

# Cancer ecology and evolution

*N. Alcalá*

Rare Cancers Genomics Team

November 3rd 2023

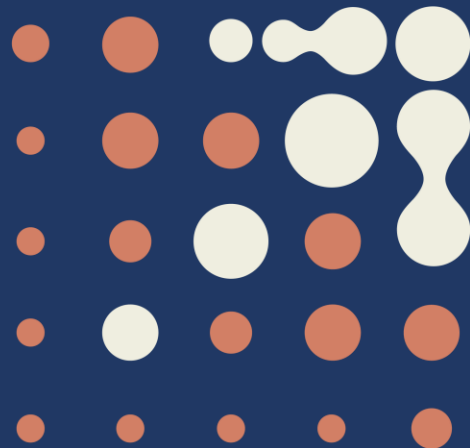
International Agency  
for Research on Cancer



World Health  
Organization



@nlalcala



# Plan

1. Introduction: **a theory of cancer**, with ecology and evolution as main conceptual frameworks
2. Reconstructing **tumor evolutionary trajectories**
3. From **microenvironmental pressures to tumor phenotypes**

# Tumor ecology & evolution | *Toward a theory of cancer*

Cell, Vol. 100, 57–70, January 7, 2000, Copyright ©2000 by Cell Press

## The Hallmarks of Cancer

## Review

Douglas Hanahan\* and Robert A. Weinberg†

"One day, we imagine that cancer biology and treatment—at present, a patchwork quilt of cell biology, genetics, histopathology, biochemistry, immunology, and pharmacology—will become **a science with a conceptual structure and logical coherence that rivals that of chemistry or physics.**"

"We foresee **cancer research developing into a logical science**, where the complexities of the disease [...] will become understandable in terms of a **small number of underlying principles.**"

# Tumor ecology & evolution | *Cancer evolutionary theory*

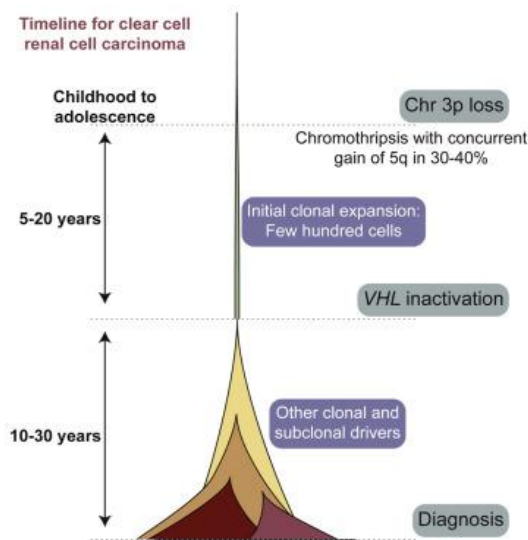
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## The Hallmarks of Cancer

## Review

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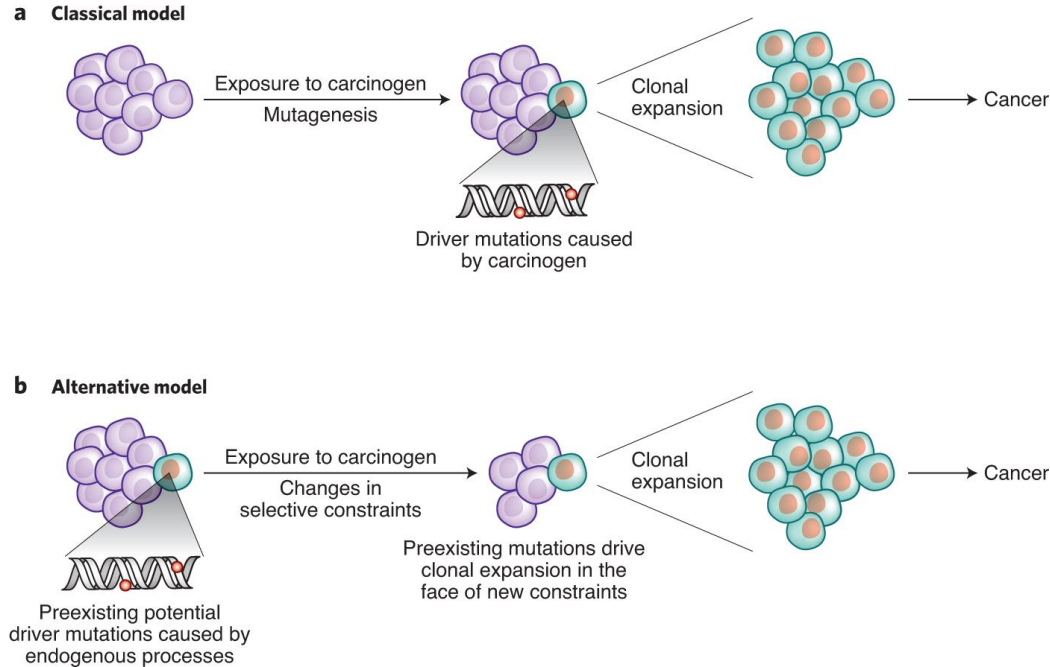
"[...] tumor development proceeds via a process formally analogous to Darwinian evolution, in which a succession of genetic changes, each conferring one or another type of growth advantage, leads to the progressive conversion of normal human cells into cancer cells."



TRACERX project (Mitchell *et al.* 2018).

# Tumor ecology & evolution | *Cancer evolutionary theory*

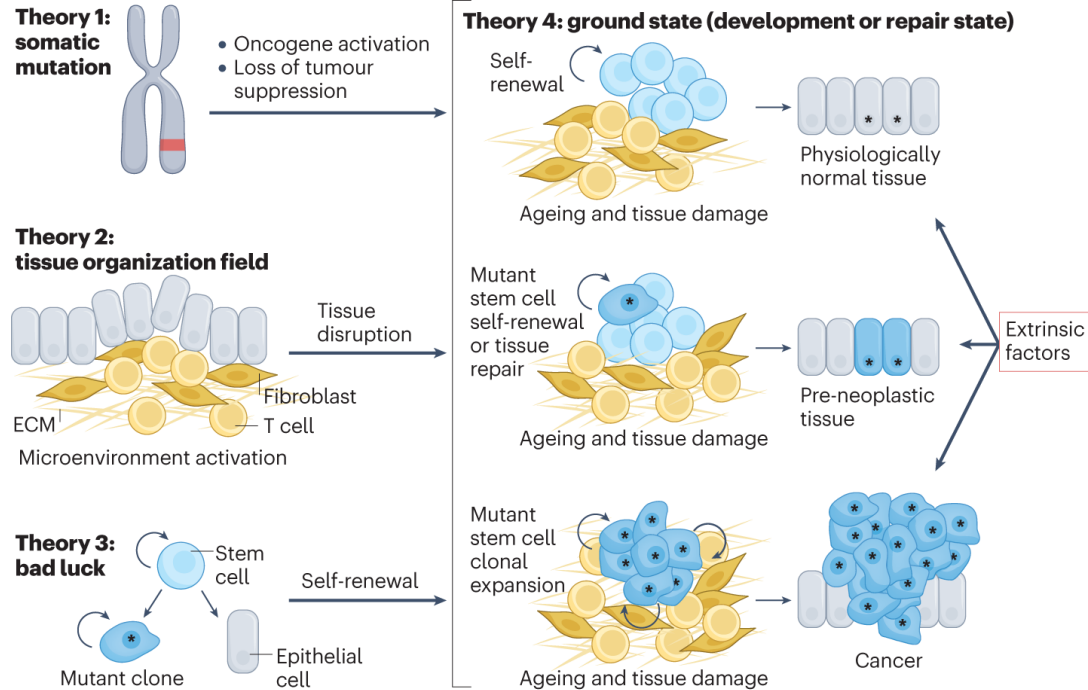
More recently, other models have been proposed



**Models of carcinogenesis. a**, classical model. **b**, promoter model (Lopez-Bigas and Gonzalez-Perez, *Nat Genet* 2020)

# Tumor ecology & evolution | *Cancer evolutionary theory*

More recently, other models have been proposed

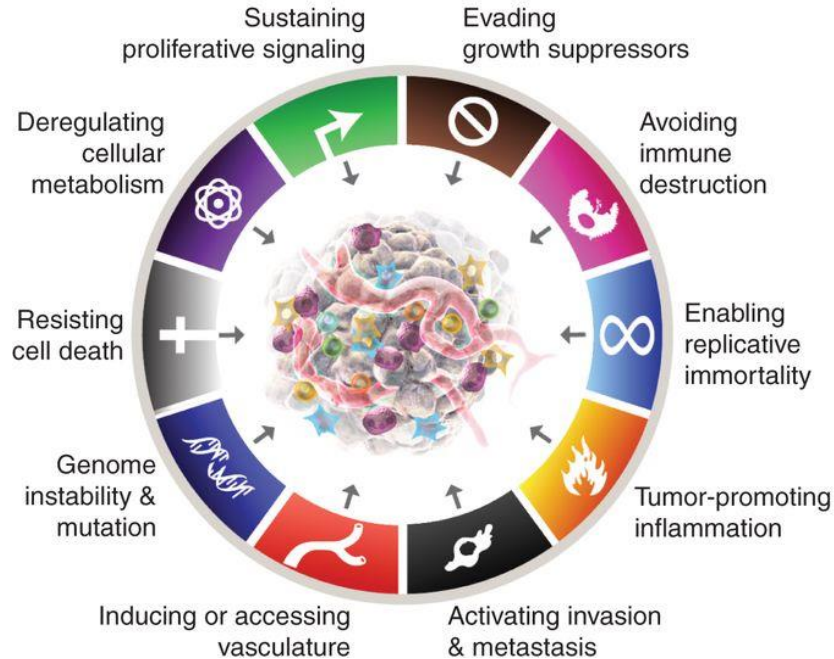


**Models of carcinogenesis.** 1, mutations in oncogenes or TSG. 2, chronic abnormal interactions between stroma and tissue. 3, tissue ground state determines phenotypic effect of mutations (Jassim *et al. Nat Rev Canc* 2023)

# Tumor ecology & evolution | *Cancer phenotypes*

Enumerating **cancer phenotypes** is central in their endeavor:

