

# Epigenomics in PanNETs

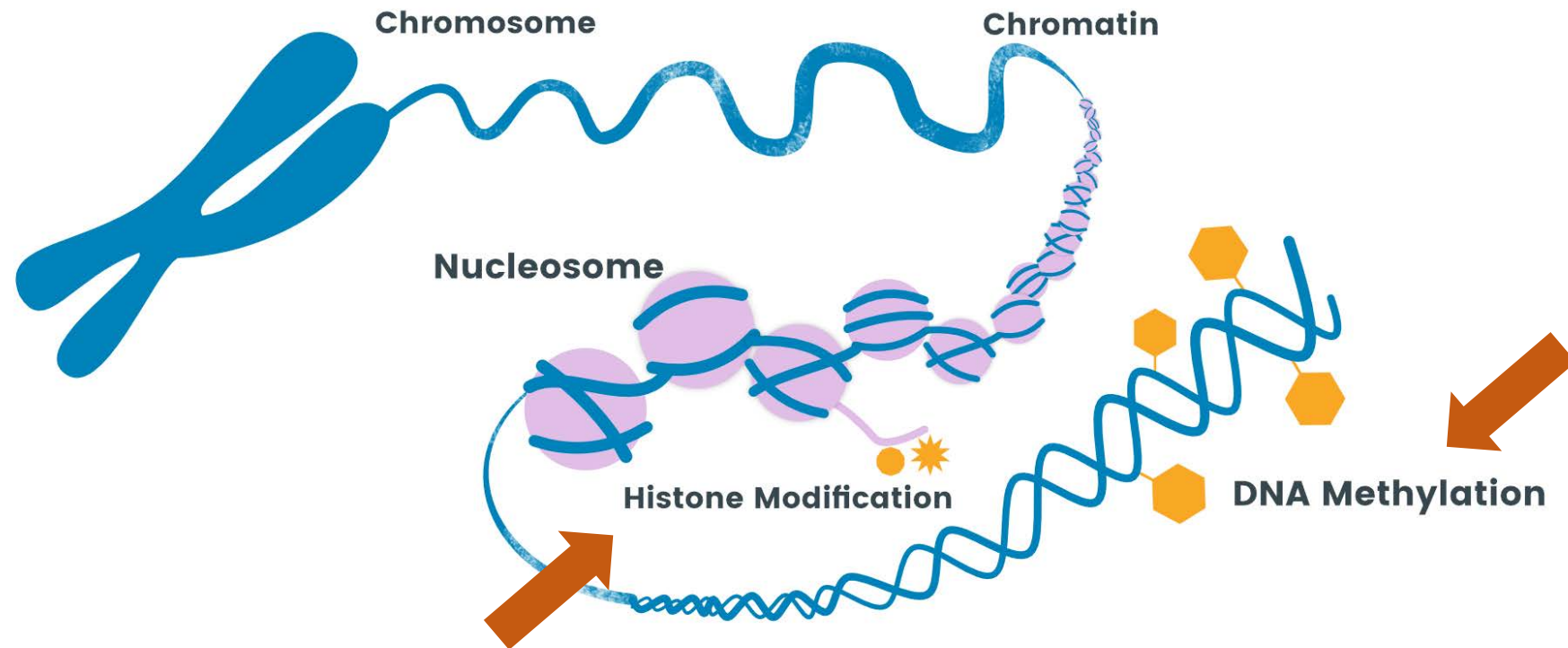
Dr. Ilaria Marinoni

XVI International Symposium GET-NE 2020

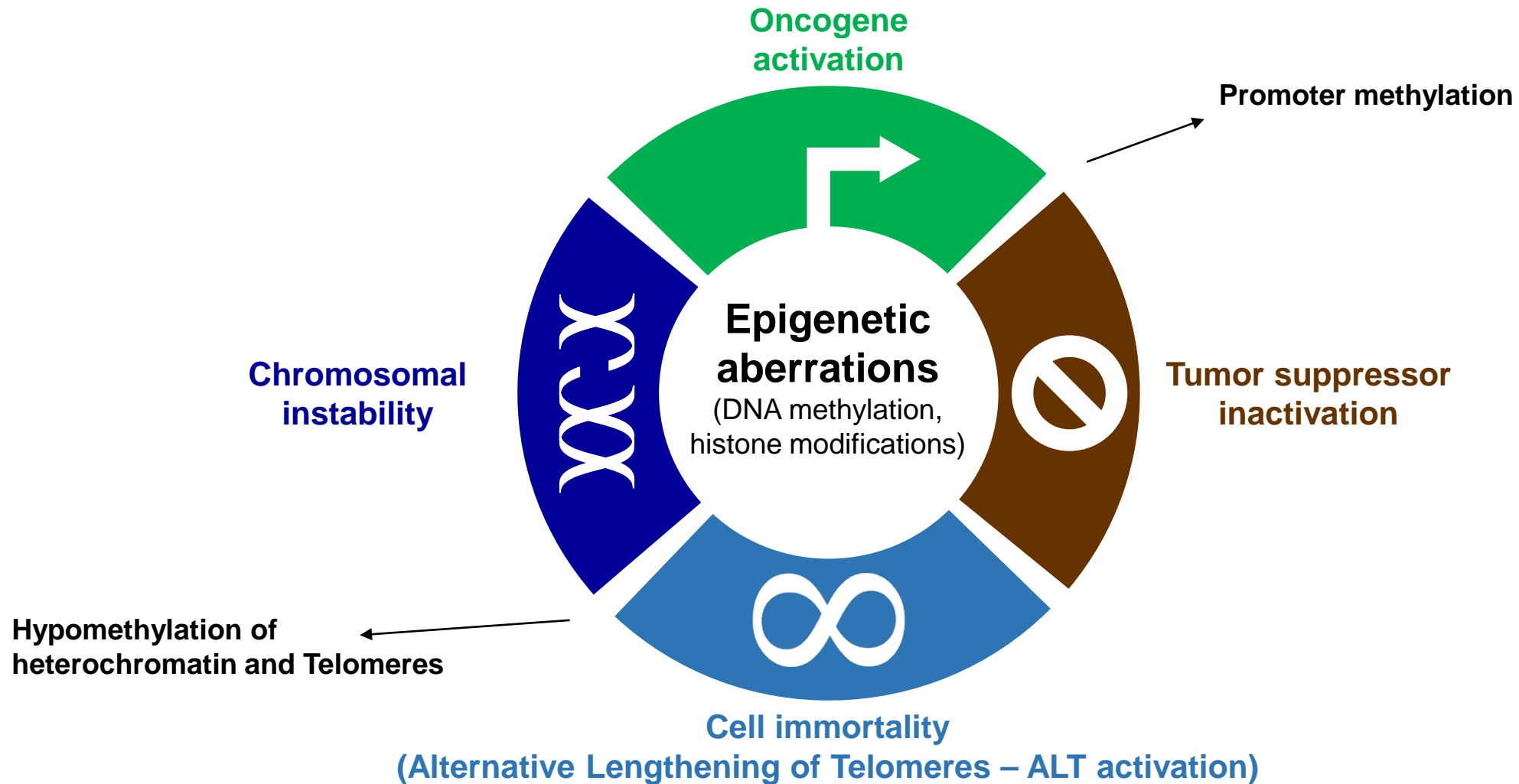
7-9 October 2020

- Literally “upon” or “on top” of genetic
- Modification of DNA which do not alter the sequence but affect chromatin structure affecting gene expression and genomic stability
- Epigenetic changes are heritable and propagated to the daughter cells in divisions

