Melanoma Cancer Progression Model:

using Capri

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Overview :

- Summary of Cancer Evolution
- Why it is important
- Overview of cancer progression models
- Melanoma progression model using CAPRI

Cancer Evolution:

Goal: To identify specific mutations or mutational patterns that drive cancer progression

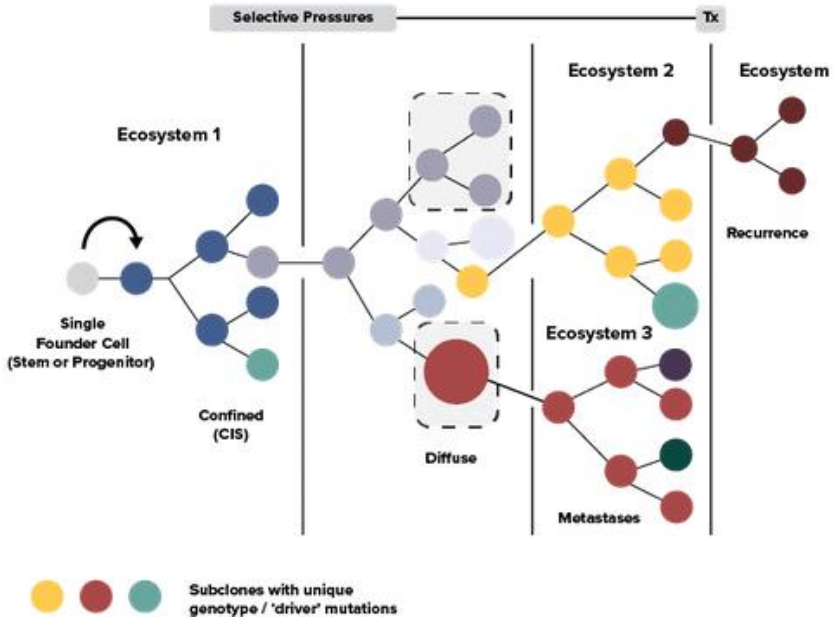


Image: Greaves & Maley, 2012. Clonal evolution in cancer. *Nature*

- Cancer development and progression closely follows a traditional clonal Darwinian evolution
- Most Cancers progress through the accumulation of alterations that affect gene functions
- This is typically driven by step-wise somatic mutations with sequential sub-clonal selections (Nowell, 1976)
- Driver mutations give cells the selective advantages that lead to further tumor progression

Cancer Evolution: Alterations

Passenger Mutations: Neutral mutations, little to no effect

Driver Mutations: Leads to tumor cell proliferation and disease progression

- Activation/deactivation of proteins
- Deregulation of cellular processes
- Leading to intra-tumor heterogeneity

Cancer Evolution: Intra-Tumor Heterogeneity

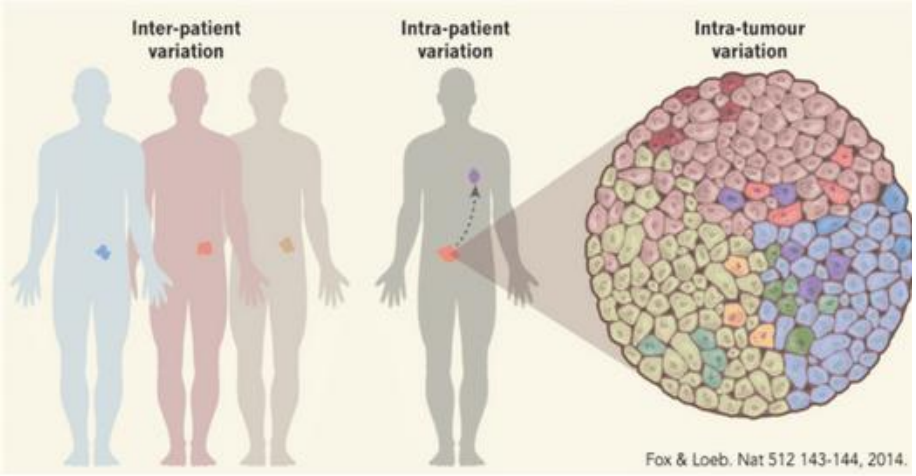
Intra-tumor heterogeneity plays an important role in how cancer develops and evolves

As time passes clones and sub-clones within a tumor continue to grow and mutate causing a variation of different cells within a tumor

Intra-tumor heterogeneity is seen in -

- Cellular morphology
- Genetic variation
- Gene expression

Knowing the heterogeneity of a tumor can serve as a clinically valid molecular marker (Gerashchenko, et al., 2013)



Cancer Evolution: Why it's important

Knowing the evolution and progression of cancer is critical in drug development and treatment decisions:

- Patients at different stages and with different mutational evolutions will respond differently to various treatments
- Treatments that target dominant tumor clones will cause a shift in selective pressures, allowing certain minor sub-clones to multiply which can result in disease relapse.