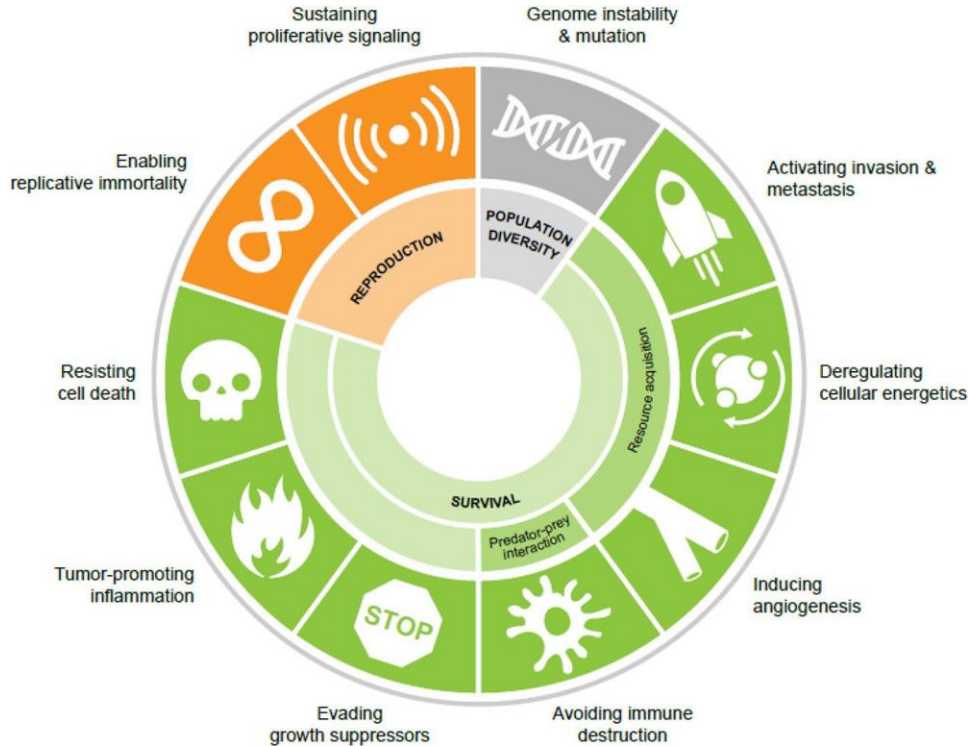
> 10 **hallmarks of cancer**, biological capabilities that enable malignant growth



The hallmarks of cancer (Weinberg and Hanahan 2015).

# Tumor ecology & evolution | *Phenotypes and ecology*

Hallmarks of cancer can be interpreted as **ecological strategies**



Hallmarks of cancer as ecological fitness parameters (Somarelli *Front. Ecol. Evol.* 2021).

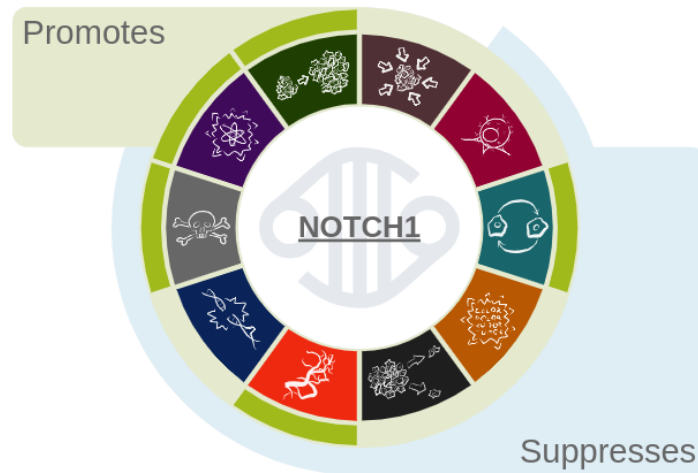


# Tumor ecology & evolution | *From genotypes to phenotypes*

Cancer phenotypes result from a **breach of anticancer mechanisms** because of genetic or epigenetic **alterations in cancer genes**

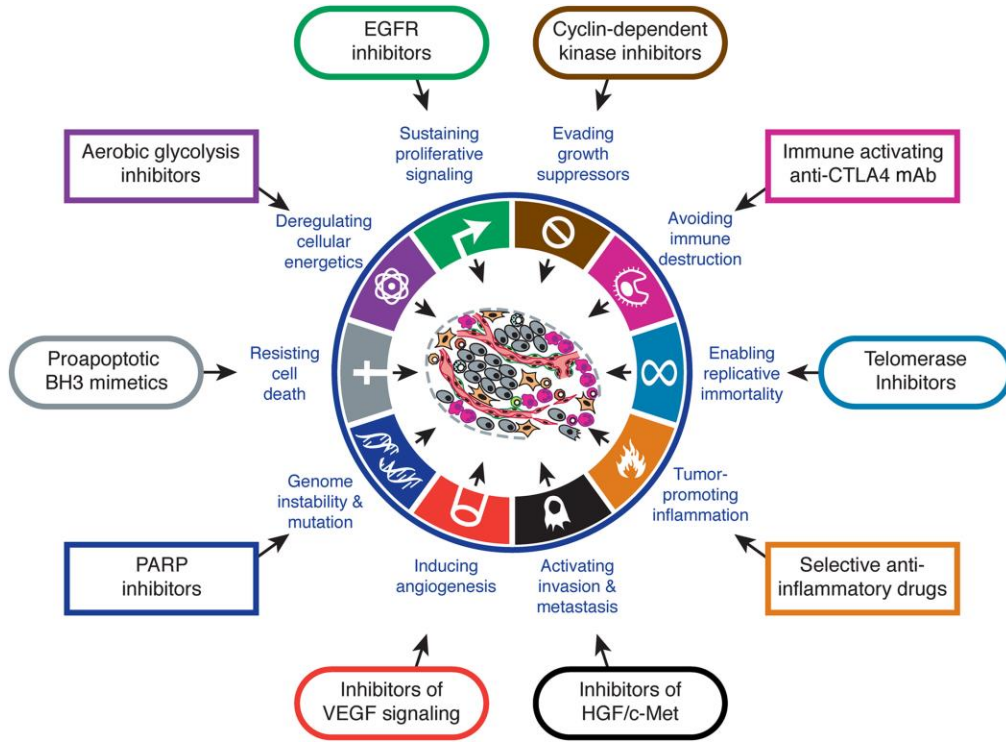
Two types of "cancer genes":

- **Oncogenes**, proto-genes that can be switched on by genetic or epigenetic alteration
- **Tumor suppressor genes**, "house keeping" genes whose inactivation promote cancer



*NOTCH1* Hallmarks profile (COSMIC database).

# Tumor ecology & evolution | *Phenotype and treatment*



Therapeutic targeting of the hallmarks of cancer (Weinberg and Hanahan *Cell* 2015).

# Reconstructing tumor evolutionary trajectories

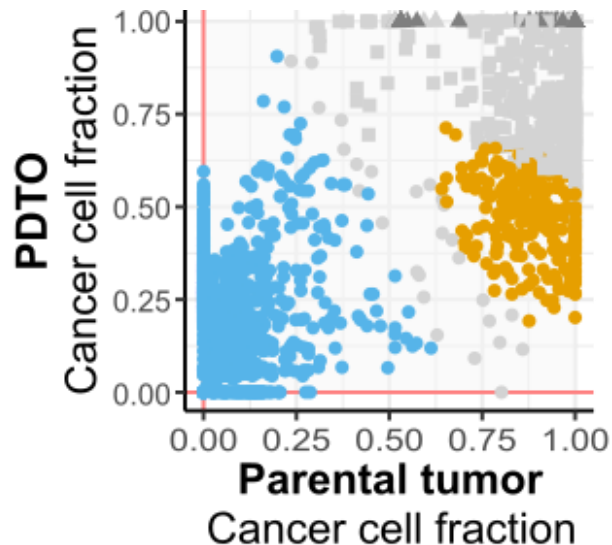
# Intra-tumor diversity | *Evolutionary trajectory inference*

Theoretical frameworks are based on

- **molecular phylogenetics**: the study of mutations in species—at macro-evolutionary time-scales
- **population genetics**: the study of mutations in populations—at micro-evolutionary time-scales

# Intra-tumor diversity | *Phylogenetic approaches*

1. Cluster mutations based on the **cancer cell fraction** (proportion of cells carrying this mutation, determined in NGS by the allelic fraction, the proportion of reads with the ALT allele)