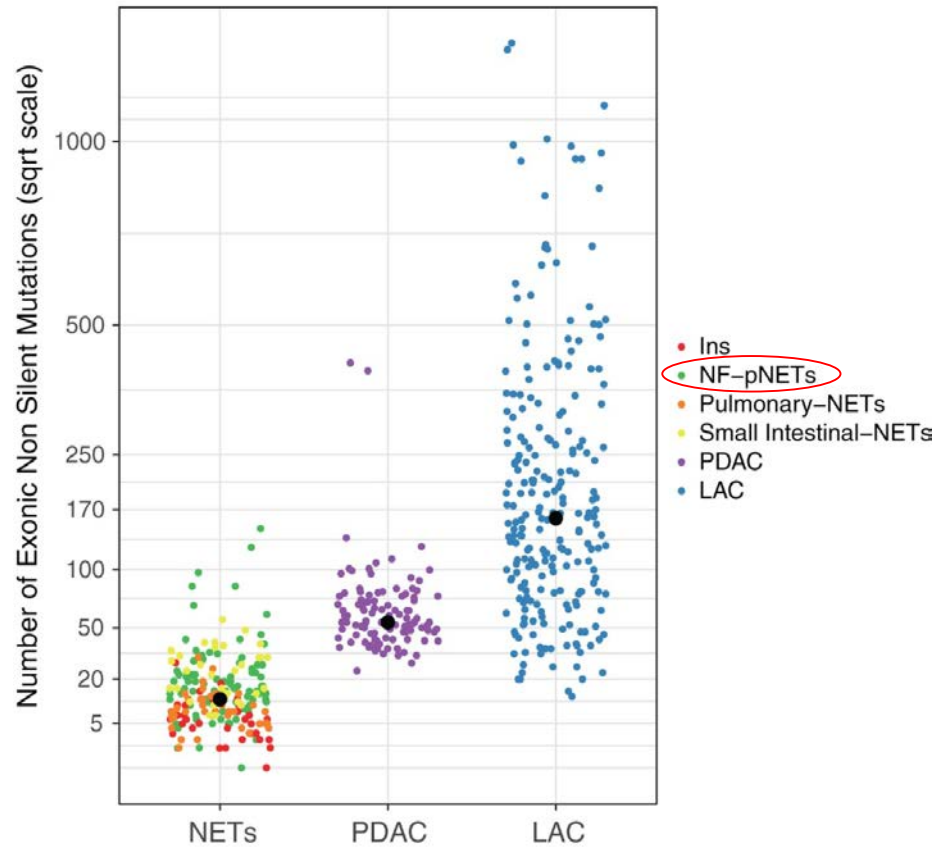
# Epigenome



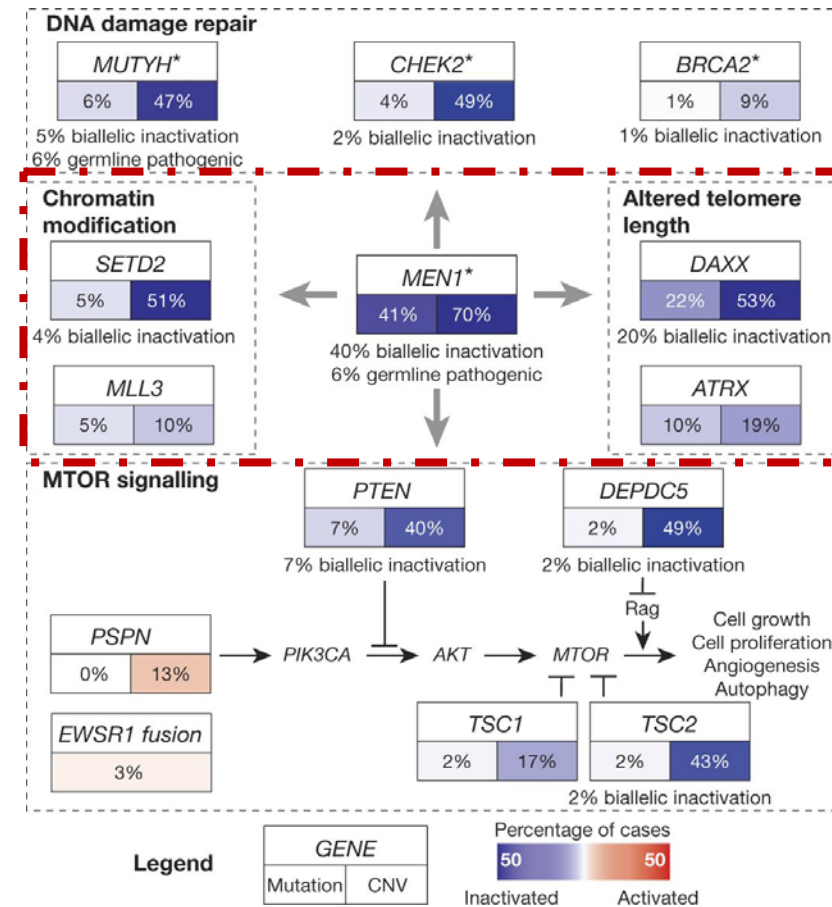
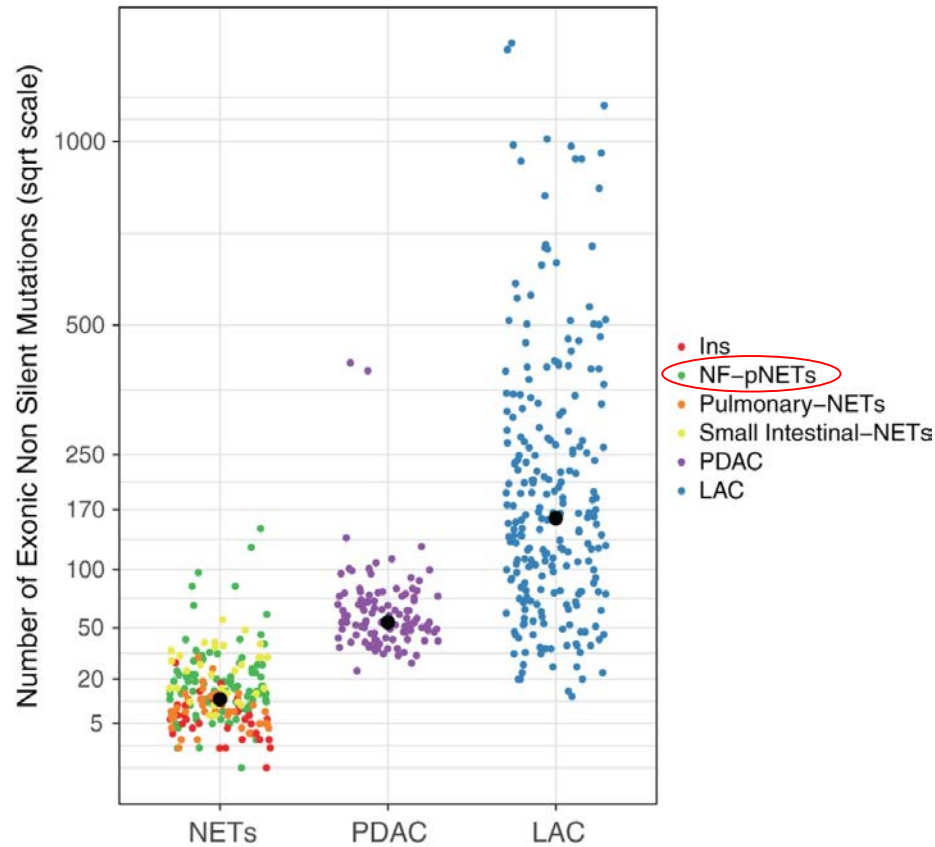
# Epigenetic and Cancer



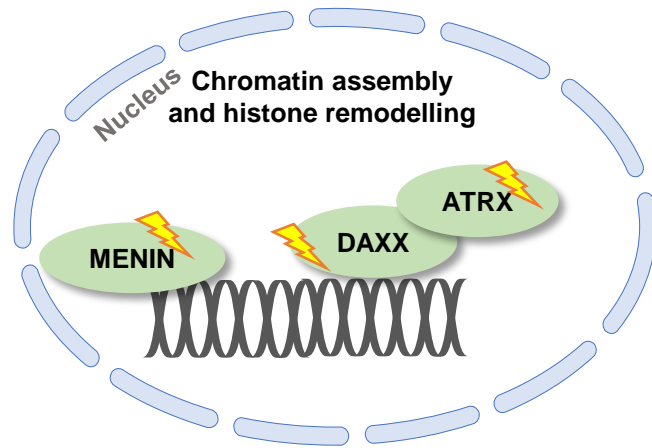
# DNA mutations in PanNETs



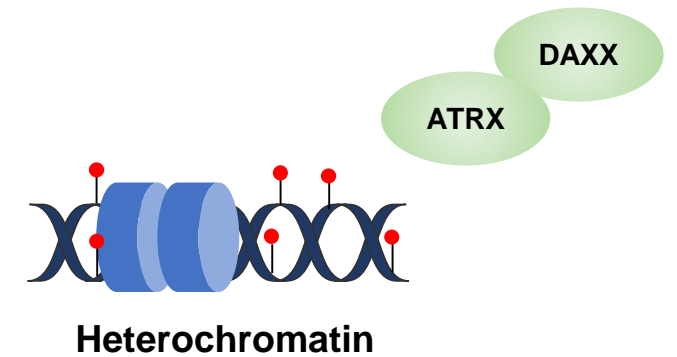
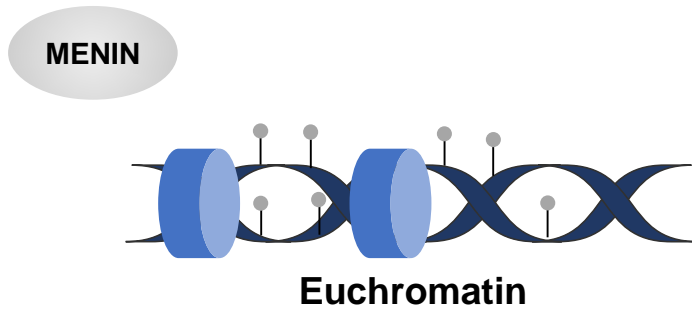
# DNA mutations in PanNETs