Cancer Evolution: Progression Models

Predicting the path of cancer progression can be critical in diagnostic, prognostic, and treatment options.

There are many progression models available that use a variety of methods; including – Conjunctive Bayesian networks, DAGS, Phylogenetic tree models, etc.

Goals:

- Identifying driver alterations and inferring progression
- Determine the mutational landscape of the tumor genome
- Determine differences at various phases of disease, i.e. recurrence/ metastasis
- Evaluate impact of therapies/ treatments

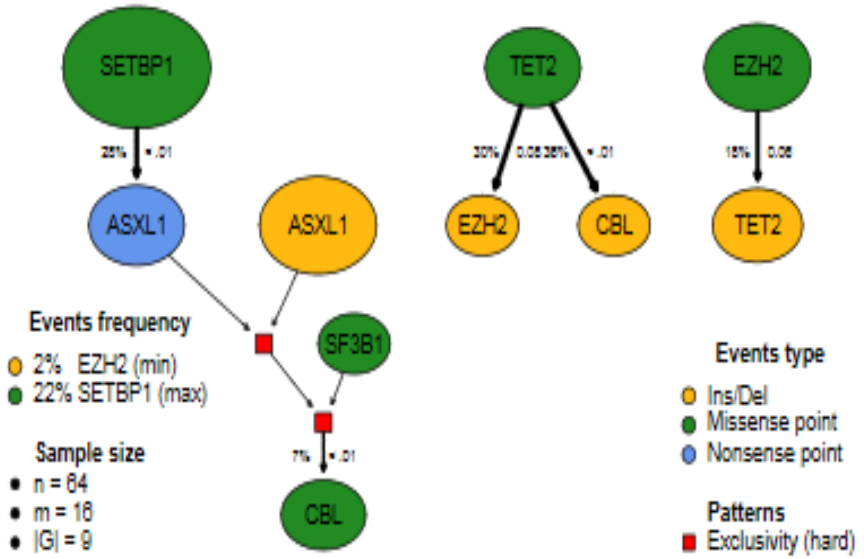
Examples –

- **CAPRI** (Cancer Progression Inference) – DAG producing model – Ensemble data
- **CAPRESE** (Cancer Progression Extraction w/ Single Edges) – Bulk data - Tree producing model
- **TRAIT** (Temporal Order of Individual Tumors) – Bulk data - Tree producing model
- **LACE** (Longitudinal Analysis of Cancer Evolution) – Single cell - tree producing model

CANCER EVOLUTION: **CAPRI** (CANCER PROGRESSION INFERENCE)

Based on Suppes' Theory of causality – uses selective advantage relationship between genetic events to reconstruct a set of prima facie 'causal' relationships

- **The CAPRI algorithm**:
  - **Step 1**: Data input (MAF or Gistic file)
  - **Step 2**: Preprocessing – produce an m x n Boolean matrix with an optional set of hypotheses
  - **Step 3 & 4**: DAG node and edge selection
  - **Step 5**: DAG labeling
  - **Step 6**: Likelihood fit – to reduce spurious hits
- (Ramazzotti, et al., 2015)