## Clonality

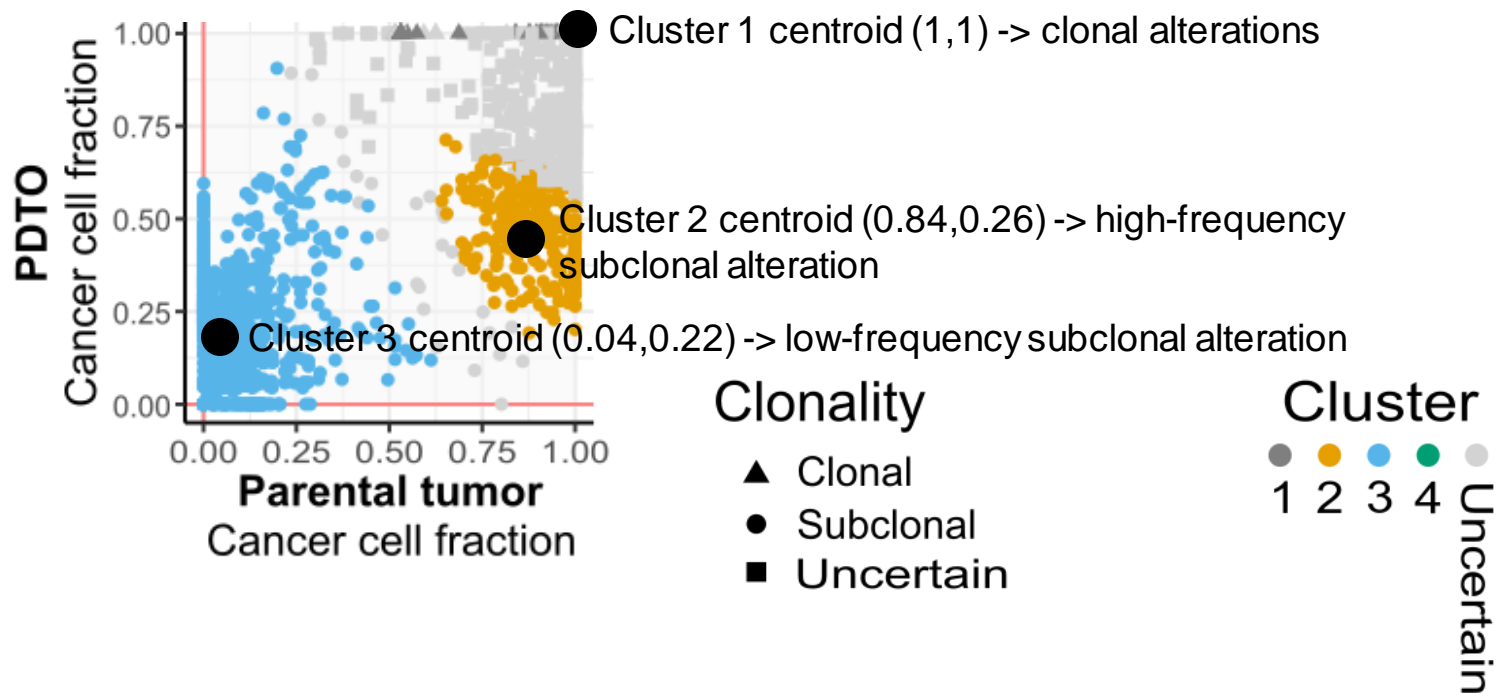
- ▲ Clonal
- Subclonal
- Uncertain

## Cluster



# Intra-tumor diversity | *Phylogenetic approaches*

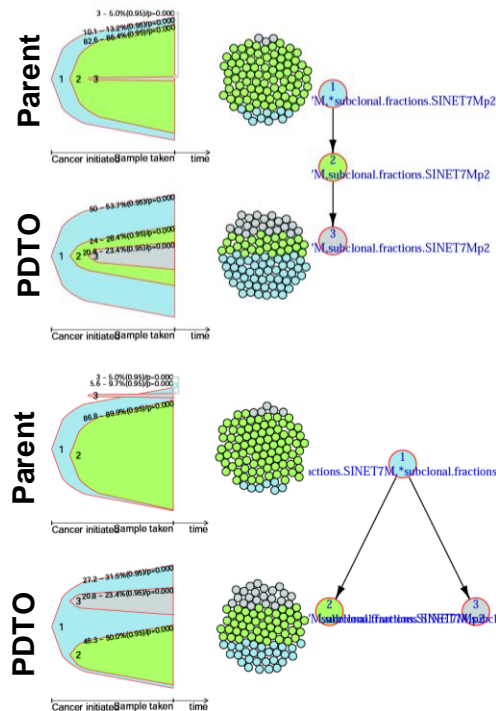
1. Cluster mutations based on the **cancer cell fraction** (proportion of cells carrying this mutation, determined in NGS by the allelic fraction, the proportion of reads with the ALT allele)



# Intra-tumor diversity | *Phylogenetic approaches*

2. Enumerate possible **parsimonious phylogenetic trees** using "pidgeonhole principle": sum of CCF of subclones cannot exceed that of parental clone

- Cluster 1 centroid (1,1) -> clonal alterations
- Cluster 2 centroid (0.84,0.26) -> high-frequency subclonal alteration
- Cluster 3 centroid (0.04,0.22) -> low-frequency subclonal alteration

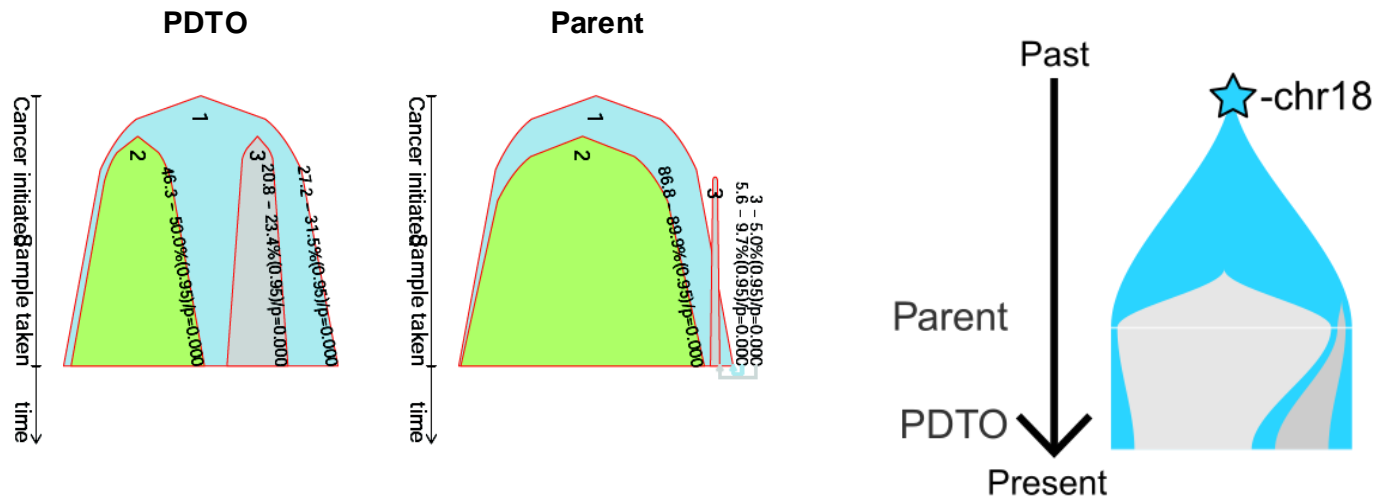


Solution 1: linear evolution

Solution 2: branching evolution

# Intra-tumor diversity | *Phylogenetic approaches*

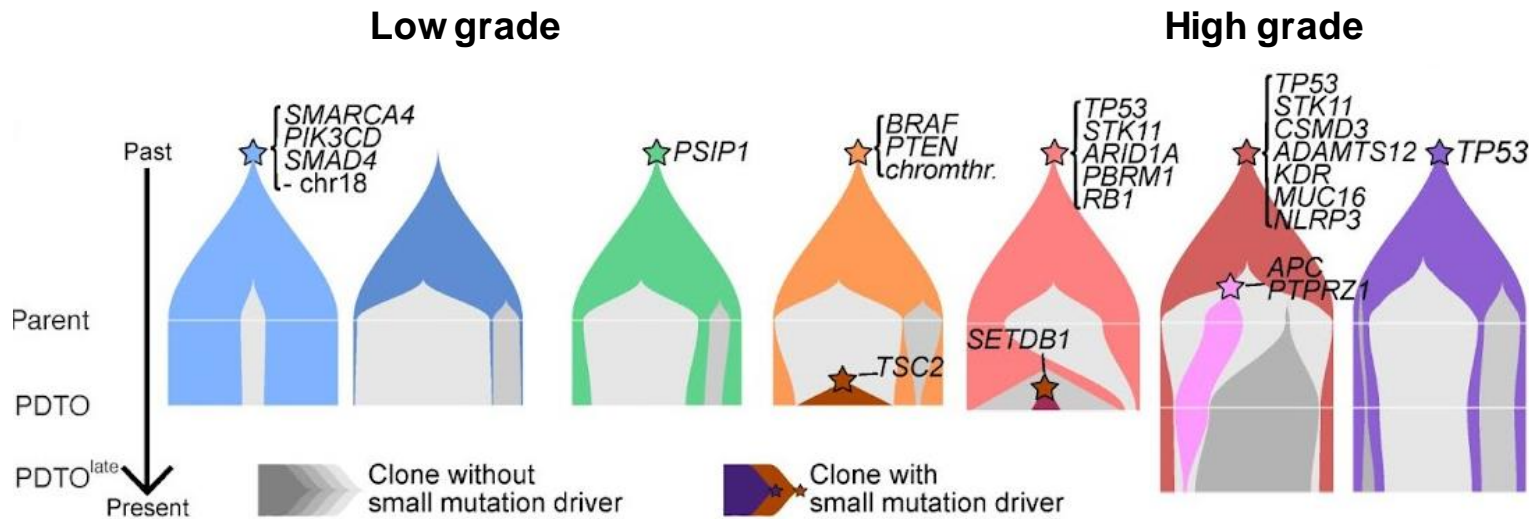
3. Reconcile multiple samples (regions / time-points) to infer spatial or temporal changes in clone frequencies





# Intra-tumor diversity | *Phylogenetic approaches*

3. Reconcile multiple samples (regions / time-points) to infer spatial or temporal changes in clone frequencies



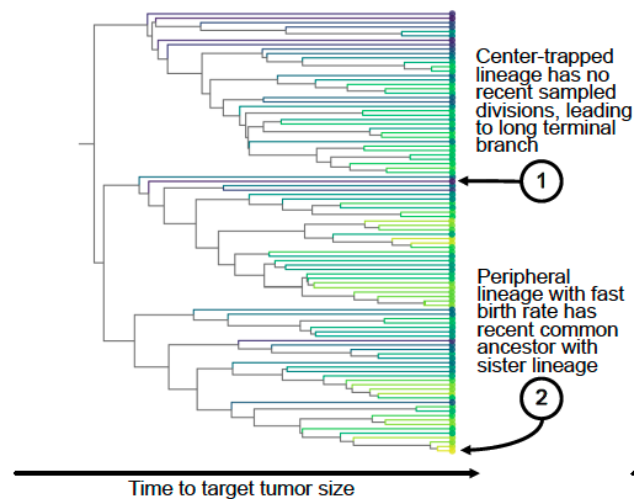
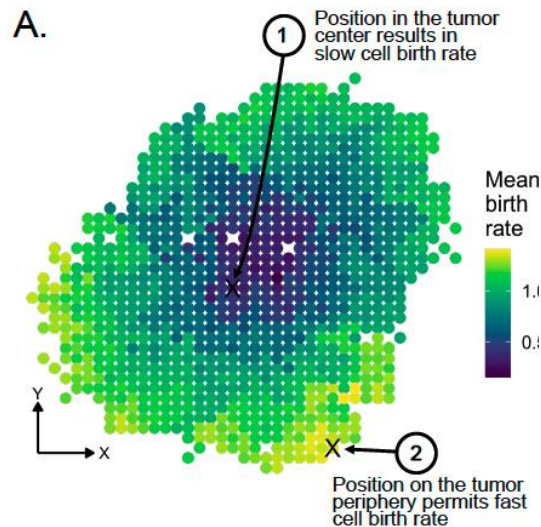
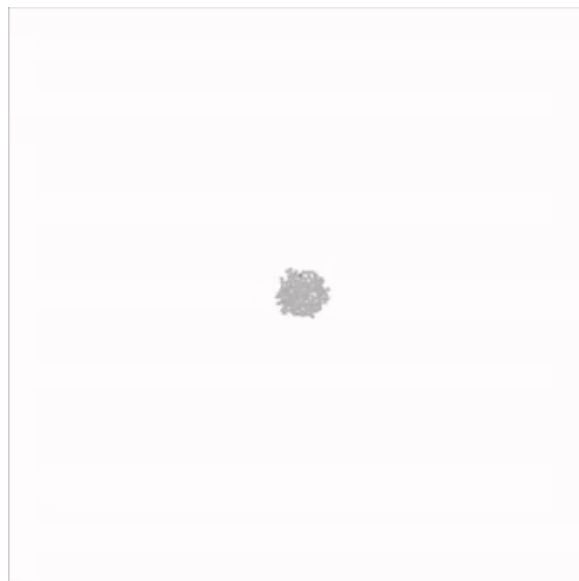
# Intra-tumor diversity | *Evolutionary trajectory inference*

