



# R2 Genomics Analysis & Visualization platform

Illustration of a multi omics journey  
into the unraveling of cancer biology



Jan Koster PhD.

Department of OncoGenomics

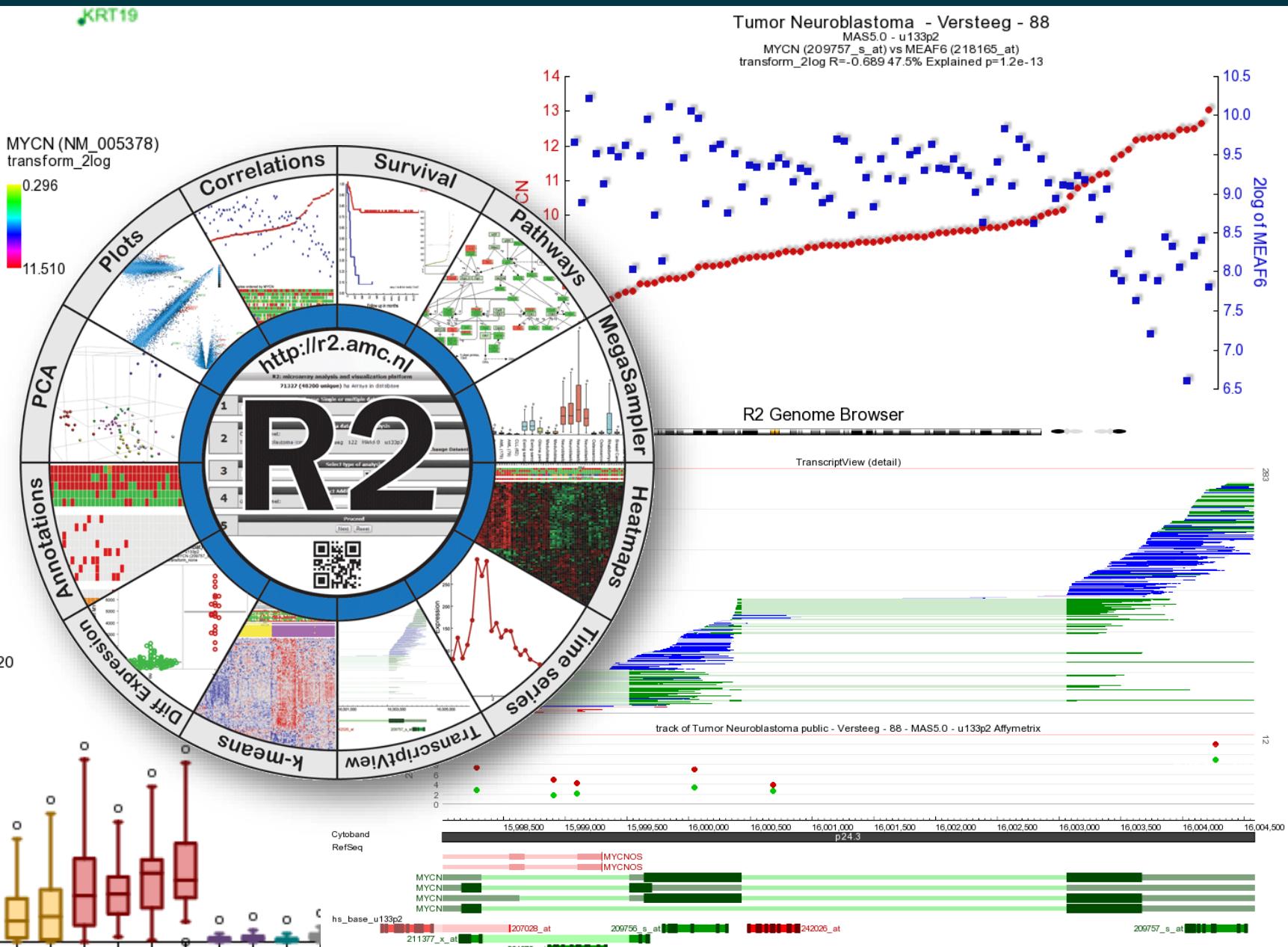
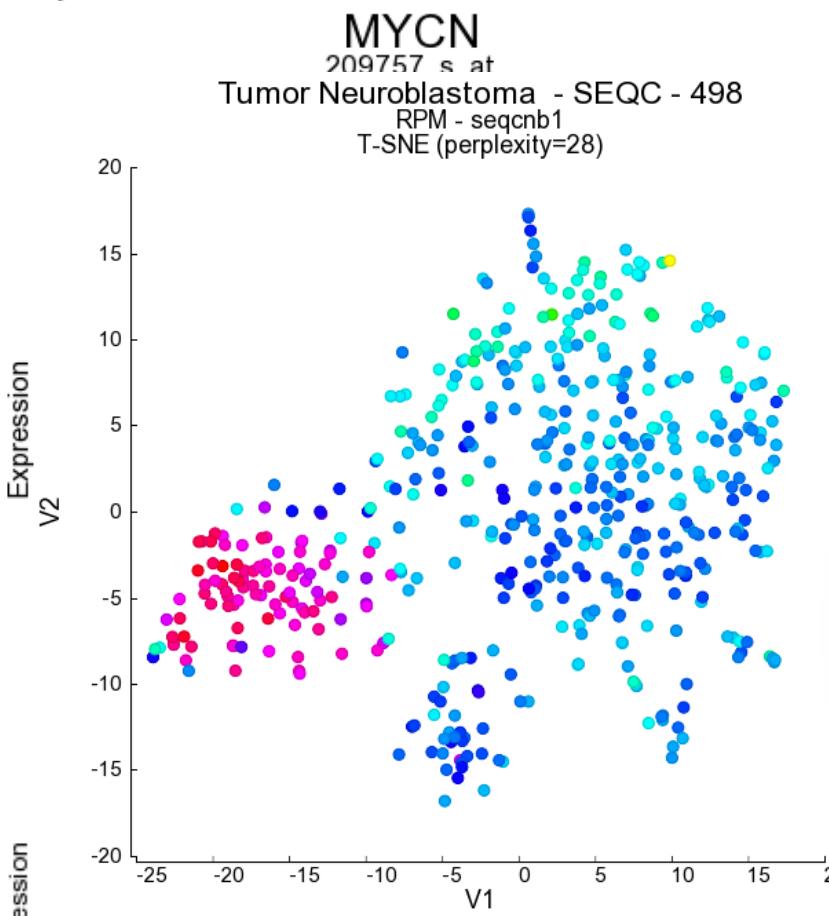
Amsterdam University Medical Centers (AUMC)