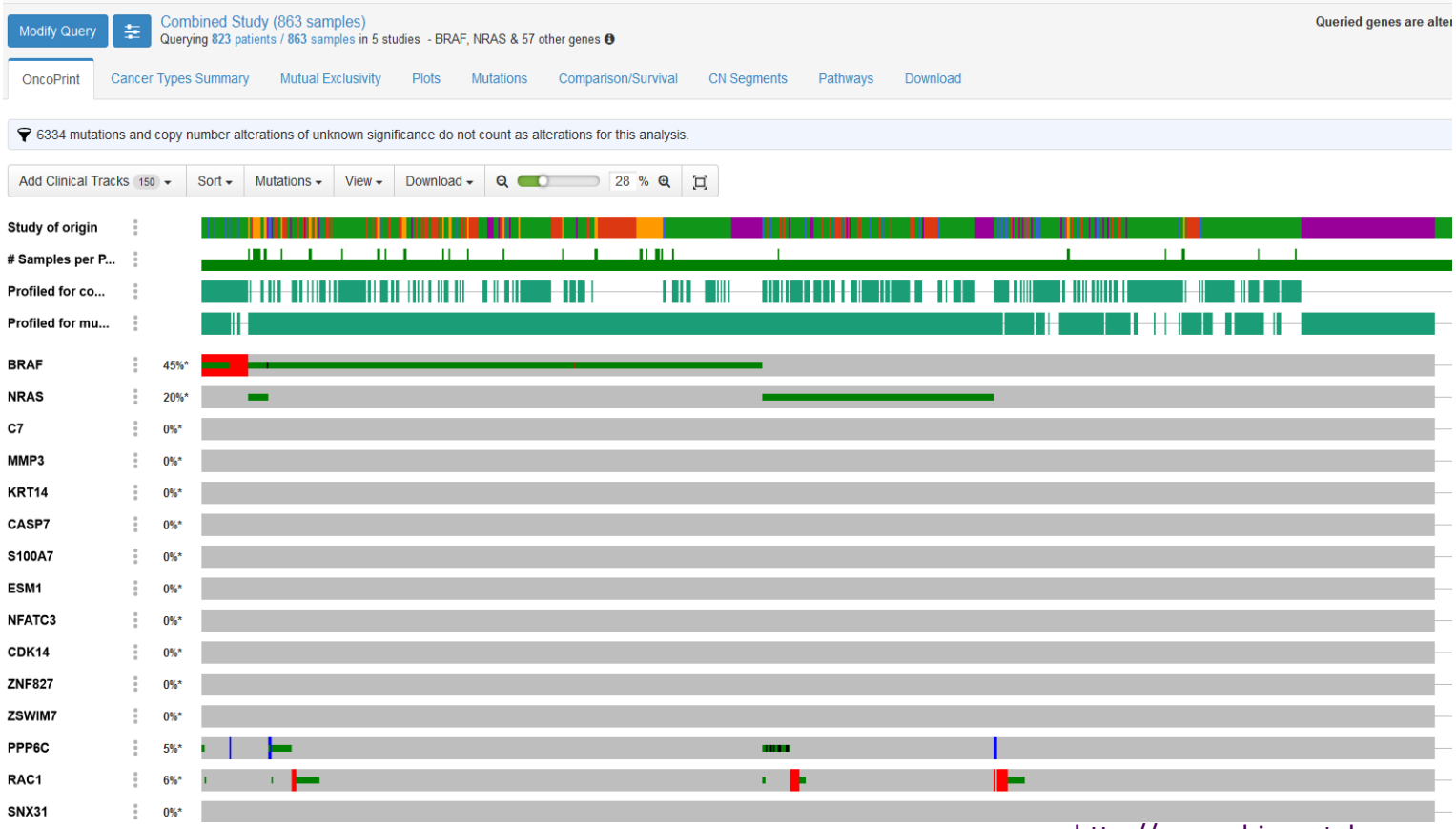
Cancer Evolution: Melanoma

Why investigate Progression of Melanoma -

- Melanoma incidences are increasing - with about 100,350 new cases estimated to be diagnosed in 2020 (American Cancer Society)
- While there have been many recent discoveries, events that drive tumor progression in melanoma are still largely unknown.
- Melanoma often has a low sensitivity to chemotherapy with many patients unable to achieve remission

Cancer Evolution: CAPRI Melanoma Data

Input Data - Clinical cutaneous melanoma cross-sectional patient data from TCGA. Obtained directly from the Genomic Data Commons portal (https://portal.gdc.cancer.gov/)



http://www.cbioportal.org

Cancer Evolution: CAPRI Melanoma Data

Event Selection: Literature review and Cbioportal analysis -

- Most models show that founder ***BRAF*** or ***NRAS*** mutations are often first detected in precursor lesions that give rise to melanoma by accumulation of additional mutations(Shain et al., 2015).
- Mutations noted in literature review and from cbioportal: *BRAF', 'NRAS', 'PPP6C', 'RAC1', 'PTEN', 'GRIN2A', 'KMT2C', 'TP53', 'TP63', 'NF1', 'KDR', 'CDKN2A', 'AGO2', 'KMT2A', 'MUC16', 'MUC15', 'RASA2', 'RAC1', 'ARID2', 'TERT', 'PI3K', 'KIT', 'MET', 'MAP2K1'*