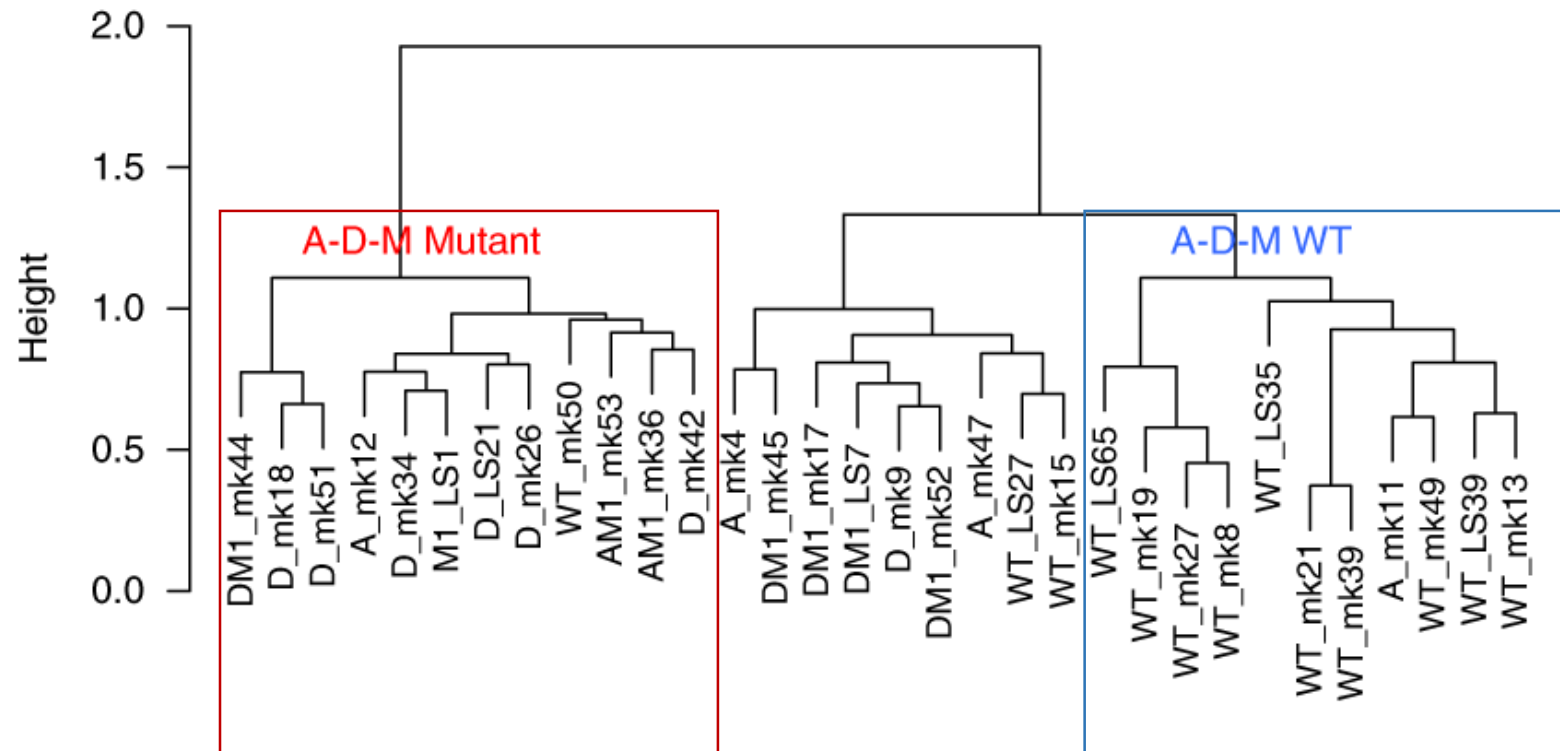
# DAXX/ATRAX and MEN1 functions



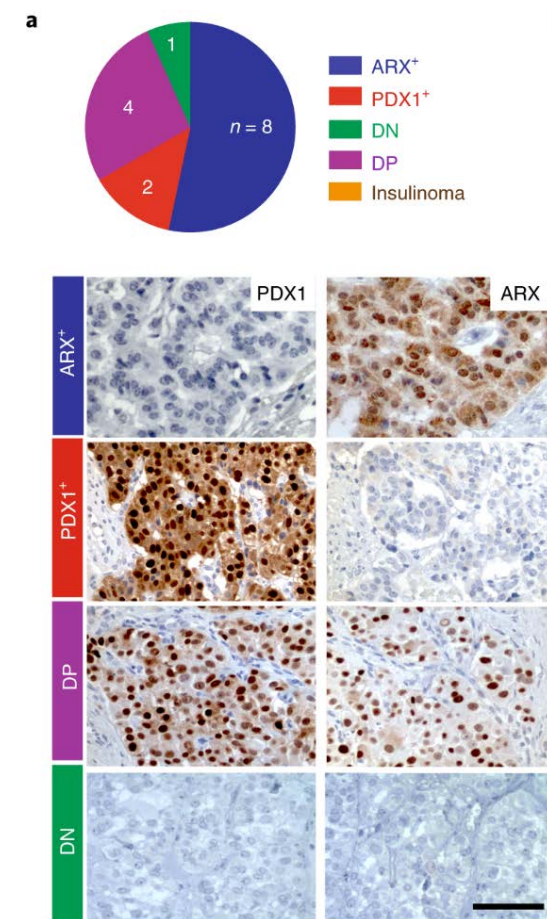
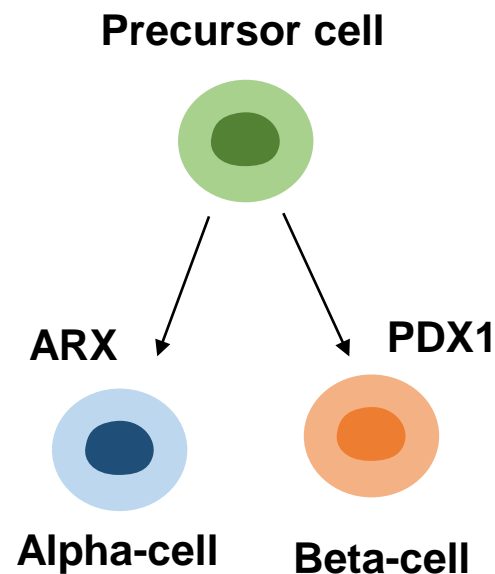
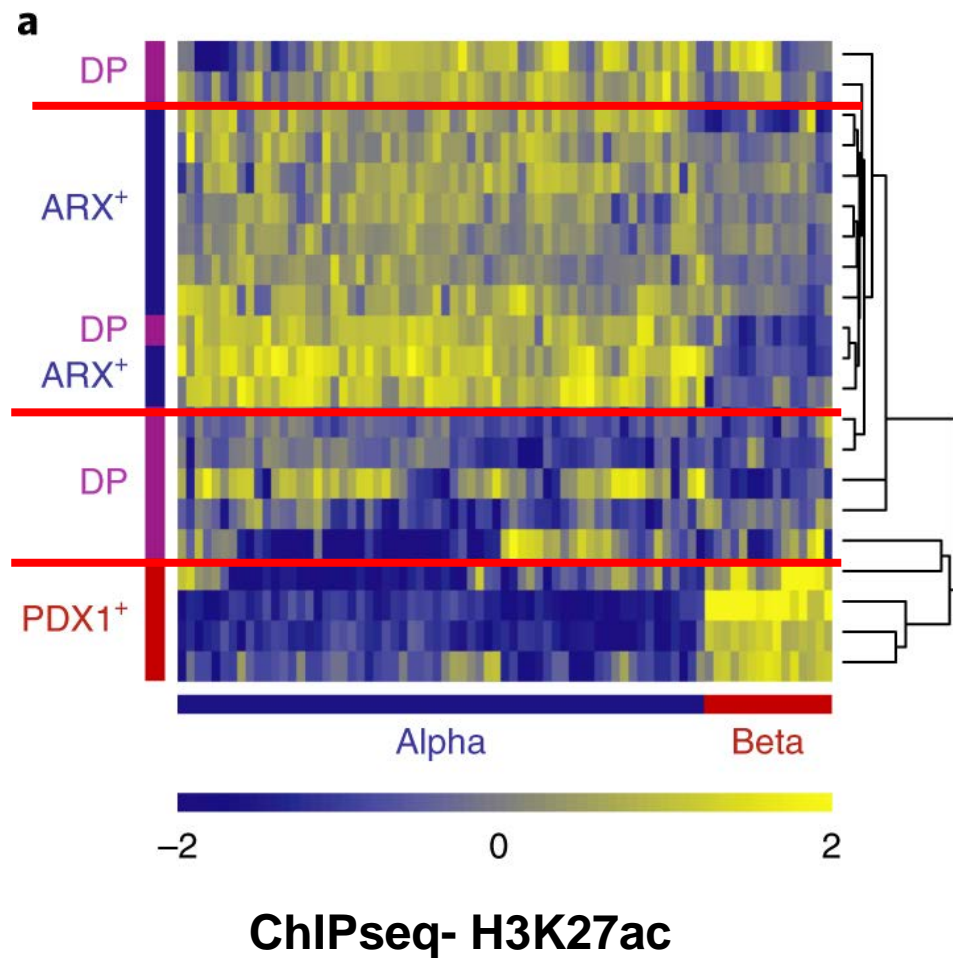
|                   |     |
|-------------------|-----|
| <i>MEN1</i>       | 41% |
| <i>DAXX, ATRX</i> | 32% |



# DAXX/ATRX/MEN1 mutated PanNETs have distinct DNA methylation profiles

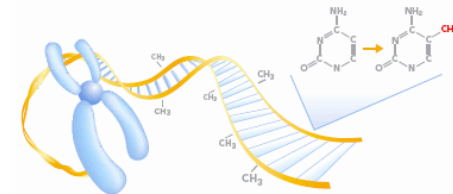
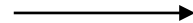


# Distinct PanNETs enhancer profiles

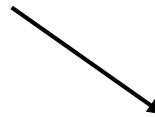
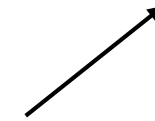