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- **population genetics**: the study of mutations in populations—at micro-evolutionary time-scales

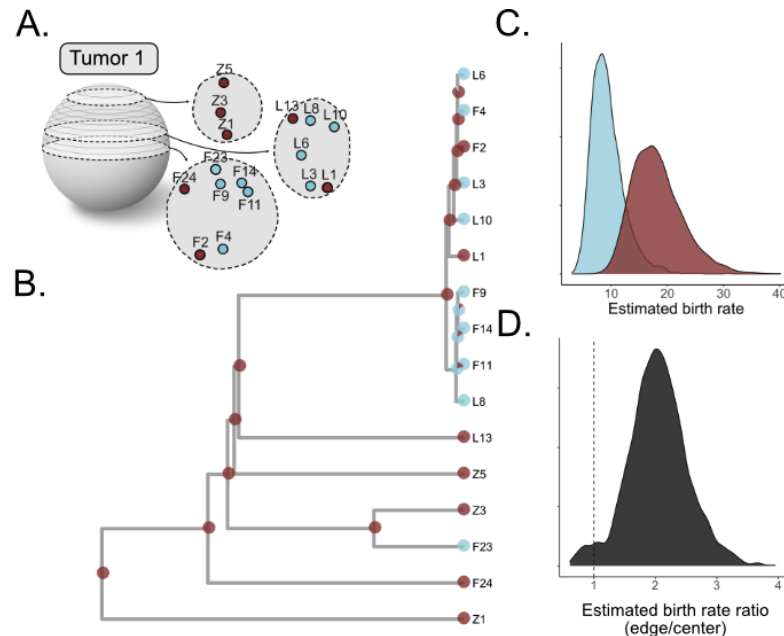
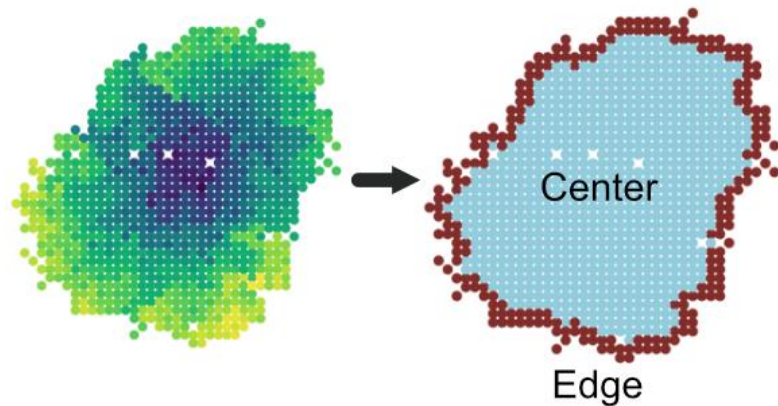
# Intra-tumor diversity | *Population-genetic approaches*

1. Model tumor growth
2. Build cell genealogical trees from tumor



# Intra-tumor diversity | *Population-genetic approaches*

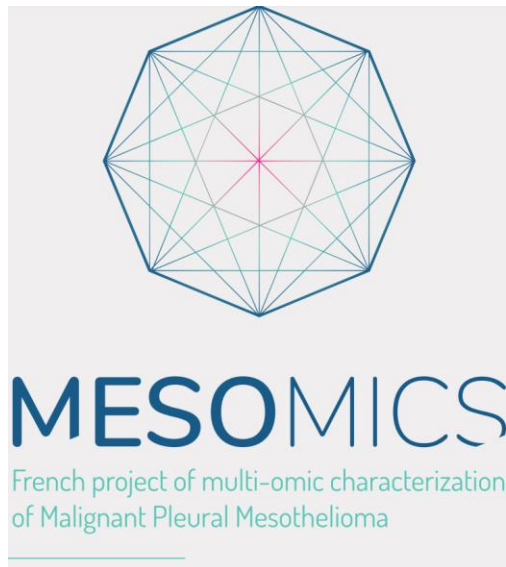
3. Use model predictions (link birth-rate / position) to infer spatial position of cells from genealogies and growth advantage of edge cells



Application to hepatocellular multi-regional sequencing

# From micro-environment to tumor phenotype

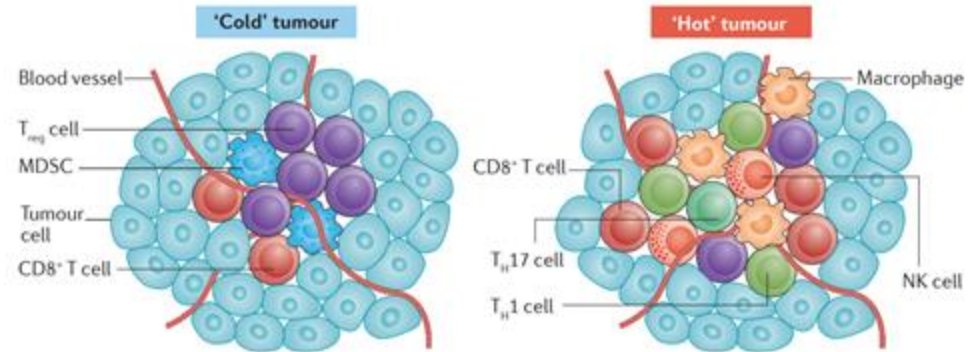
*Example of malignant pleural mesothelioma phenotypes*



# From environment to phenotype | *Heterogeneity*

## Tumors contain a mixture of cells

> we sequence simultaneously different tumor cell populations, immune "predator" cells, neighbouring tissue, ...



Tumors differ in their level of infiltration. Source: Nagarsheth et al. *Nat Rev Immun* 2017.

# From environment to phenotype | *Heterogeneity*

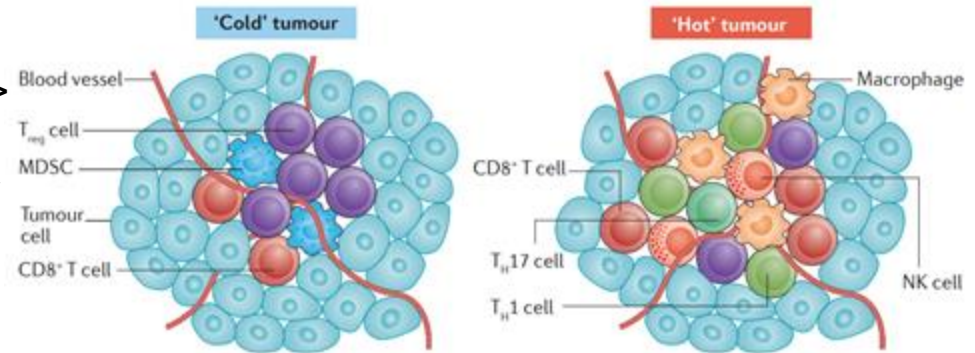
## Tumors contain a mixture of cells

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> **multi-omic sequencing** provides an opportunity to learn about the tumor genotype, phenotype, and the tumor micro-environment:

>> genomic data -> tumor genotype, what is "hard-wired" in the tumor

>> transcriptomic data/epigenomic data -> which cells are present in the micro-environment, what pathways are tumor cells activating (phenotypes), how do they interact with the environment

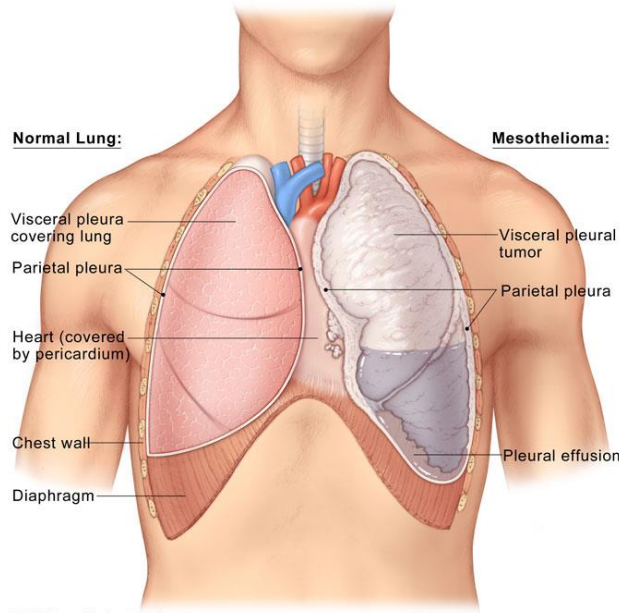


Tumors differ in their level of infiltration. Source: Nagarsheth et al. *Nat Rev Immun* 2017.