Cancer Evolution: CAPRI Melanoma Data

Step 1: Input Data

```
library(TCGAbiolinks)
library(TRONCO)

# Import MAF data set
maf <- GDCquery_Maf("SKCM", pipelines = "mutect2")
```

Step 2: Refine the data

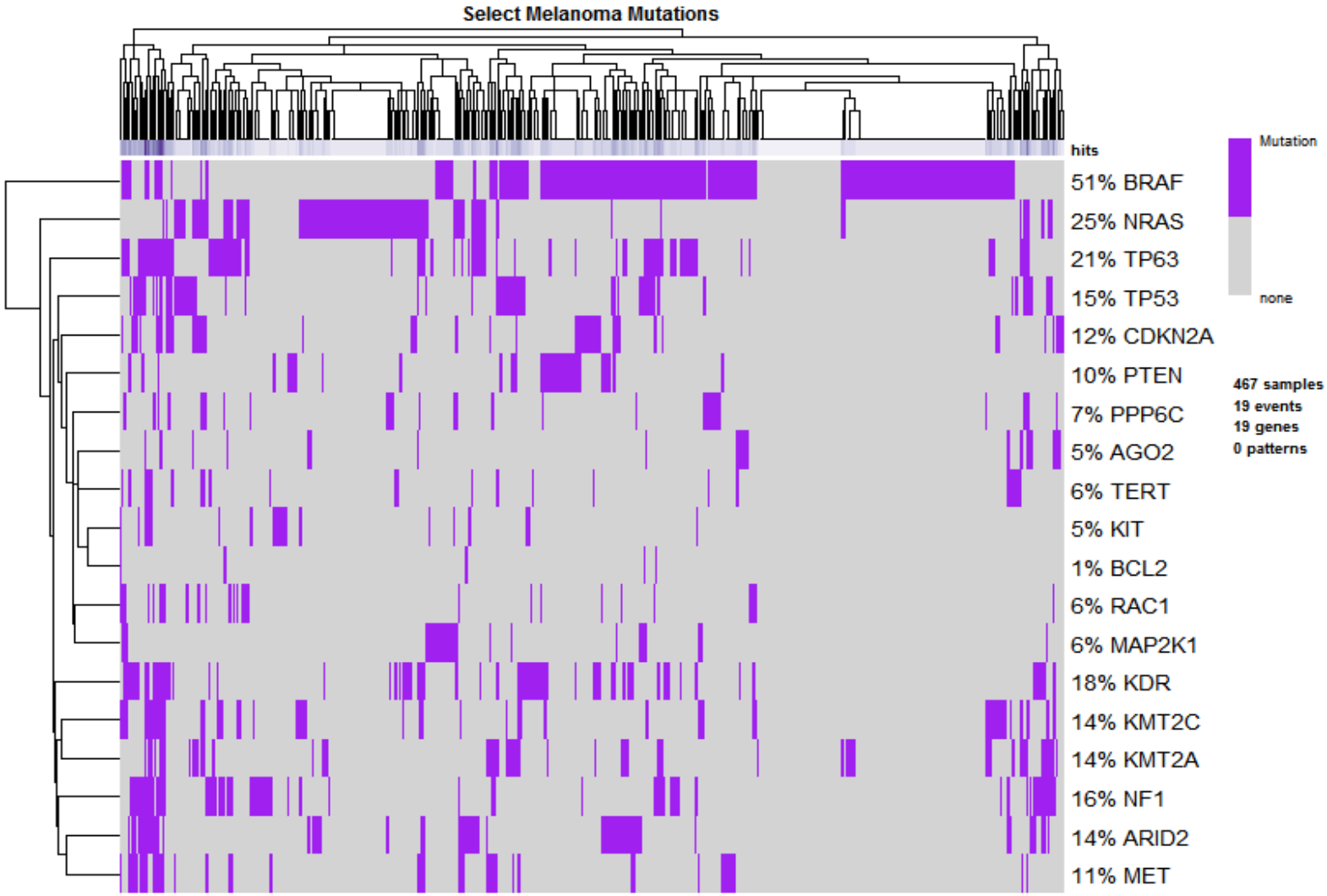
```
## import the maf file into the TRONCO package
maf<- as.data.frame(maf)