# Investigation of PanNETs methylome: experimental set up

Garvan Institute, Sydney



DNA methylation data  
Illumina – 450K array



Cell of origin

Genetic background

Clinical characteristics

125 primary PanNETs

# Three main PanNETs subtype

Beta cells

Alpha cells

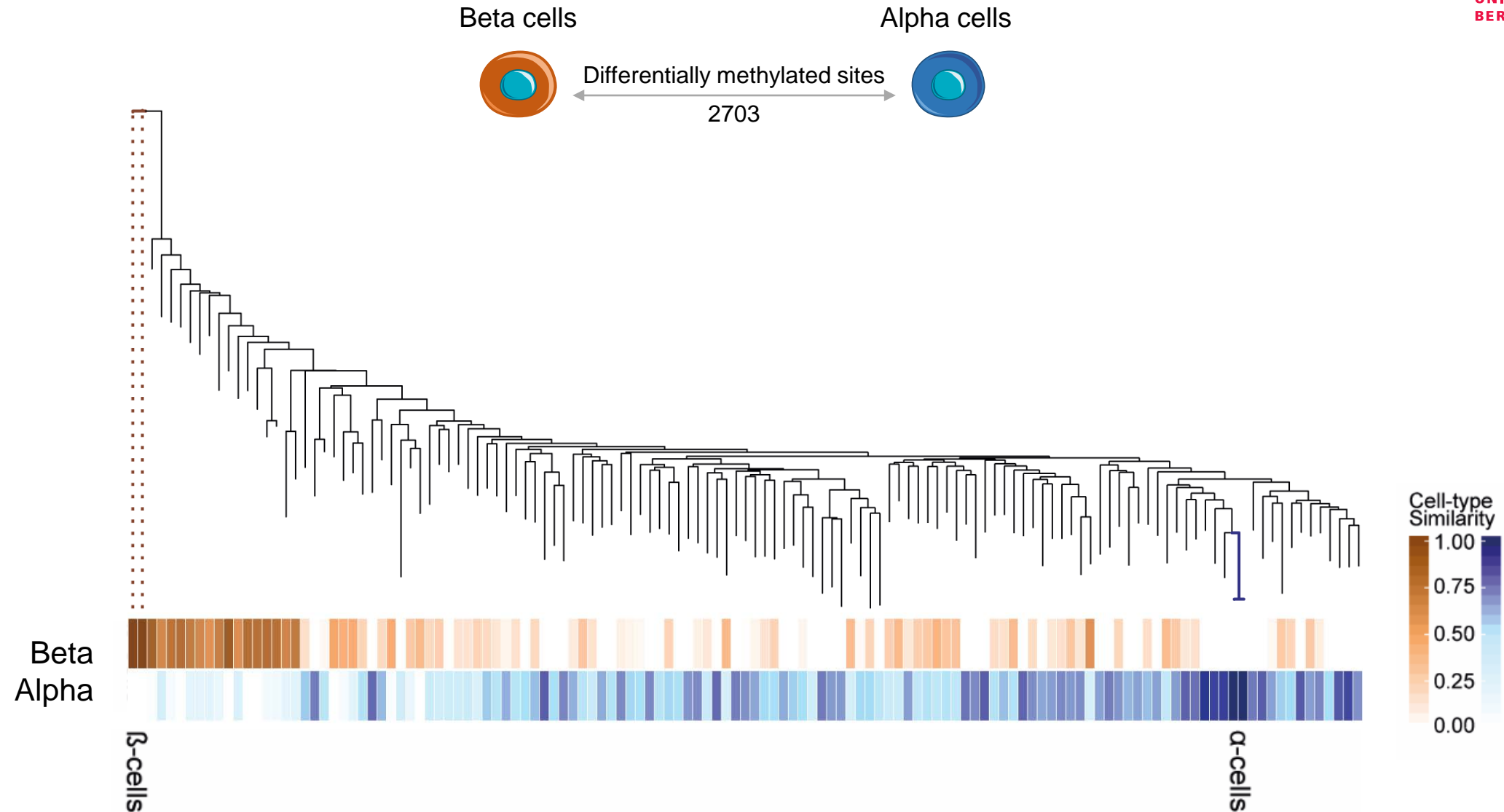


Differentially methylated sites

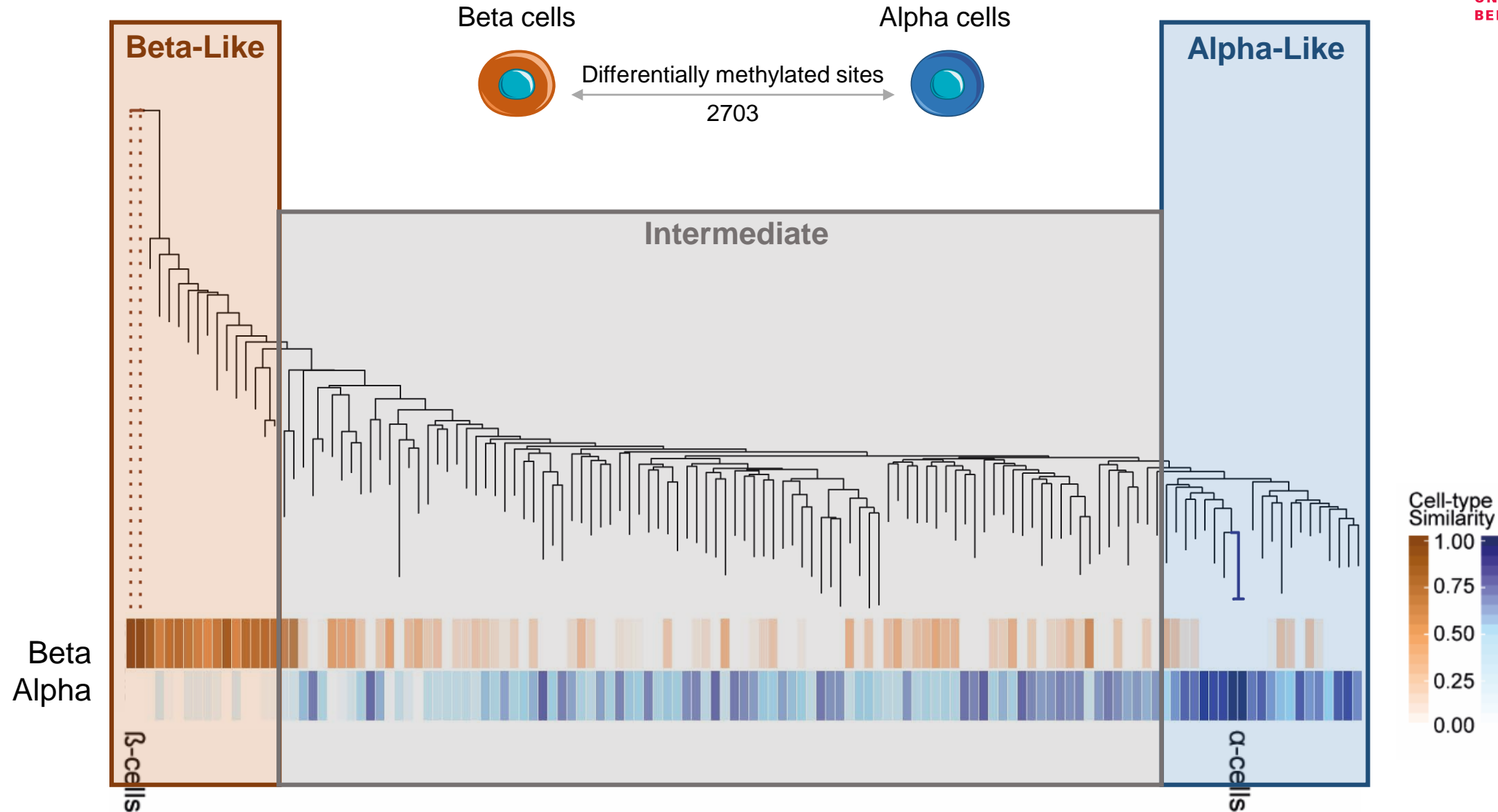
2703



# Three main PanNETs subtype



# Three main PanNETs subtype



# DNA methylation of $\alpha$ - or $\beta$ -specific Transcription factor check points

Analysis of regulatory regions (mainly annotated enhancer regions)

| Alpha        |                |
|--------------|----------------|
| <i>ARX</i>   | <i>MAFB</i>    |
| <i>FEV</i>   | <i>PGR</i>     |
| <i>HMGB3</i> | <i>PTGER3</i>  |
| <i>IRX2</i>  | <i>RFX6</i>    |
| <i>LDB2</i>  | <i>SMARCA1</i> |

| Beta           |             |
|----------------|-------------|
| <i>MAFA</i>    | <i>SIX3</i> |
| <i>PDX1</i>    | <i>SIX2</i> |
| <i>SMAD9</i>   | <i>MNX1</i> |
| <i>CDKN1C</i>  | <i>BMP5</i> |
| <i>TFCP2L1</i> | <i>PIR</i>  |