# From environment to phenotype | *The case of mesothelioma*

## Malignant pleural mesothelioma

- Rare and deadly cancer arising in the linings of the lung (pleura)
- Mostly associated with **asbestos exposure**
- Asbestos is banned in many countries but **lag between exposure and disease ~30-40 years** ⇒ incidence still rising



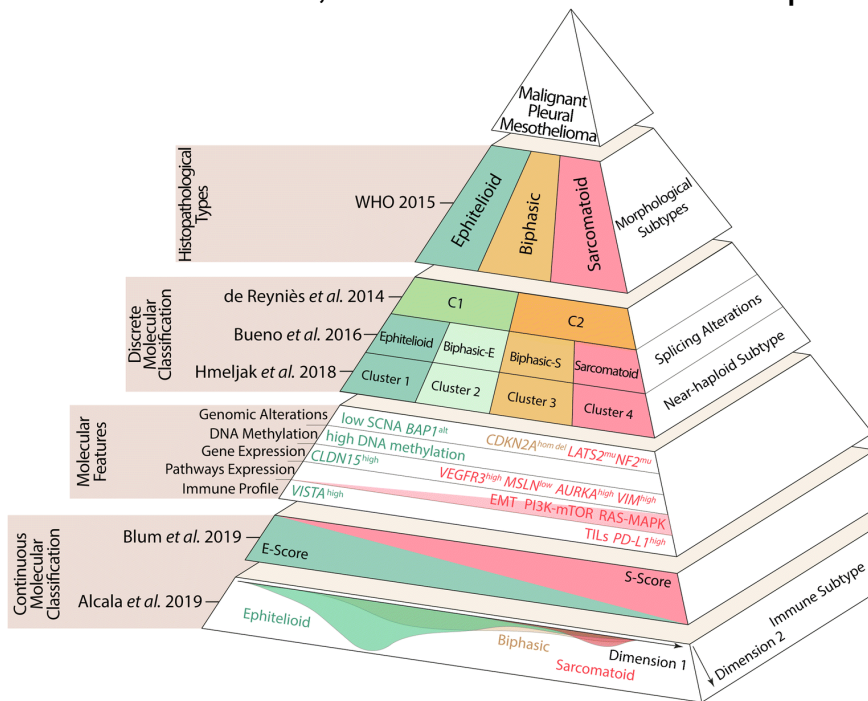
Stanford Medicine Dept of Surgery



# From environment to phenotype | *The case of mesothelioma*

## Malignant pleural mesothelioma (MPM)

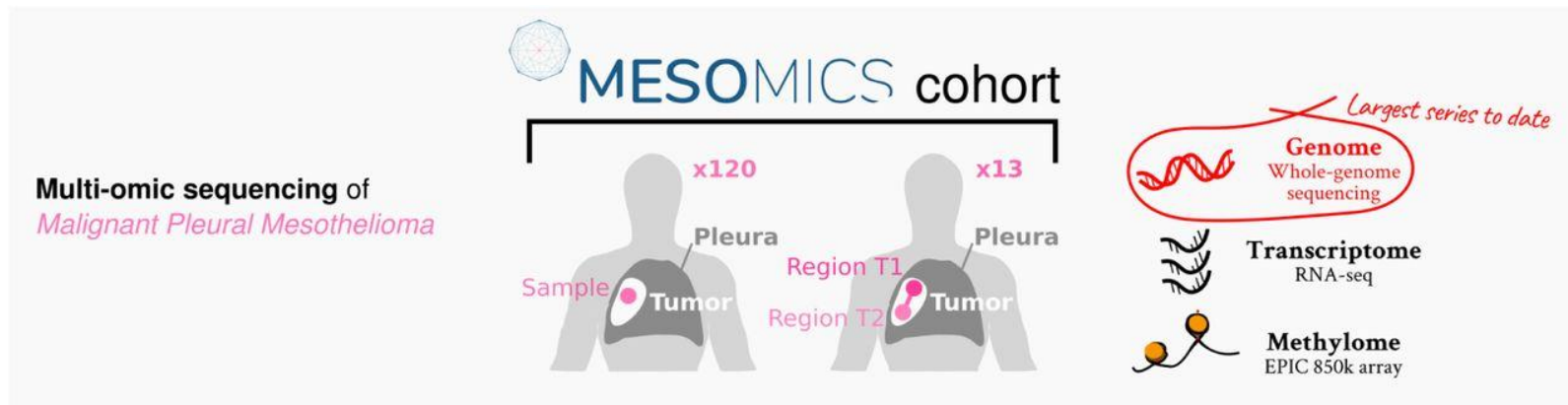
- Current WHO classification considers 3 types; molecular classifications further subdivide them
- Nevertheless, the extent of molecular phenotypes is unknown



Schematic representation of current MPM classifications (Fernandez-Cesta et al. Virchows Archive 2021)

# From environment to phenotype | *The case of mesothelioma*

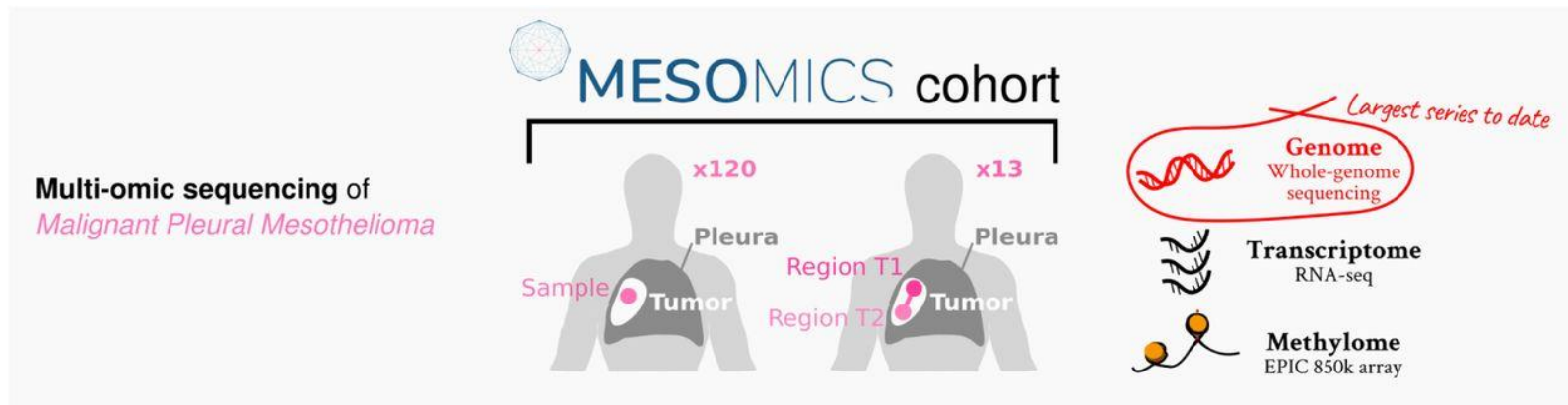
**MESOMICS study:** further define inter-patient molecular variation



Mangiante\*, Alcalá\*, Sexton-Oates\*, Di Genova\*,  
et al. (Nature Genetics 2023)

# From environment to phenotype | *The case of mesothelioma*

**MESOMICS study:** further define inter-patient molecular variation



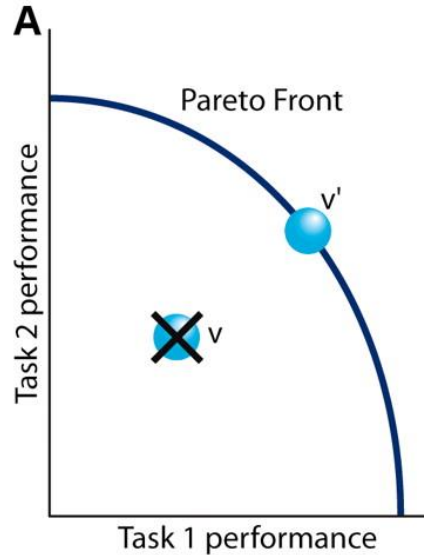
Mangiante\*, Alcalá\*, Sexton-Oates\*, Di Genova\*,  
et al. (Nature Genetics 2023)

1. Find molecular phenotypes
2. Associate microenvironment and phenotypes
3. Find genomic alterations associated with phenotypes
4. Generate evolutionary hypotheses

# From environment to phenotype

## Step 1: build a phenotypic map of MPM

Method: Archetypal analysis

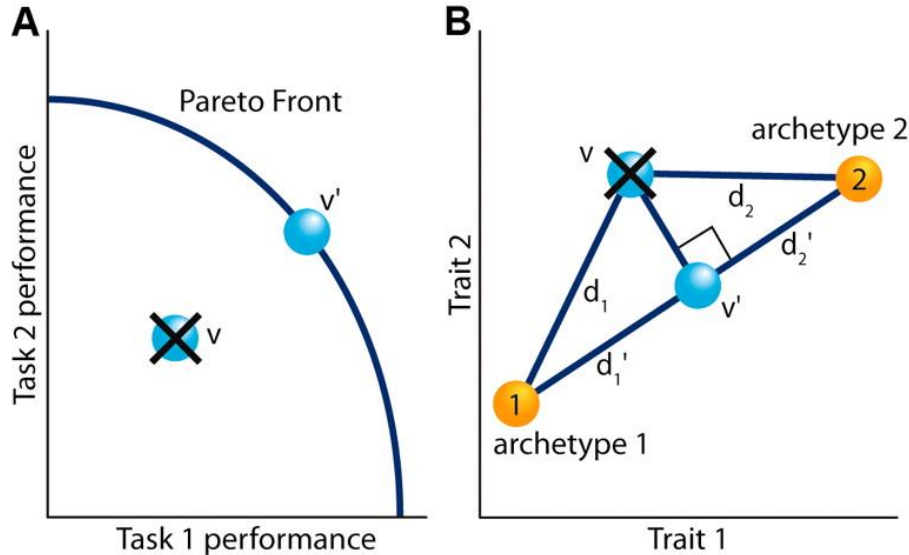


Evolutionary trade-offs and the geometry of phenotypic space (Shoval et al. Science 2012)

# From environment to phenotype | *Molecular phenotypes*

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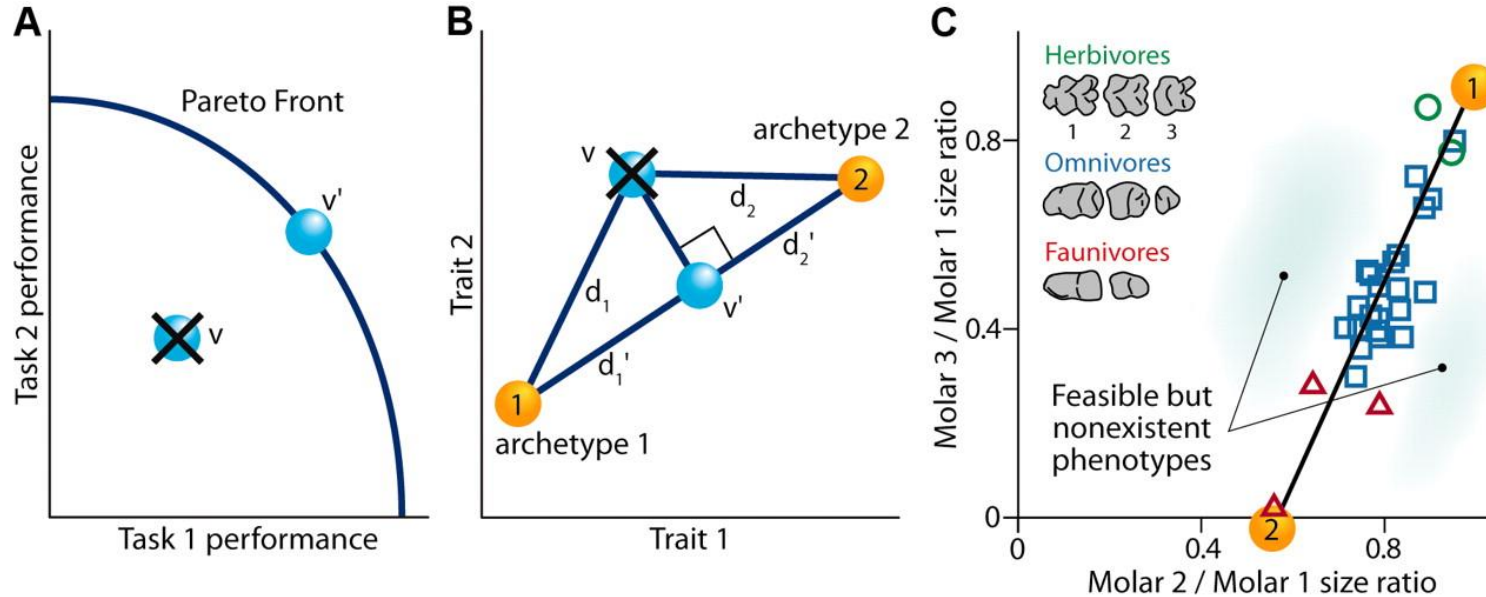