maf_data = import.MAF(maf, merge.mutation.types = T)  # to seperate mutations based on type set to FALSE

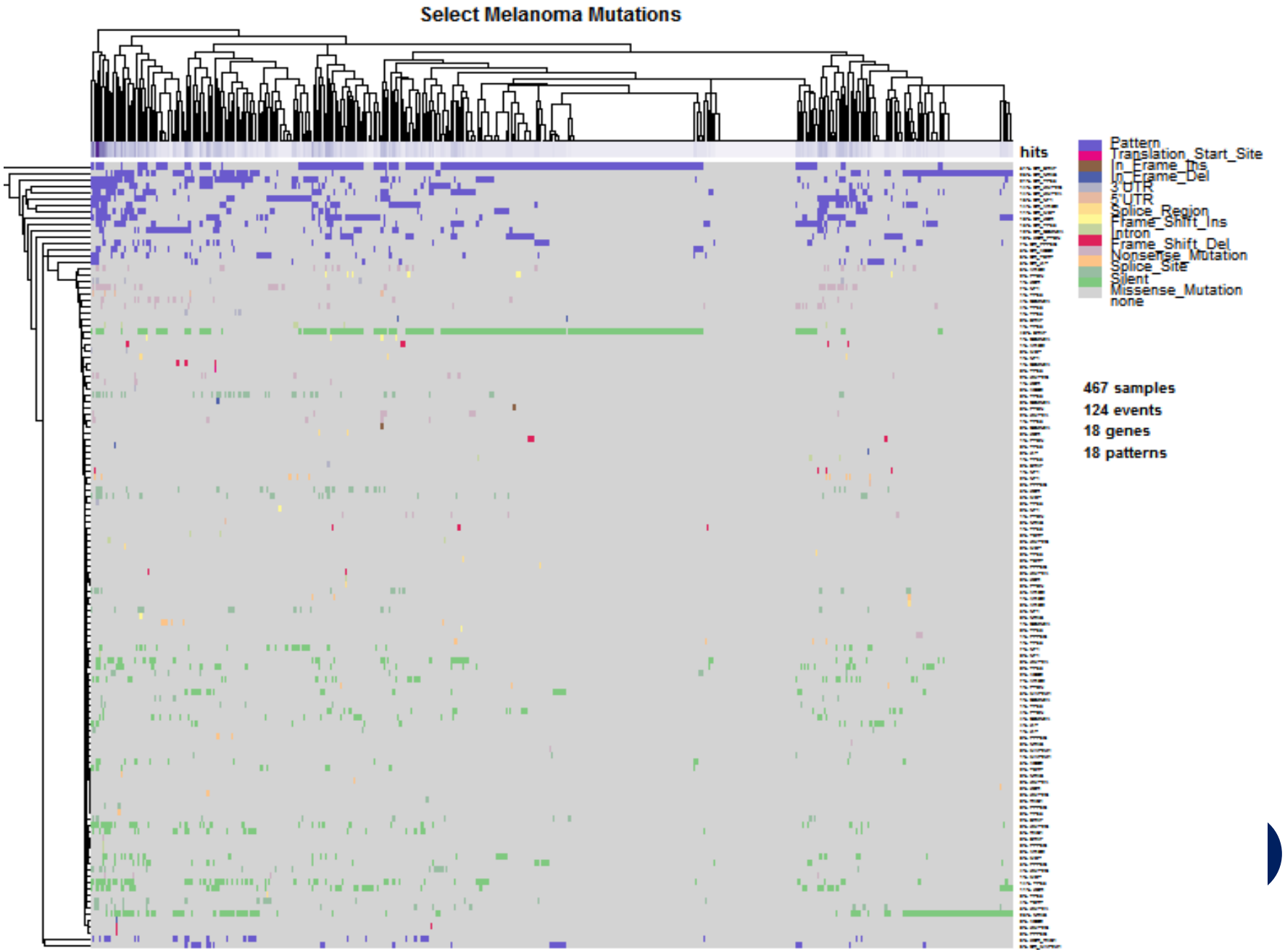
# filter the data to select events based on literature review/ cbioportal etc.
drivers<- c('BRAF', 'NRAS', 'PPP6C', 'RAC1', 'PTEN', 'KMT2C', 'TP53','TP63', 'NF1', 'KDR', 'CDKN2A','AGO2', 'KMT2