# DNA methylation of $\alpha$ - or $\beta$ -specific Transcription factor check points

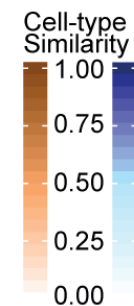
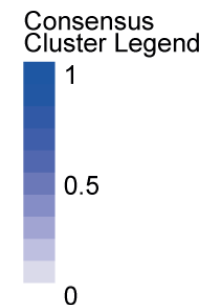
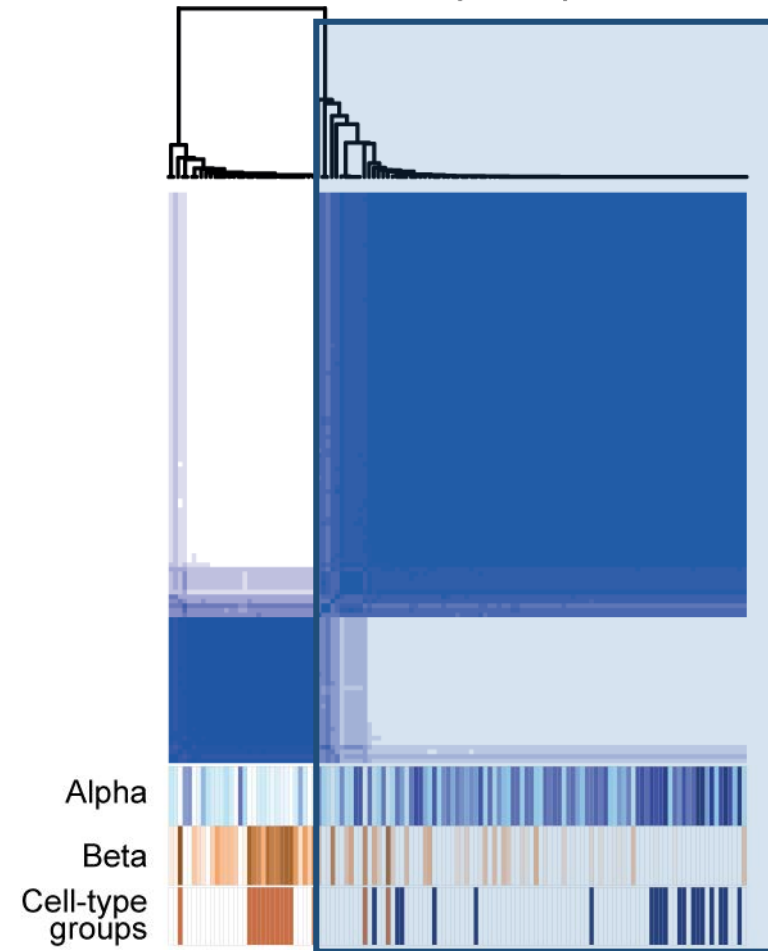
Analysis of regulatory regions (mainly annotated enhancer regions)

consensus matrix Alpha specific TFs k=2

| Alpha |         |
|-------|---------|
| ARX   | MAFB    |
| FEV   | PGR     |
| HMGB3 | PTGER3  |
| IRX2  | RFX6    |
| LDB2  | SMARCA1 |

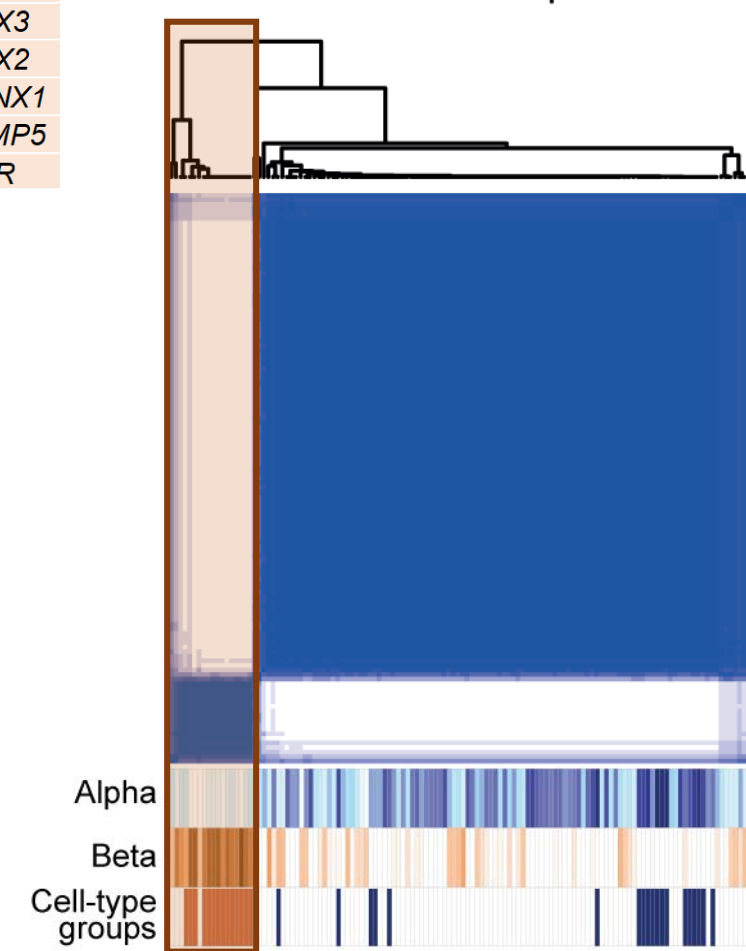
| Beta    |      |
|---------|------|
| MAFA    | SIX3 |
| PDX1    | SIX2 |
| SMAD9   | MNX1 |
| CDKN1C  | BMP5 |
| TFCP2L1 | PIR  |