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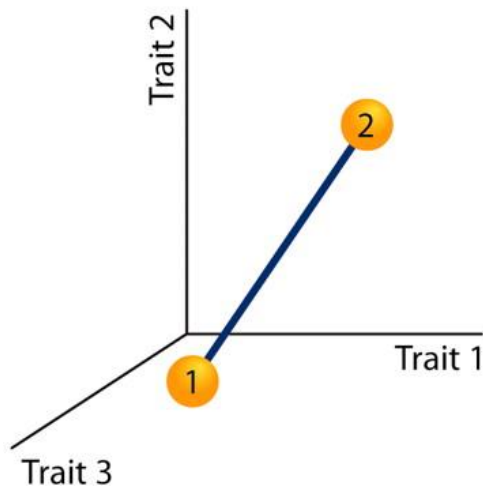
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# Tumor ecology & evolution | *A genotype-phenotype map*

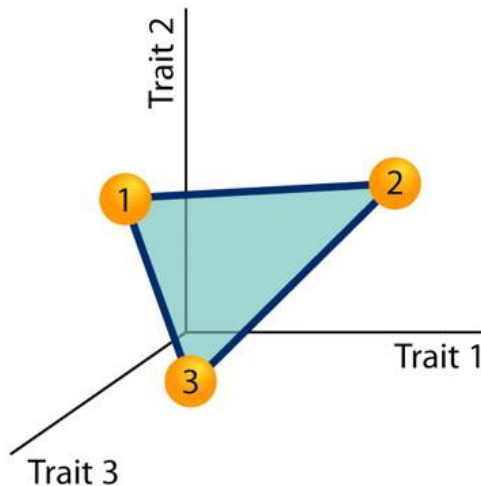
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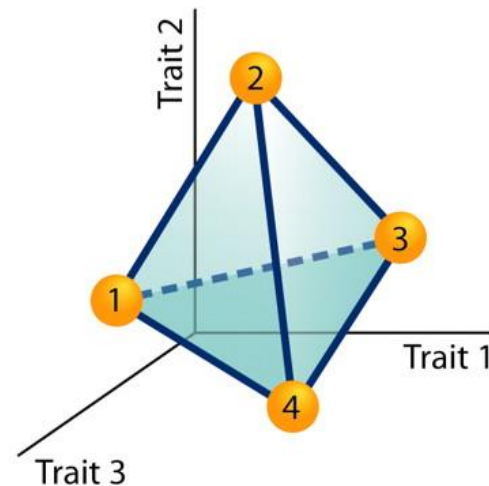
**A** 2 tasks



**B** 3 tasks



**C** 4 tasks

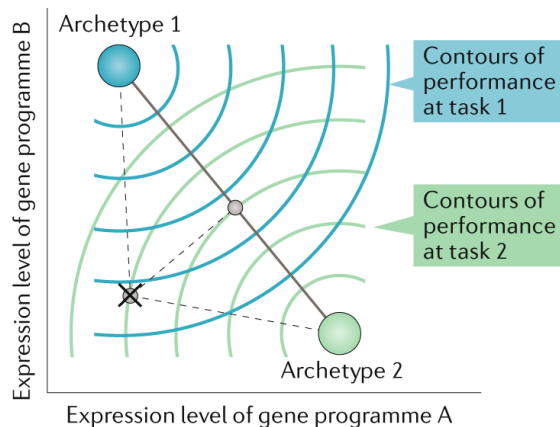


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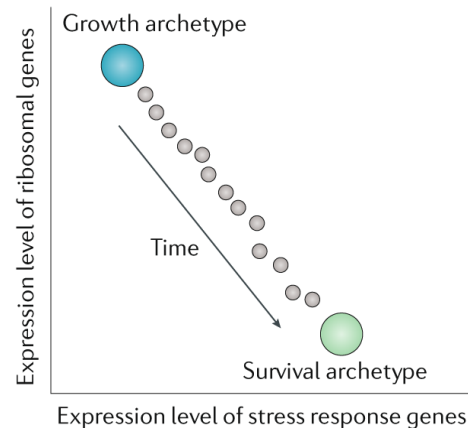
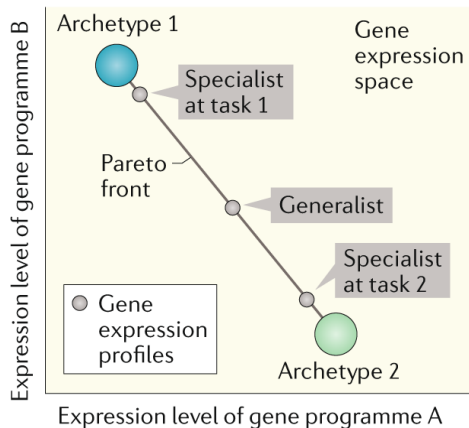
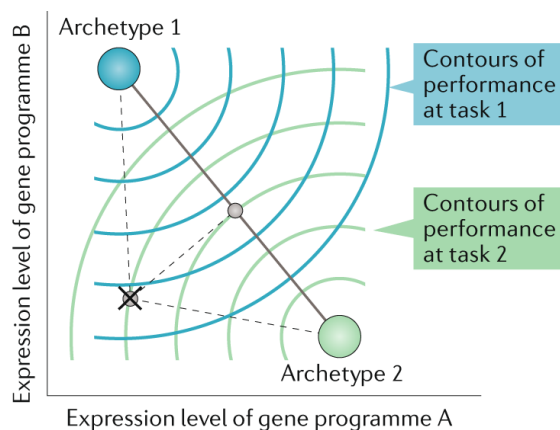
Evolutionary trade-offs in cancer. Expression is considered informative about cell function and thus a proxy for cancer phenotype (Hausser and Alon Nat Rev Cancer 2020)



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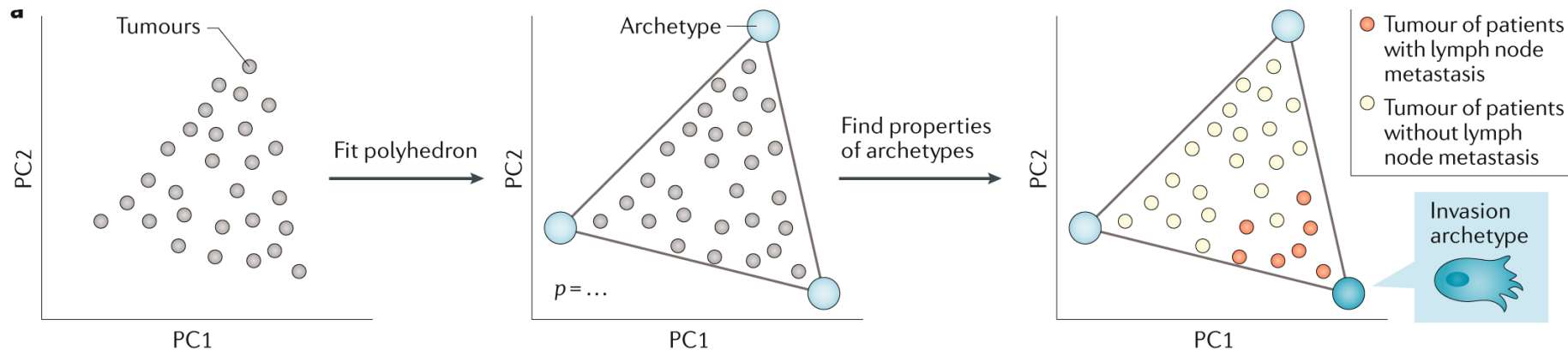


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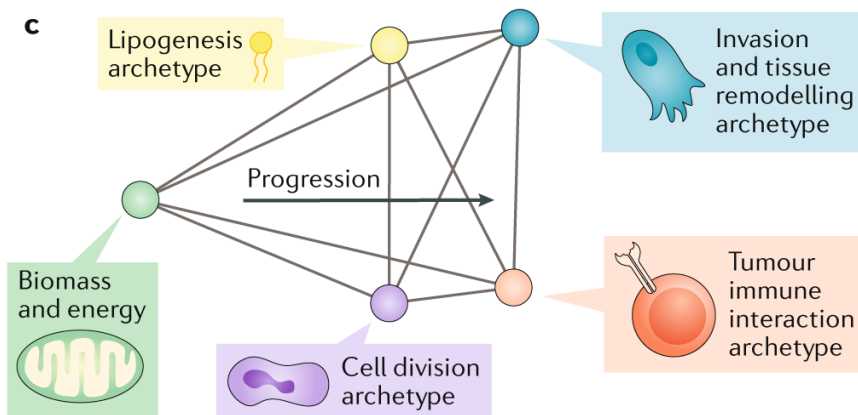
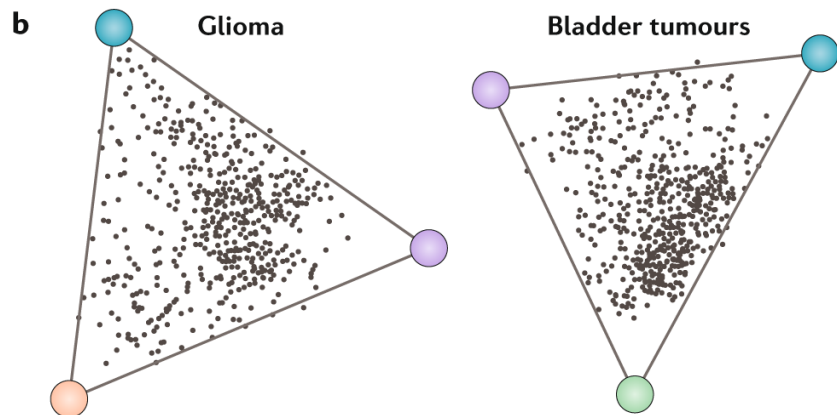