A', 'RAC1', 'ARID2', 'TERT', 'PI3K', 'KIT', 'MET', 'MAP2K1')

# exclusivity events
gene_hypothesis<- hypothesis.add(gene_hypothesis, 'NRAS xor BRAF', XOR('NRAS', 'BRAF'))

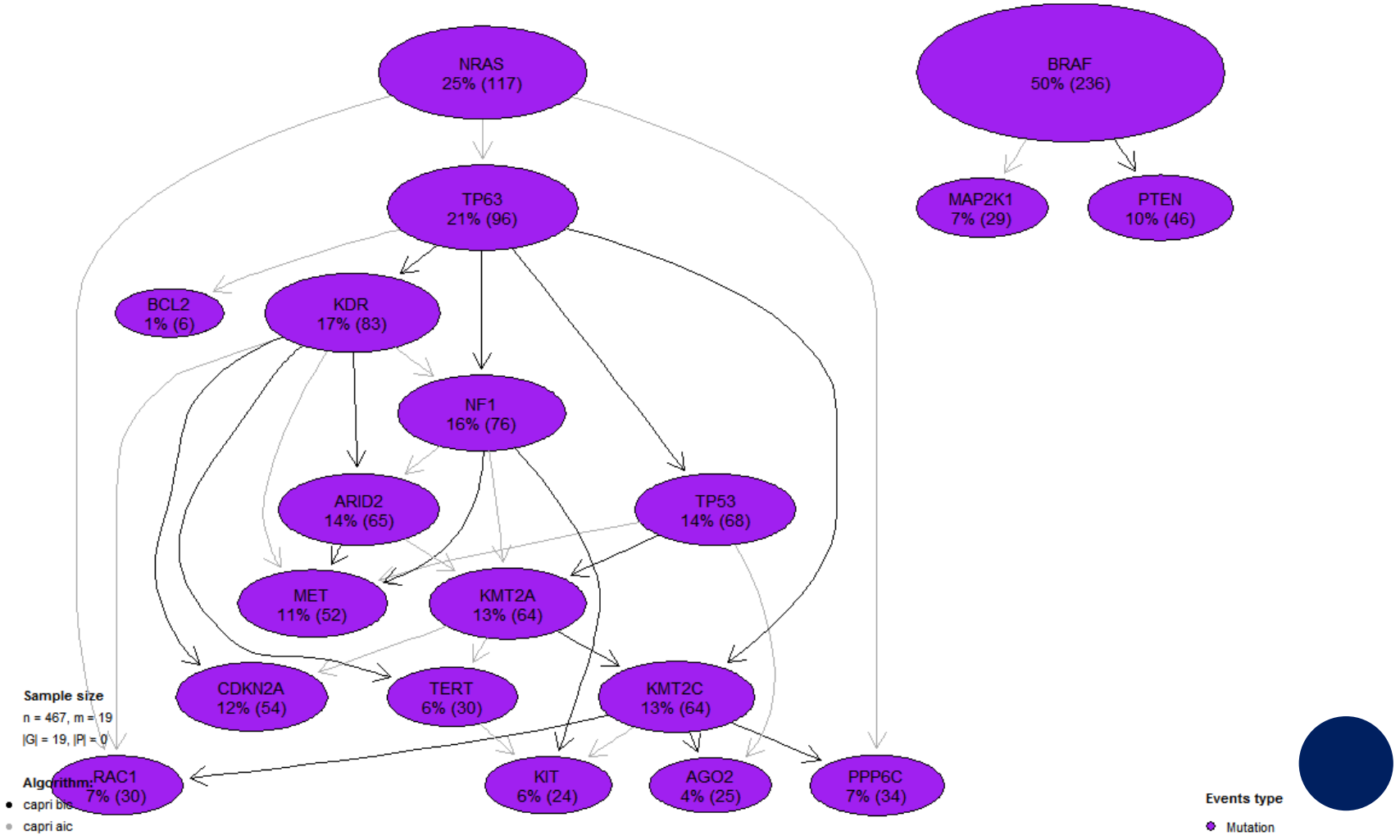
# select the events to run
skcm_dataset<- events.selection(maf_data, filter.in.names= drivers)
```