consensus matrix Beta specific TFs k=2

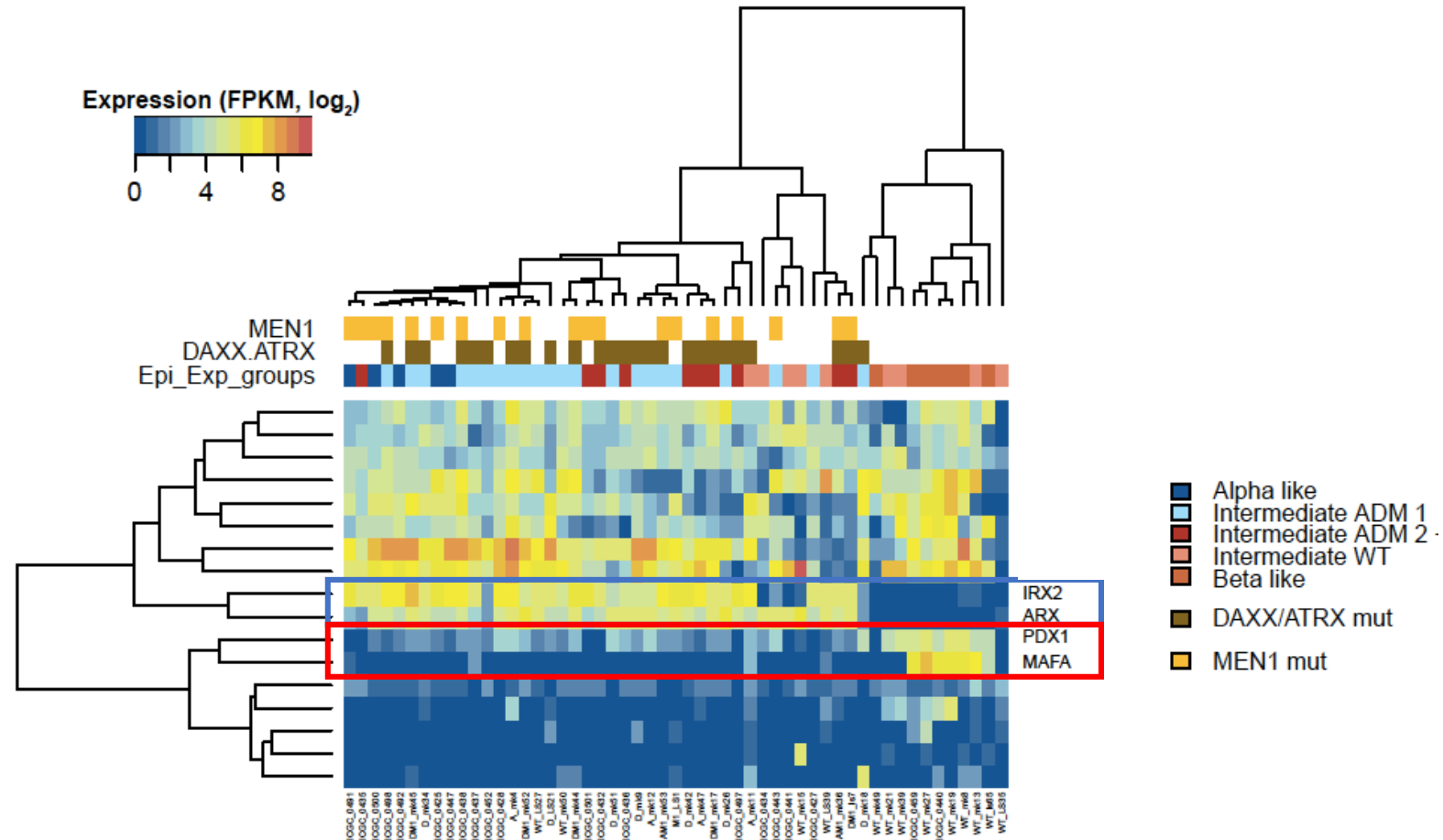


Cell-type Tumour groups

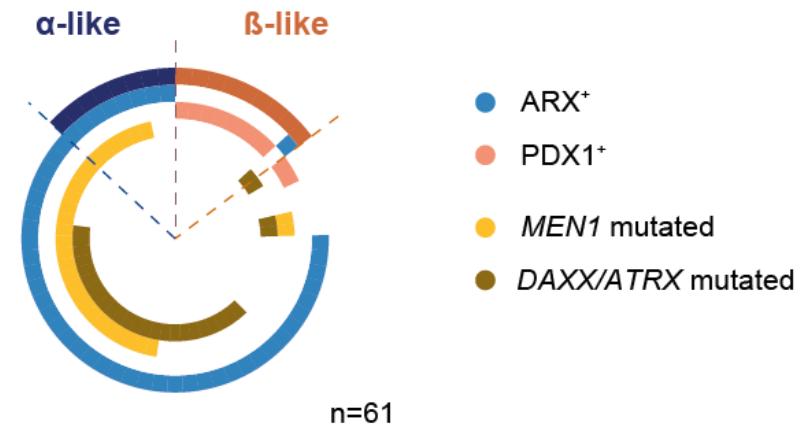
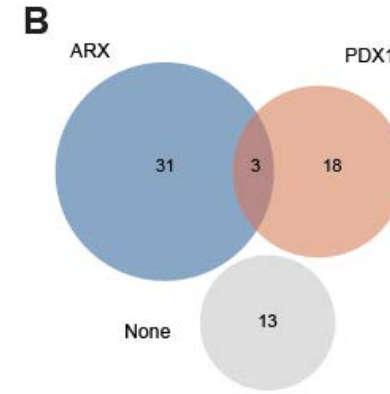
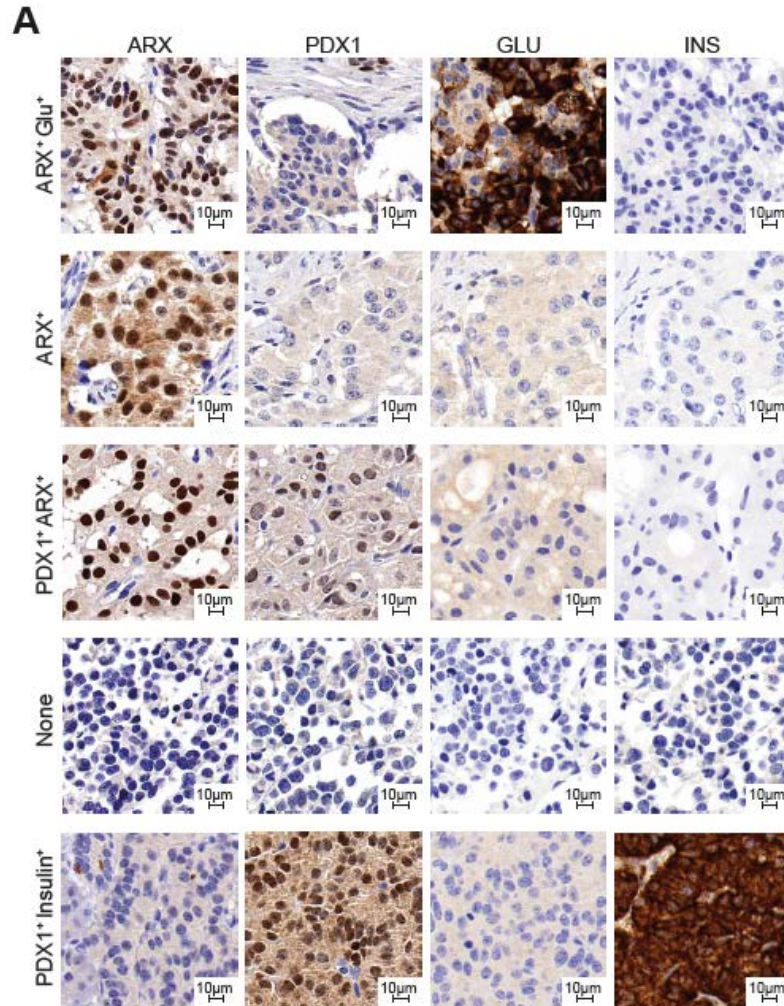
- Alpha Like
- Beta Like



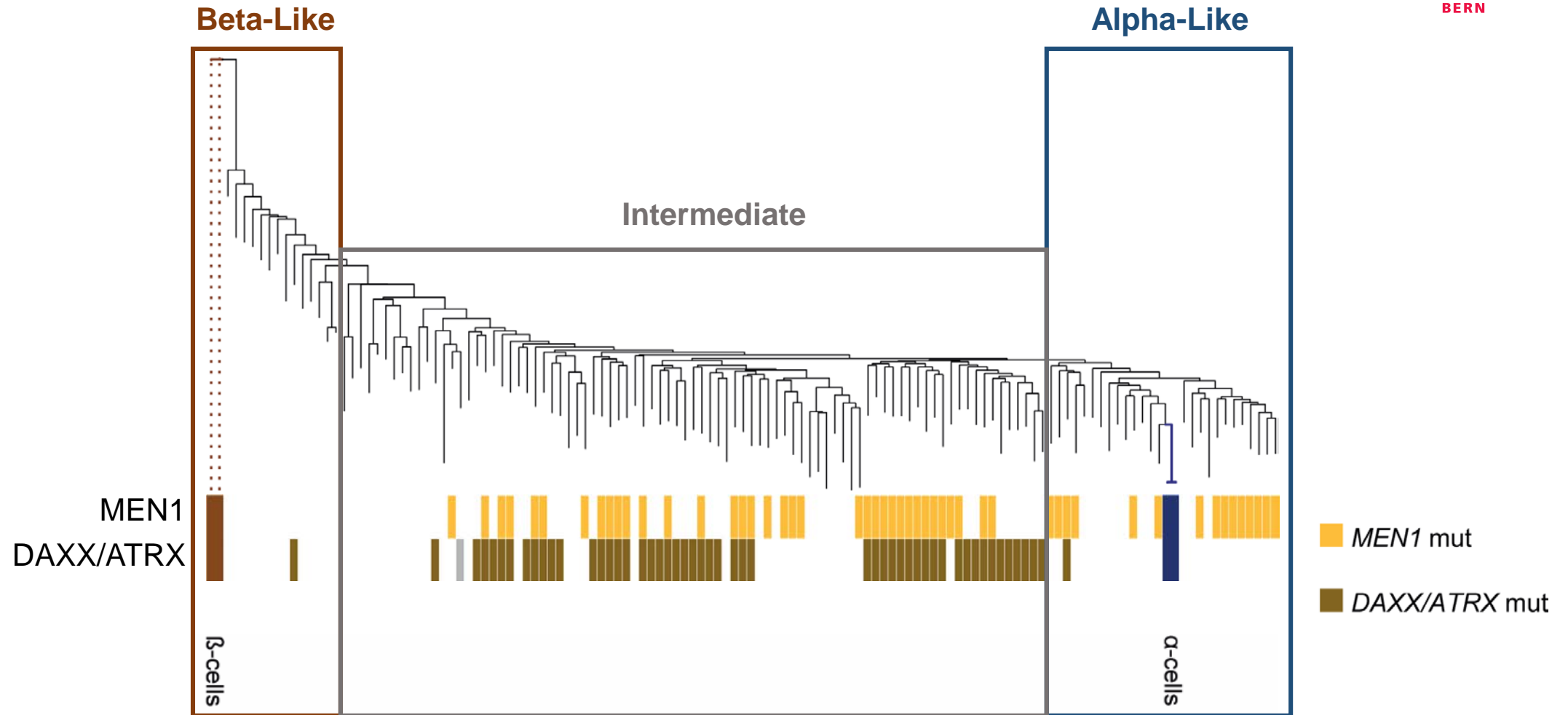
# Expression of TF



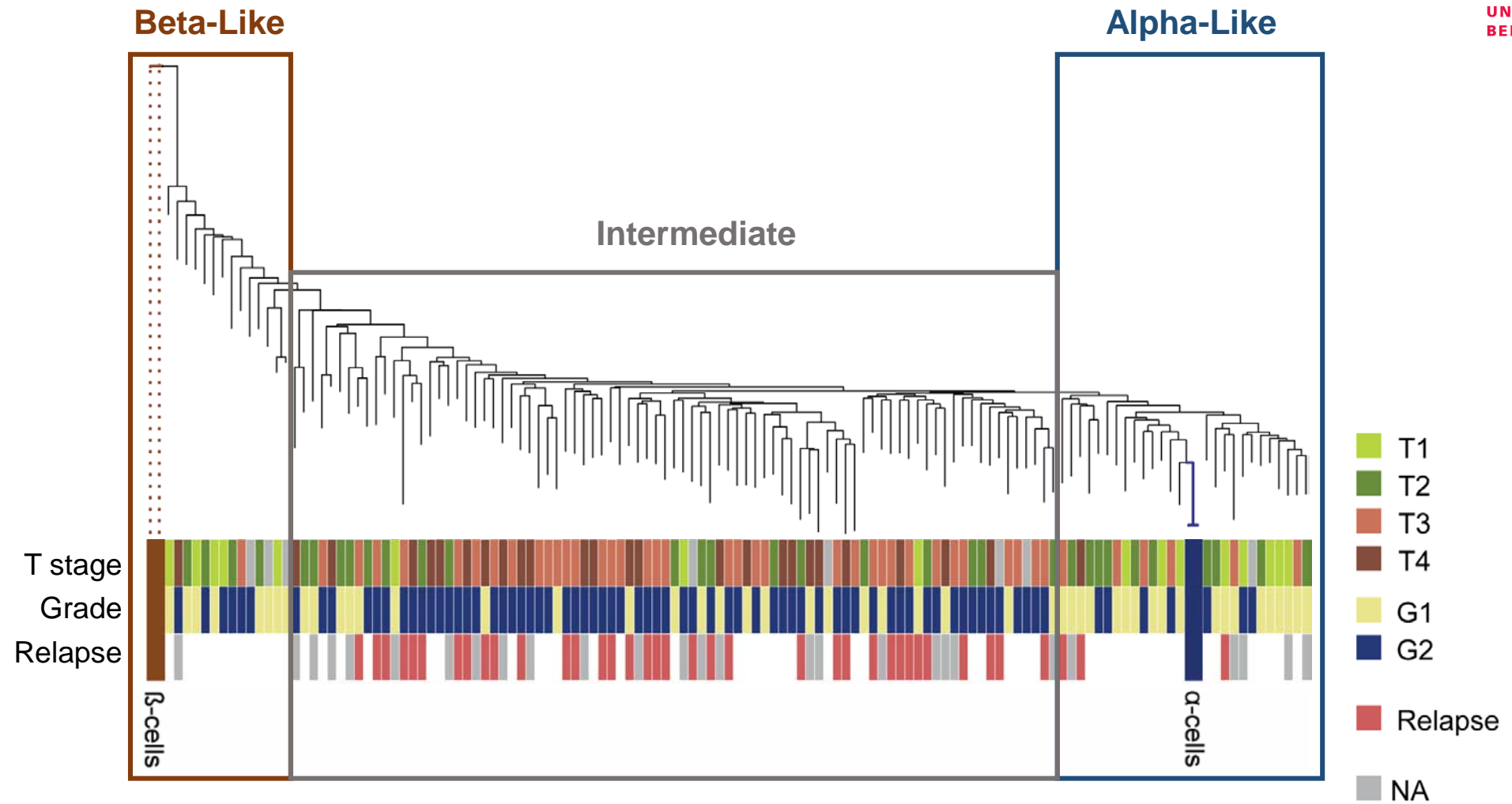
# Expression of TF and hormones confirm alpha and beta cell origin