Location AMC

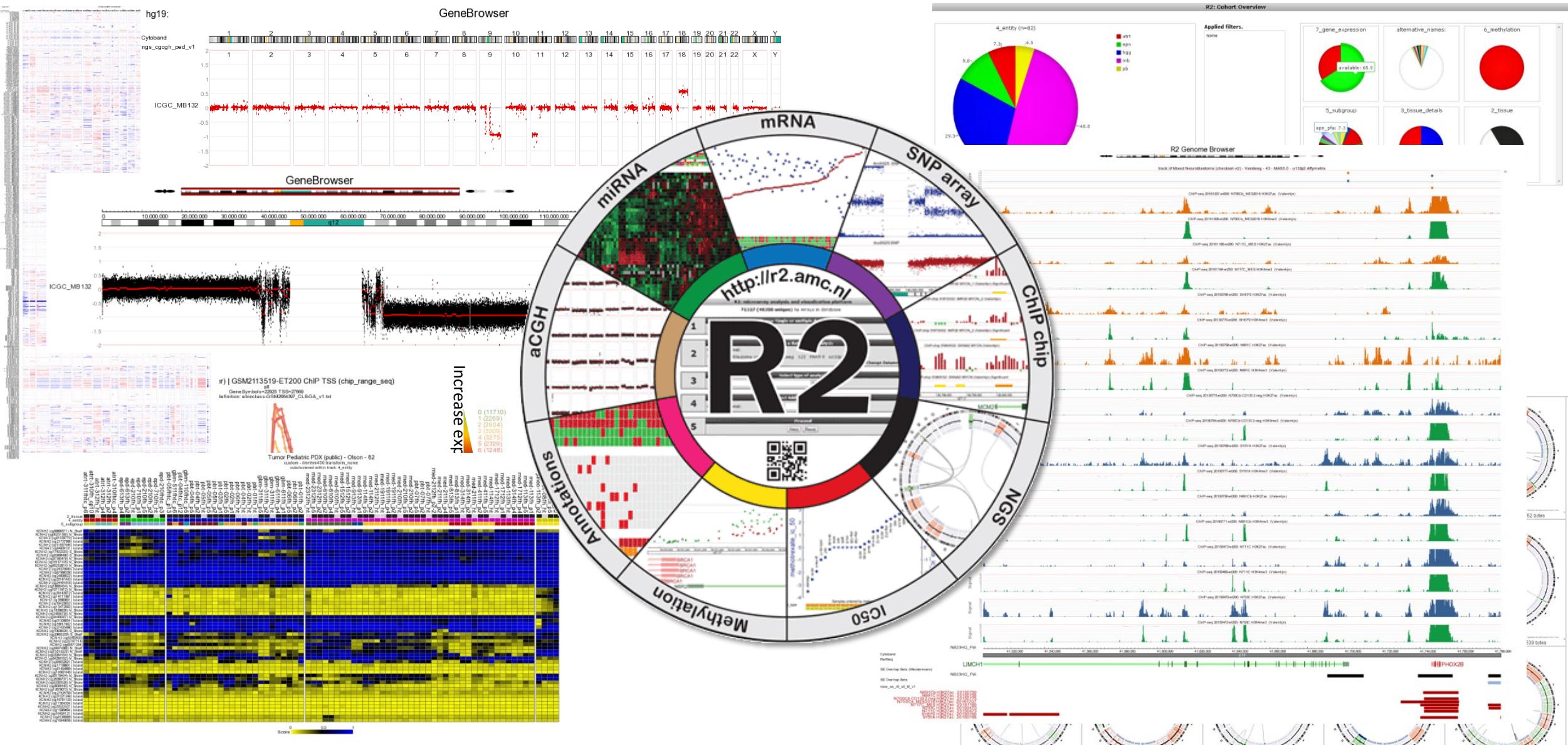
University of Amsterdam, the Netherlands