Cancer Evolution: CAPRI - Oncoprint



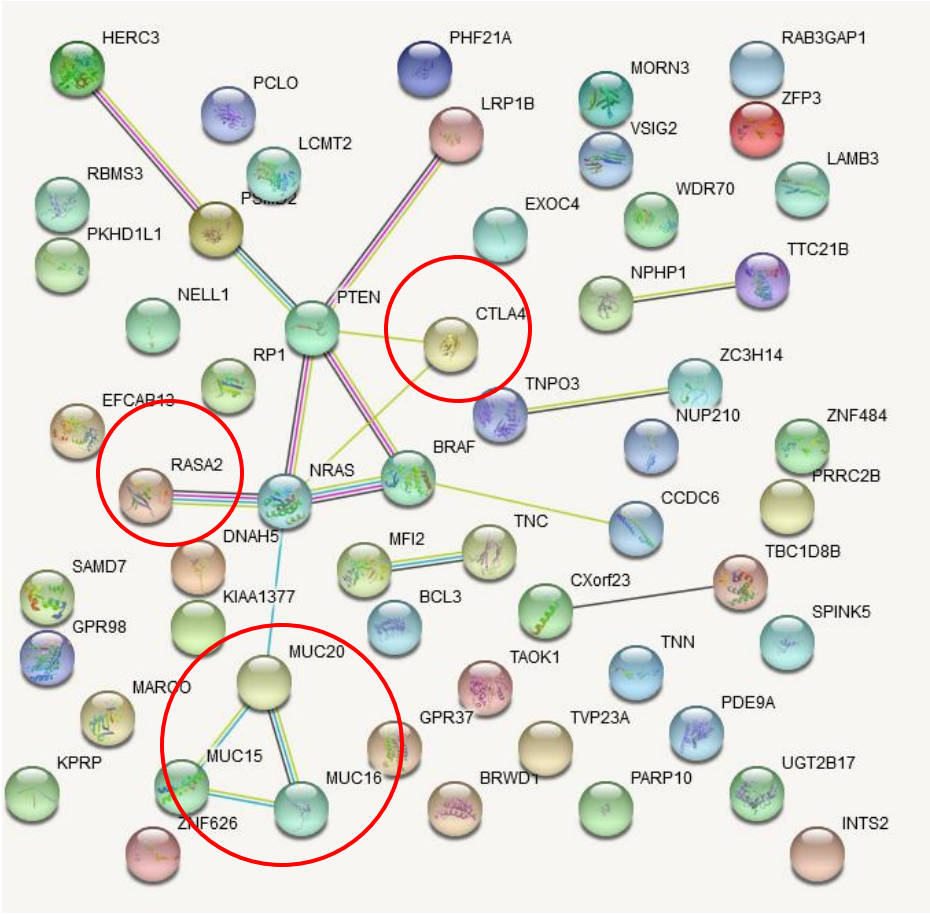

Select Melanoma Mutations
hits
Pattern
Translation_Start_Site
In_Frame_Ins
In_Frame_Del
3'UTR
5'UTR
Splice_Region
Frame_Shift_Ins
Intron
Frame_Shift_Del
Nonsense_Mutation
Splice_Site
Silent
Missense_Mutation
none
467 samples
124 events
18 genes
18 patterns