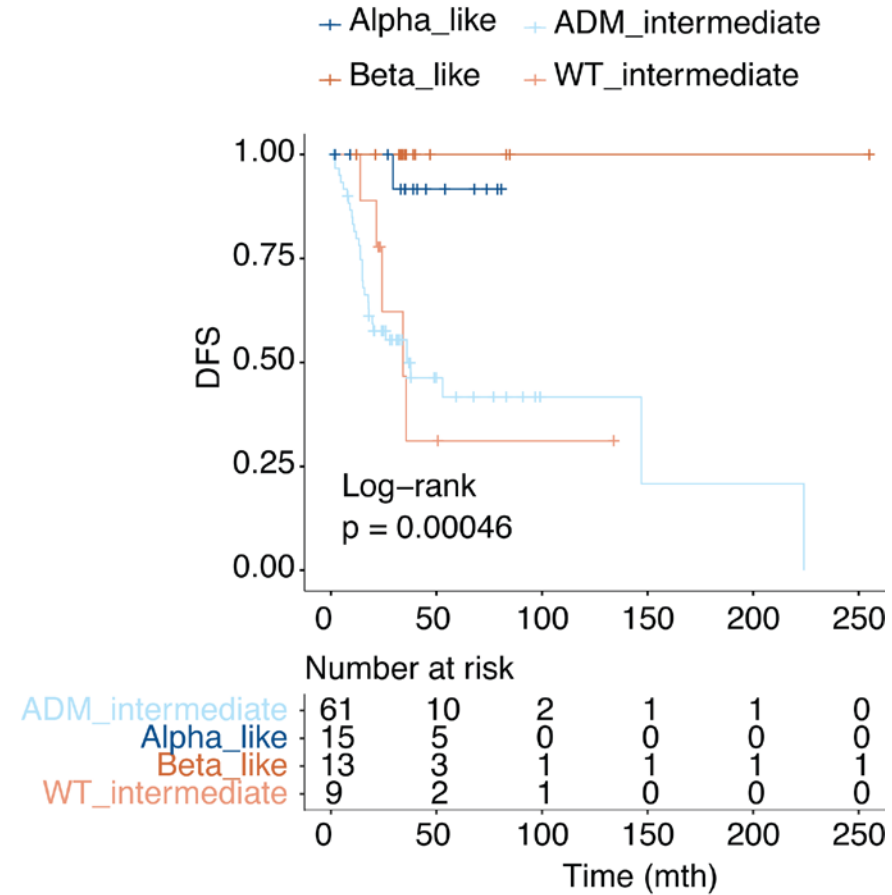
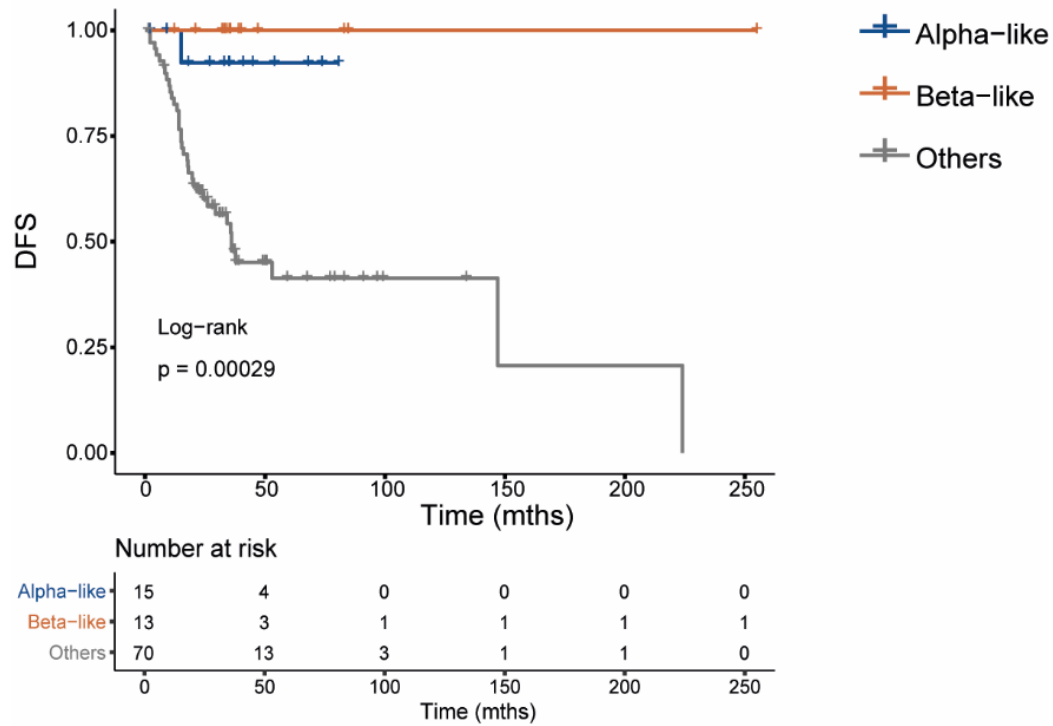
# Epigenetic subgroups of PanNET have distinct genetic background



