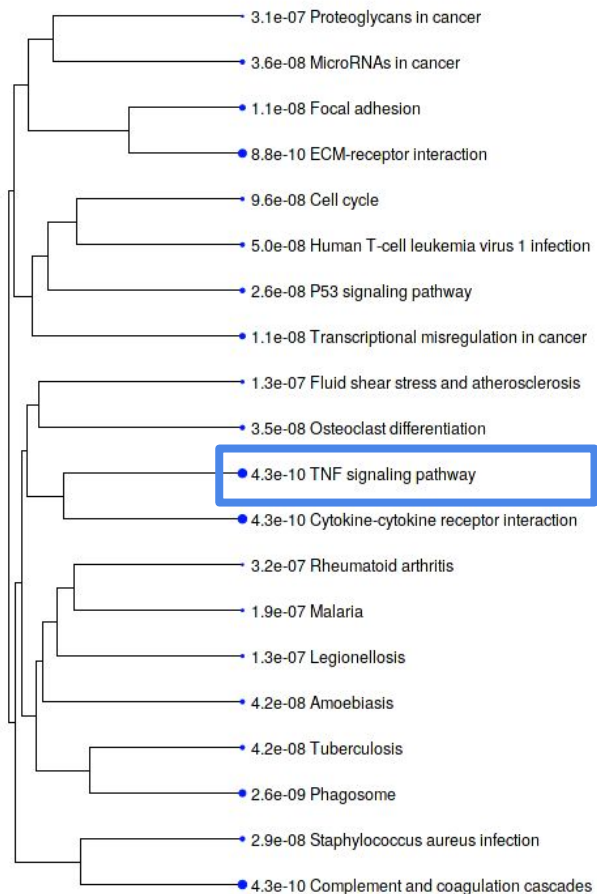
## GO Biological Processes



- **anatomical structure morphogenesis:** found to be enriched in the genes associated with overall glioblastoma survival (Serão, Nicola VL, et al, 2011)
- **P53 signaling pathway:** expression of p53 mutant correlates with worse prognosis (Zhang, Ying, et al., 2018)

# Pathway Enrichment Analysis

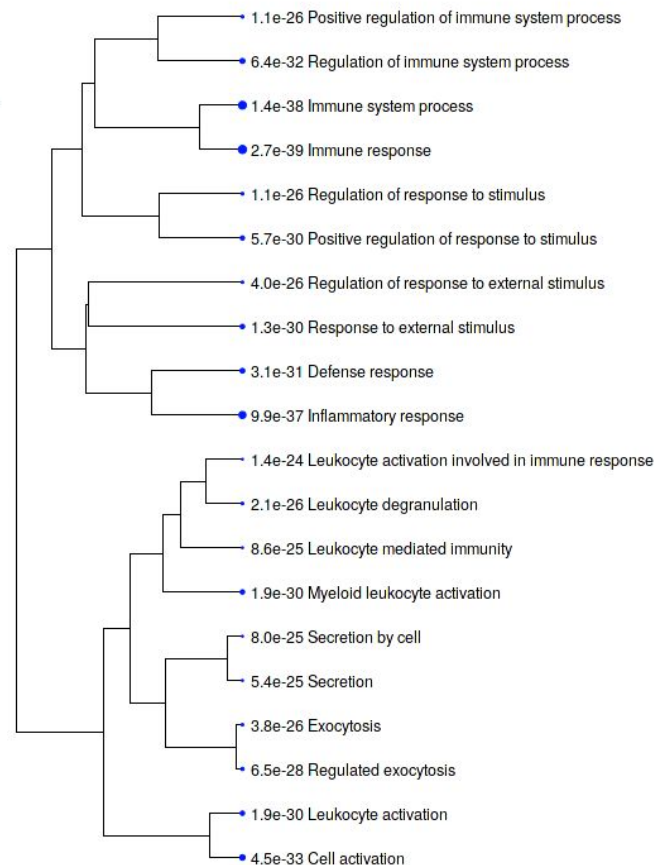
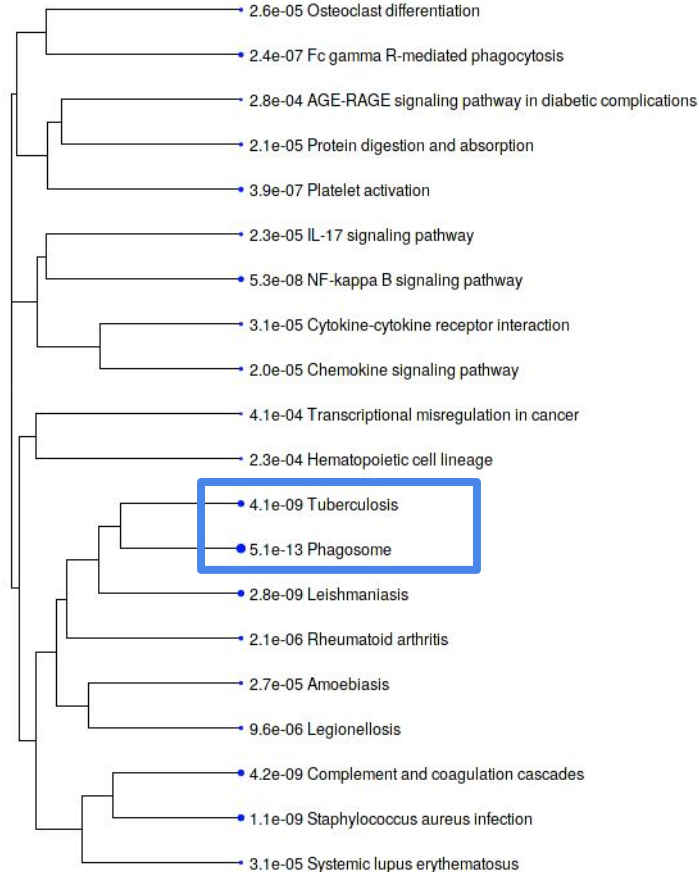
Subtype 2 vs Subtype 3



- **TNF signaling pathway:**  
TNF receptor TNFR2 found to be predictive of GBM prognosis (Zhang, Wenqing, et al. , 2017)

# Pathway Enrichment Analysis

Subtype 3 vs Subtype 1



- **Phagosome:** formed by apoptotic cells, target of chemotherapy in GBM (Trejo-Solís, Cristina, et al, 2018)
- **Tuberculosis:** possibly related to TMZ therapy (de Paiva, Tadeu Ferreira, et al, 2009)

# Future Directions

- To investigate response to other types of therapy, eg. hormone therapy, targeted drugs
- To further investigate the pathway results to reveal more detailed biological mechanisms
- To study the mechanisms of subtypes using methylation and copy number variants data

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