Cancer Evolution: CAPRI Model output



Cancer Evolution: Melanoma Progression additional insights

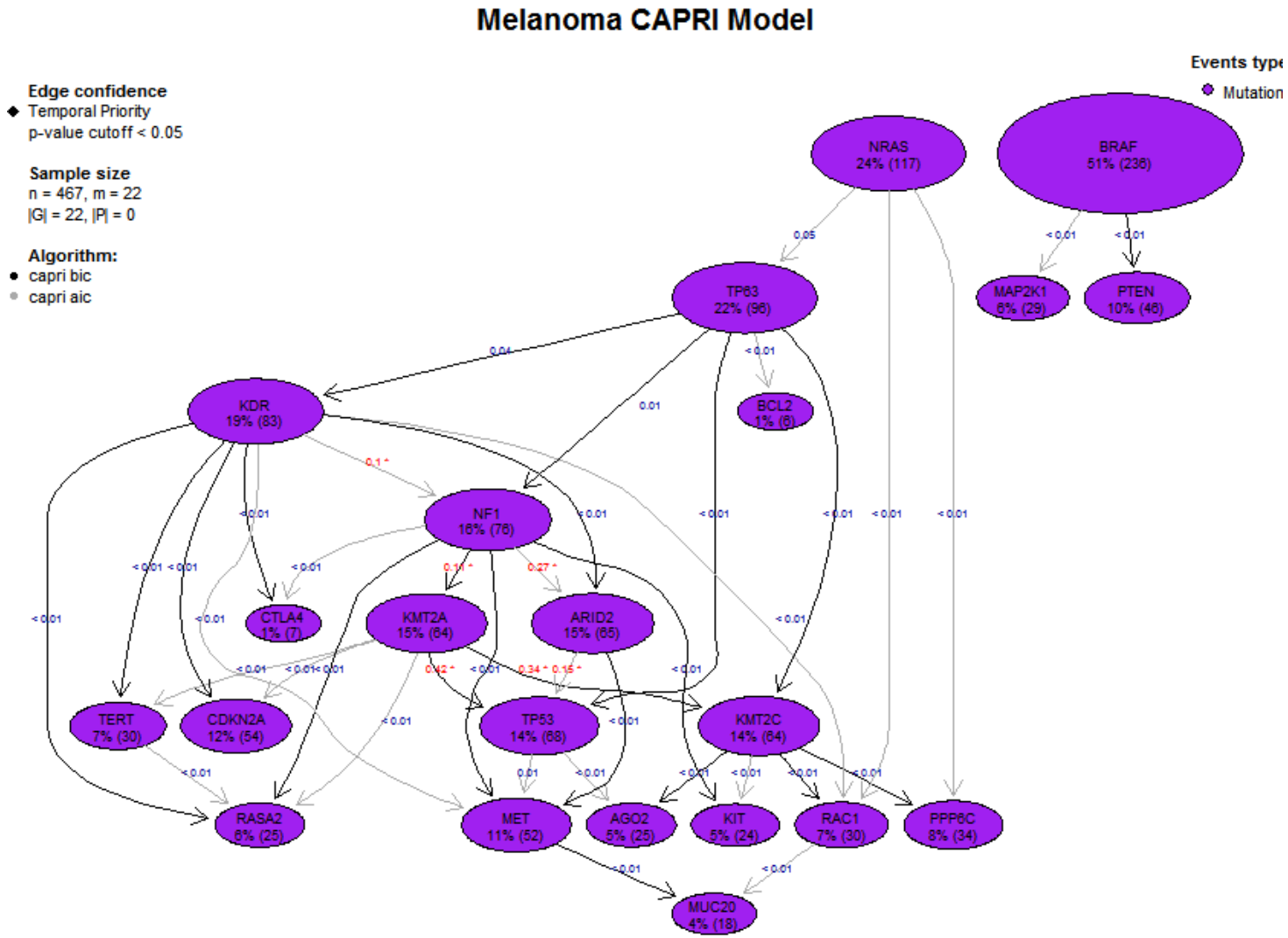
Below is a network from the String database with the top 50 occurring mutations for Melanoma -



From the network we can gain additional insights into why the NRAS pathway has a worse long-term survival rate -

- MUC16 mutation is associated with tumor mutation burden and its prognostic implications in cutaneous melanoma (Wang, et al., 2020).
- MUC20 and MUC15 are also part of a local network cluster causing familial hyperphosphatemic tumoral calcinosis (CL:26957).
- Anti- CTLA4 has been used as a successful therapy to increase survival in Melanoma (Steinberg, 2020).
- RASA2 is included in a local network of oncogenic MAPK signaling.
- MUC20 is also noted in the reactome pathway as activated MET receptor recruits the RAS signaling.

CANCER EVOLUTION: CAPRI MODEL OUTPUT ADDITIONAL INSIGHTS



Cancer Evolution: Final Thoughts & Considerations

- More data curation is needed-
  - Investigate pathway relations
  - Patterns, exclusivity etc.
  - Different hypotheses
- Overall the CAPRI model showed a good progression inference
- Can provide important information needed to create/ refine targeted treatment options

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```

Data visualization

```
# change colors, dark green is too dark
skcm_dataset<- change.color(skcm_dataset, 'Mutation', 'purple')

# see the on-coprint
oncoprint(skcm_dataset, title = 'Select Melanoma Mutations',
          samples.cluster = T,
          genes.cluster = T)
```

Step 3-6: CAPRI Model Inference

```
#### Run the Capri Model ####
model<-tronco.capri(skcm_dataset,
             nboot = 5)
# plot the model
tronco.plot(model,
            fontsize = 18,
            scale.nodes = .55,
            height.logic = 0.5,
            confidence = 'tp',
            legend.cex = 0.7,
            legend.pos = 'bottom',
            label.edge.size = 16,
            title = 'Melanoma CAPRI Model')
```

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