PCA is used as an unsupervised identification of latent expression variables that act as a proxy for cancer phenotypes (Hausser and Alon Nat Rev Cancer 2020)

# Tumor ecology & evolution | *A genotype-phenotype map*

## Step 1: build a phenotypic map of MPM

Method: Archetypal analysis

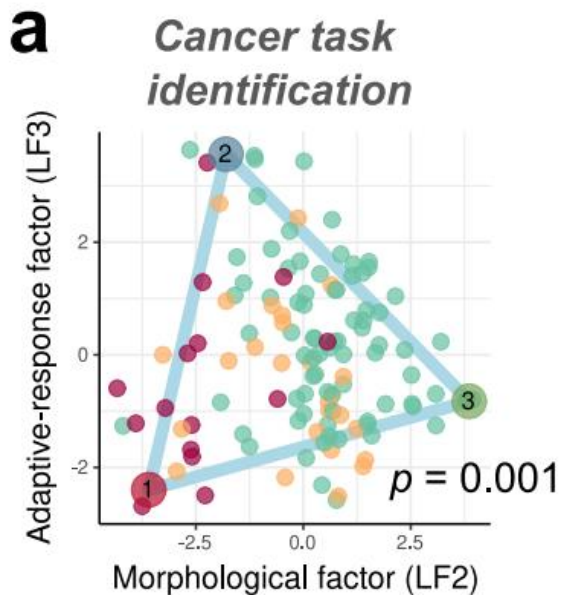


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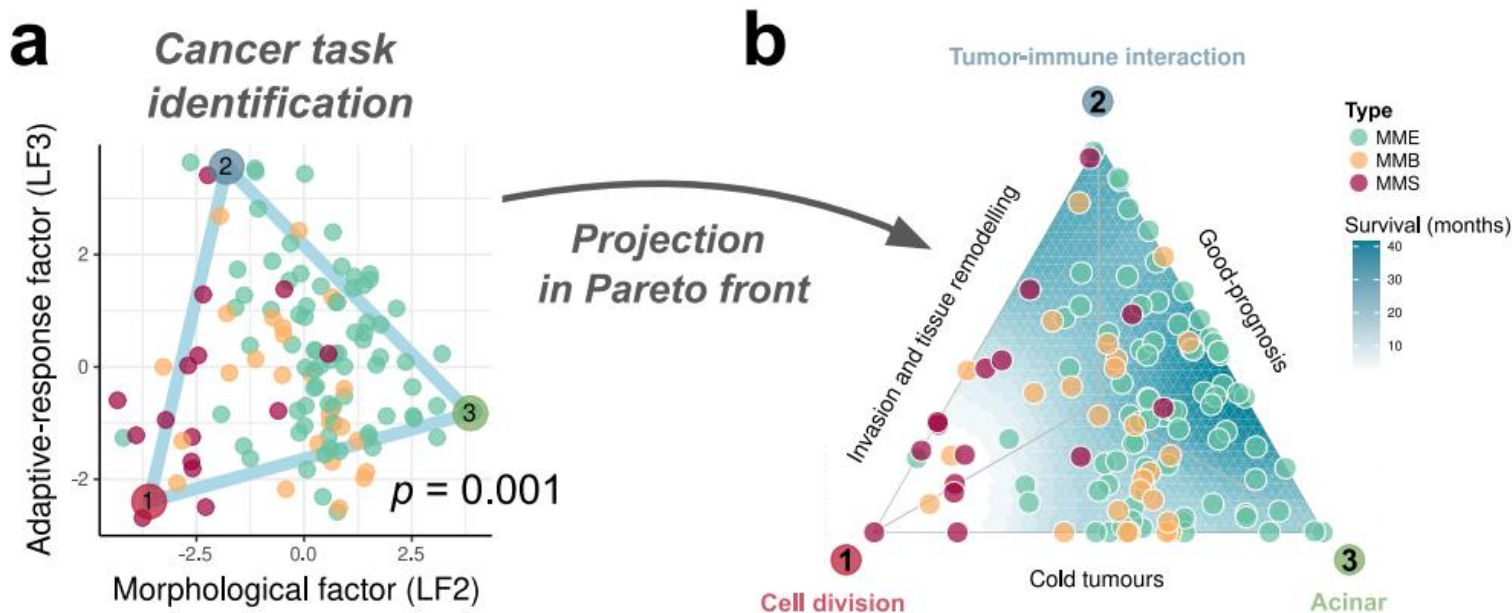


Evolutionary trade-offs in the MESOMICS cohort

# Tumor ecology & evolution | *A genotype-phenotype map*

## Step 1: build a phenotypic map of MPM

Method: Archetypal analysis

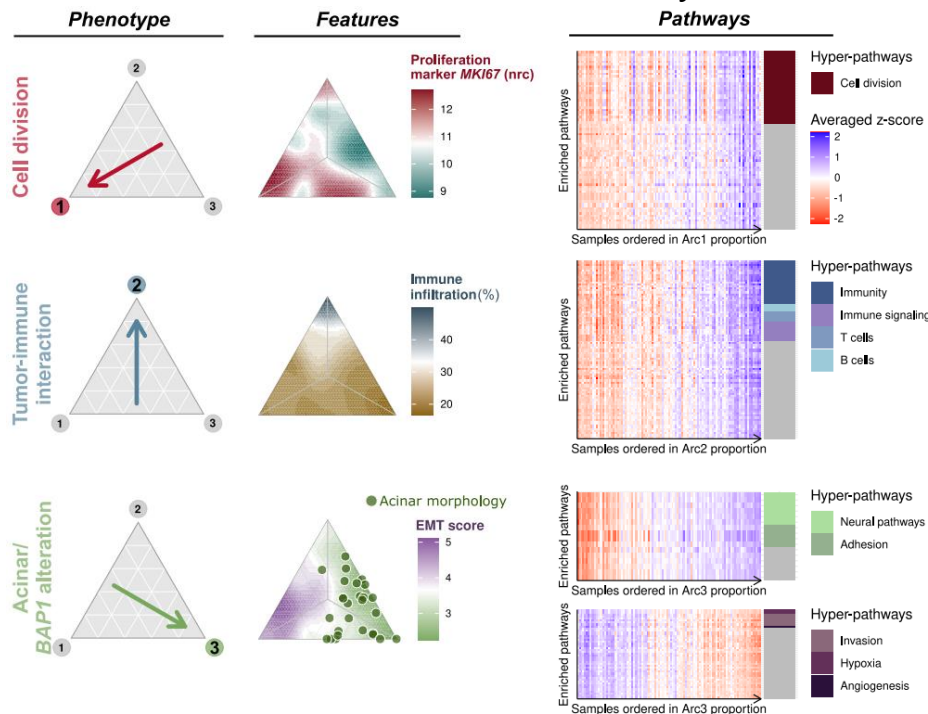


Evolutionary trade-offs in the MESOMICS cohort

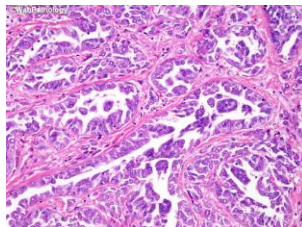
# Tumor ecology & evolution | *A genotype-phenotype map*

## Step 1: build a phenotypic map of MPM

Method: Gene set enrichment analysis of archetypes, association with cell morphology



Evolutionary trade-offs in the MESOMICS cohort

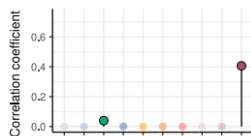
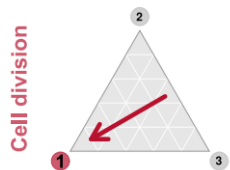


# Tumor ecology & evolution | *A genotype-phenotype map*

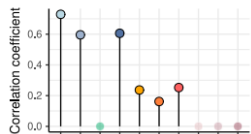
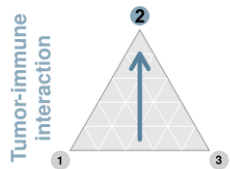
## Step 2: infer microenvironmental conditions of each phenotype

Method: Gene set enrichment analysis of archetypes, association with clinical variables

**Phenotype**                      **Microenvironment**

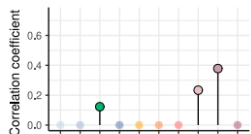
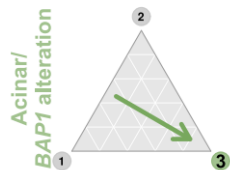


- > Scarce resources (necrosis, hypoxia)
- > Asbestos exposure (chronic inflammation?)



- > Strong immune predation
- > Asbestos exposure (chronic inflammation?)