# Epigenetic subgroups of PanNET have distinct clinical features



# Intermediate PanNETs have shorter DFS



# Model of PanNET epigenetic progression



$\beta$ -cell

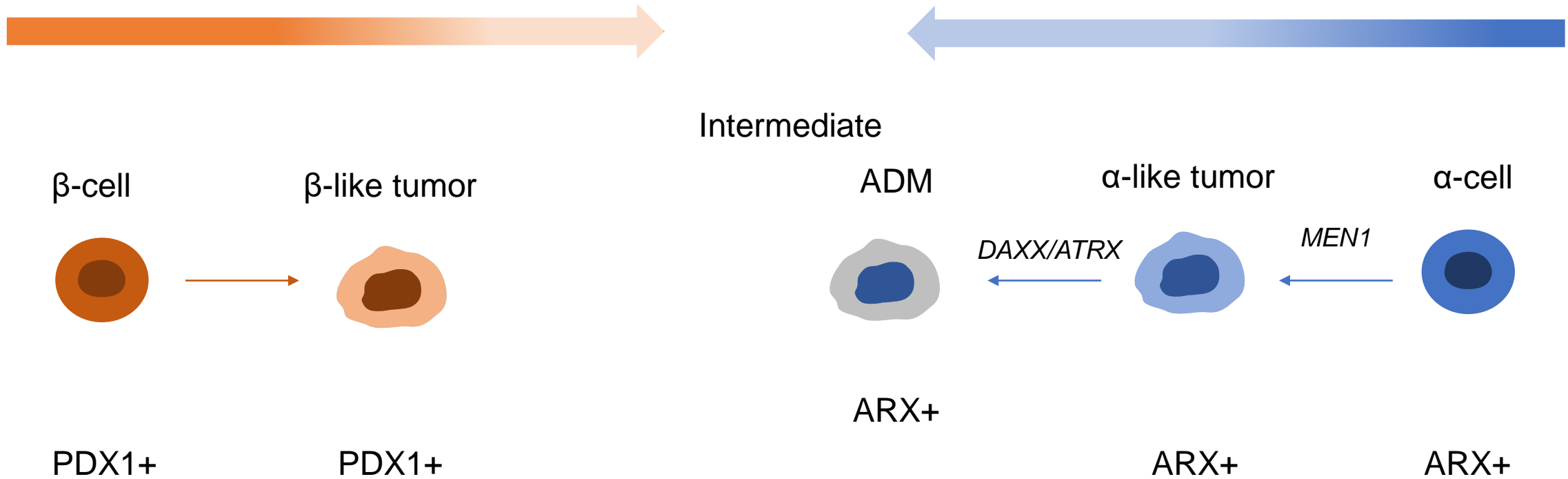
$\beta$ -like tumor



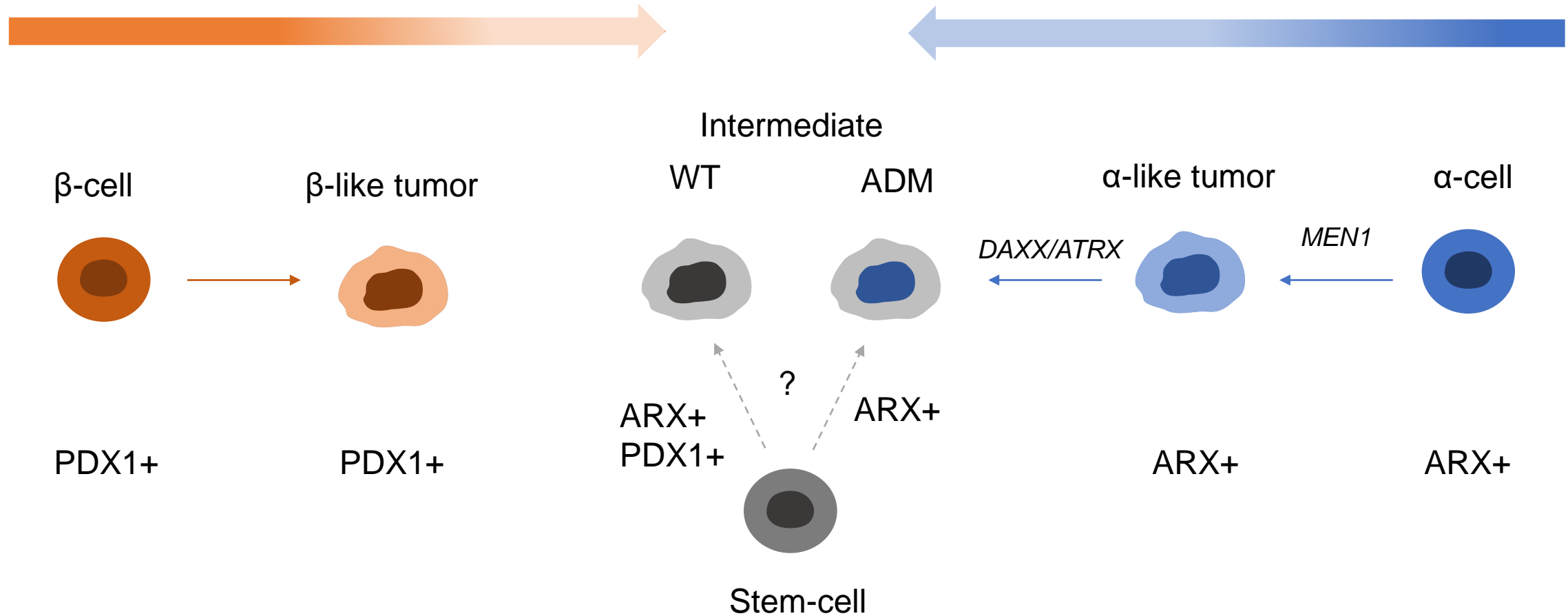
PDX1+

PDX1+

# Model of PanNET epigenetic progression



# Model of PanNET epigenetic progression

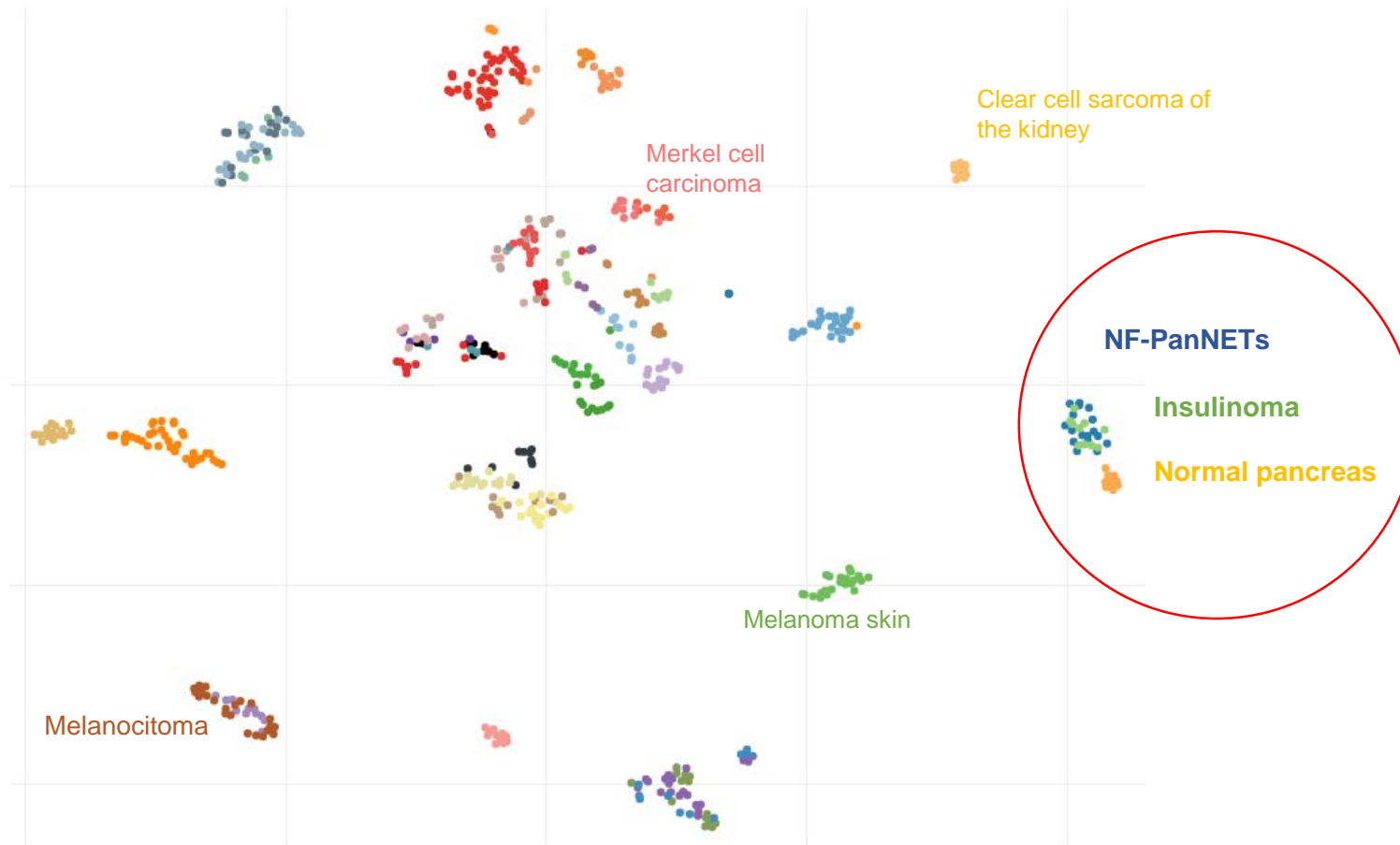


1. PanNET methylomes reveals at least 2 cells of origin for PanNETs
2. Cell-type specific epigenetic subtypes present different mutational landscape
3. We were able to identify a DNA methylation signature that defines clinically and molecularly different PanNETs

**DNA methylation is very stable in paraffin fixed samples hence it can be analysed in clinical samples and could be use for PanNET classification**

# DNA methylation for tumor classification

tSNE



# Possible future clinical application

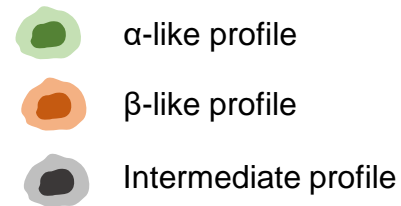
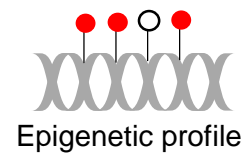
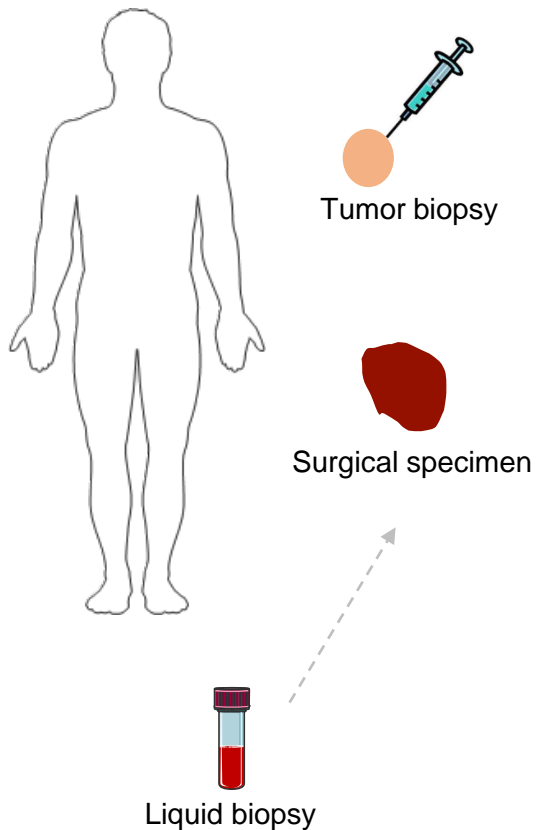
## Starting patient material

## DNA methylation

## Classification

## Future clinical application

### PanNET



- Disease Prognosis**
- Therapy Prediction**
- CUP identification**

# Acknowledgement

## Institute of Pathology, University of Bern Endocrine Pathology group

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