Microenvironments associated with MPM archetypes

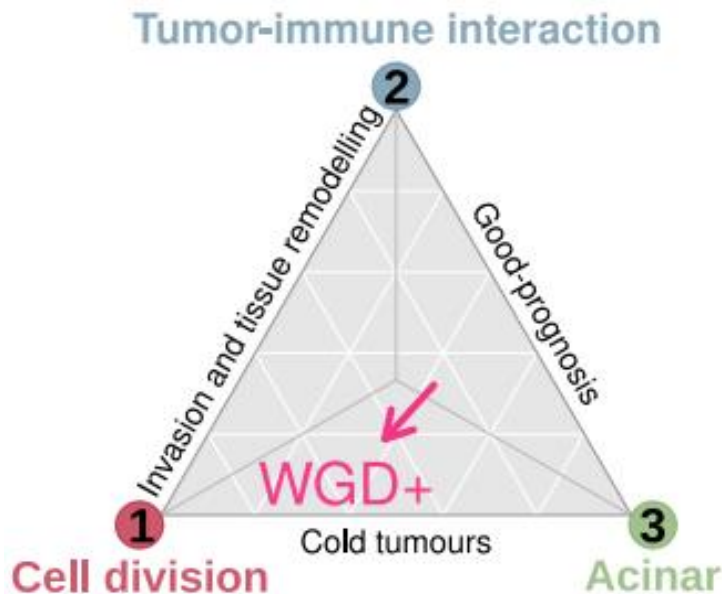


- > Favorable environment

# Tumor ecology & evolution | *A genotype-phenotype map*

## Step 3: find genomic alterations associated with each phenotype

Method: Compute “effect vectors” linking WT and altered samples in phenotypic space



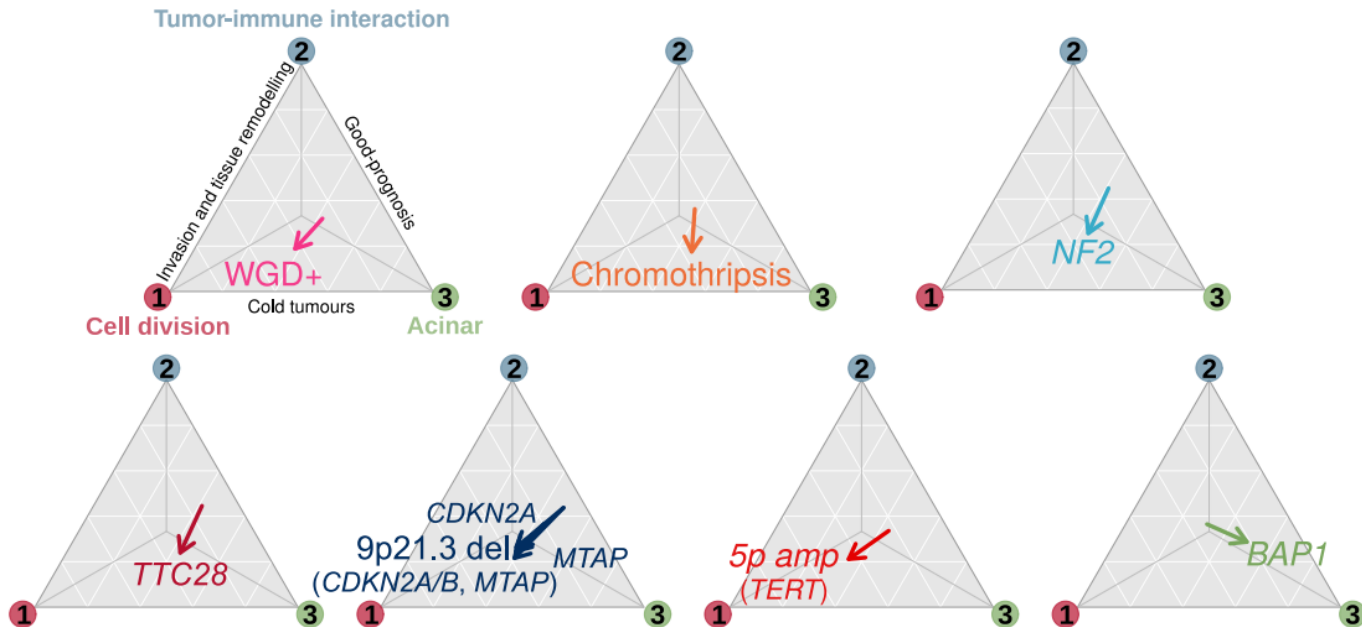
Genomic events tune phenotypic specialization in MPM



# Tumor ecology & evolution | *A genotype-phenotype map*

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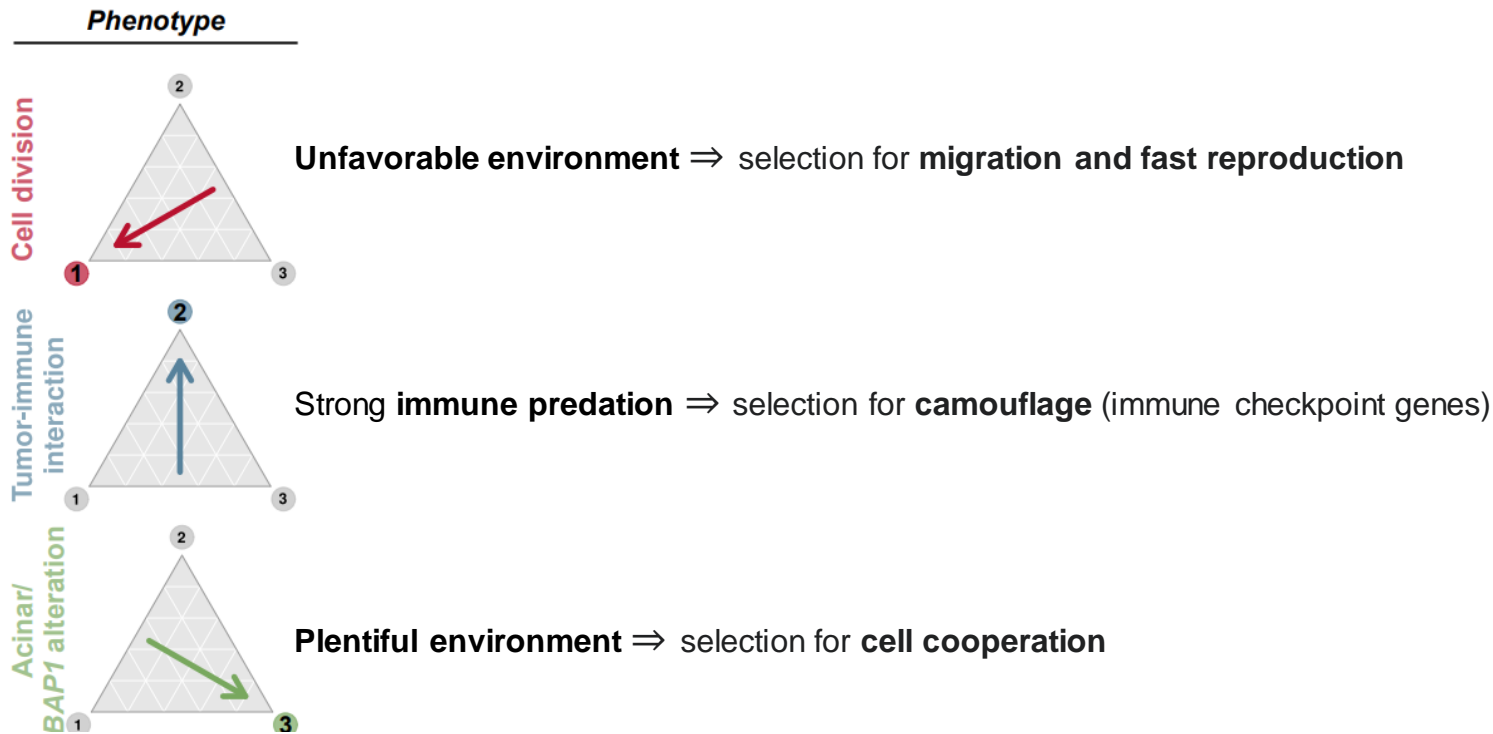
Method: Compute “effect vectors” linking WT and altered samples in phenotypic space



Genomic events tune phenotypic specialization in MPM

# Tumor ecology & evolution | *A genotype-phenotype map*

## Step 4: evolutionary hypotheses



## Tumor ecology & evolution | *Toward a theory of cancer*

- > The **convergence of experimental and theoretical models** can lead to **a theory of cancer as dreamt by Hanahan and Weinberg**
- > This novel theory of cancer will likely **have ecology and evolution at its center**

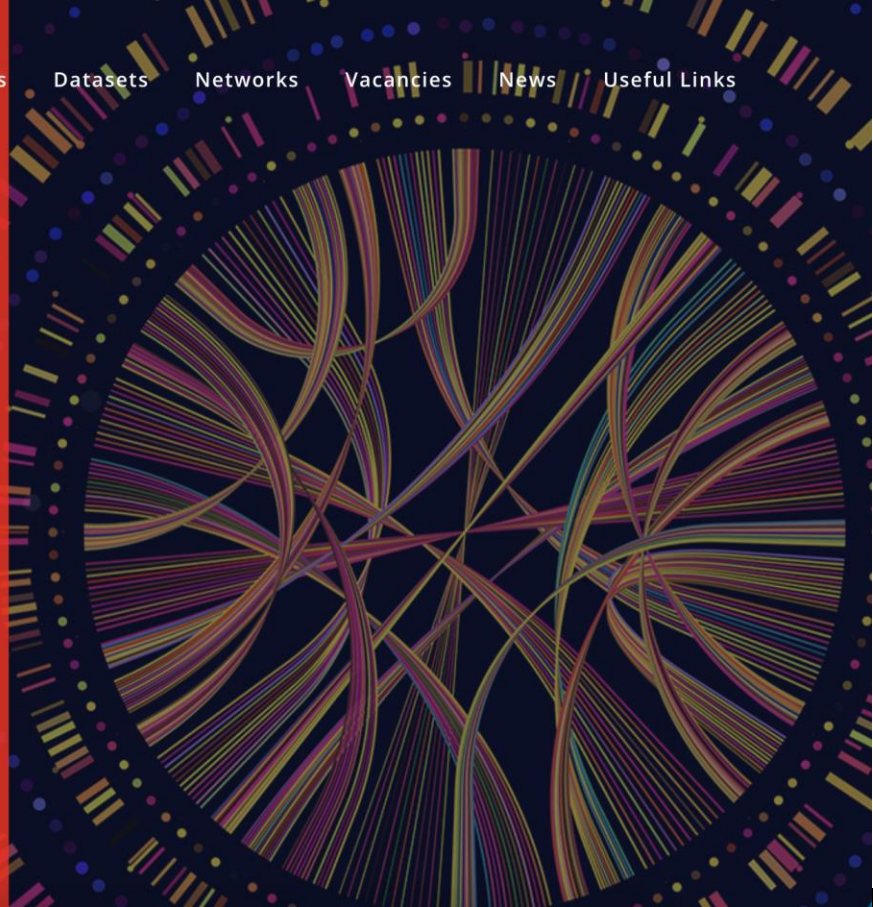
## Tumor ecology & evolution | *References*

- > Hanahan and Weinberg. The Hallmarks of cancer, *Cell* 2000
- > R scripts for tumor phylogenetics of lung NEN organoids [https://github.com/IARCBioinfo/MS\\_panNEN\\_organoids](https://github.com/IARCBioinfo/MS_panNEN_organoids)
- > R scripts for Pareto task inference of malignant pleural mesothelioma [https://github.com/IARCBioinfo/MESOMICS\\_data](https://github.com/IARCBioinfo/MESOMICS_data)
- > the <https://rarecancersgenomics.com/> initiative

[www.rarecancersgenomics.com](http://www.rarecancersgenomics.com)

# Rare Cancers Genomics

Multidisciplinary and multi-omics molecular  
characterisation of rare cancers



# THANKS