jankoster@amsterdamumc.nl

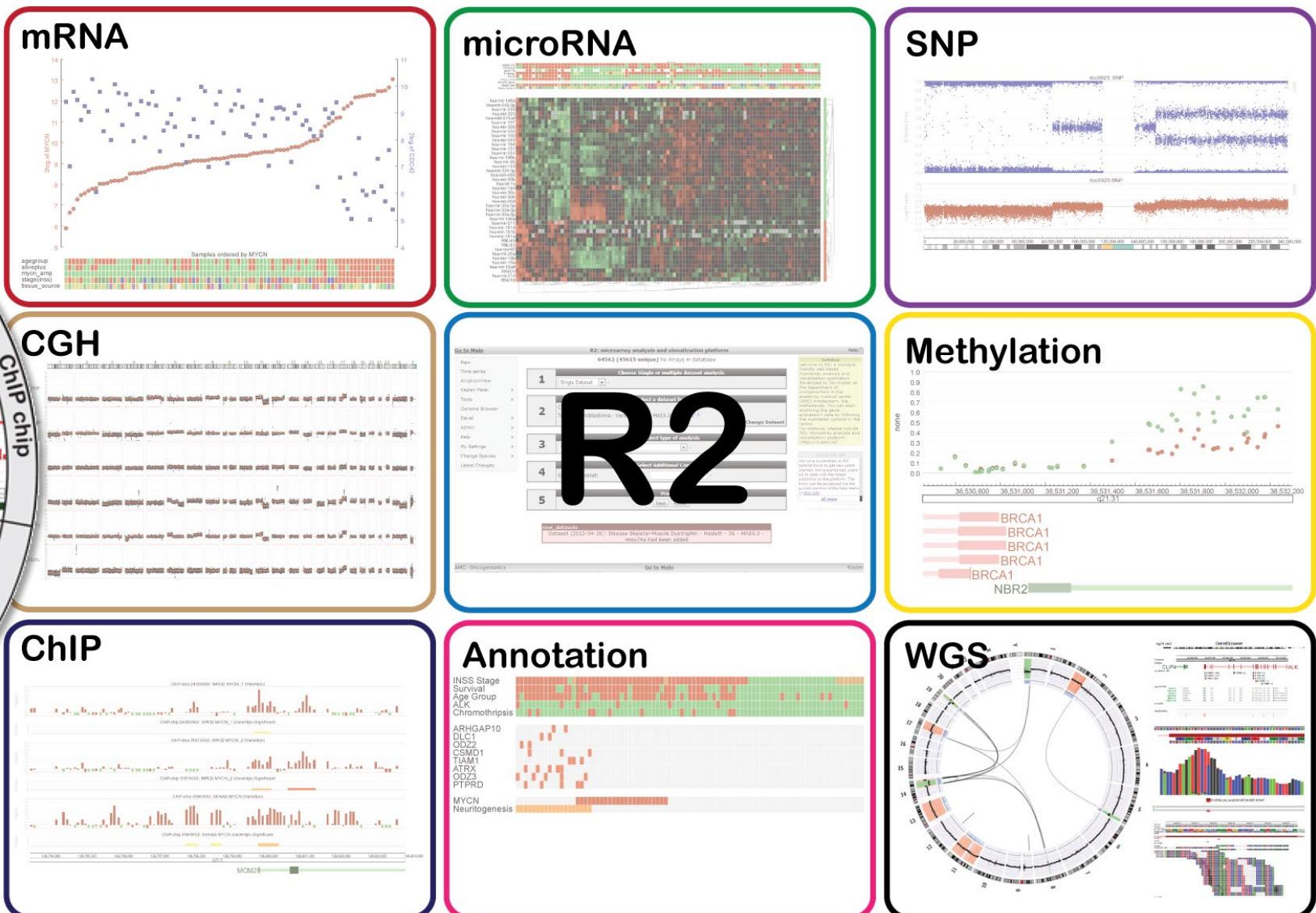
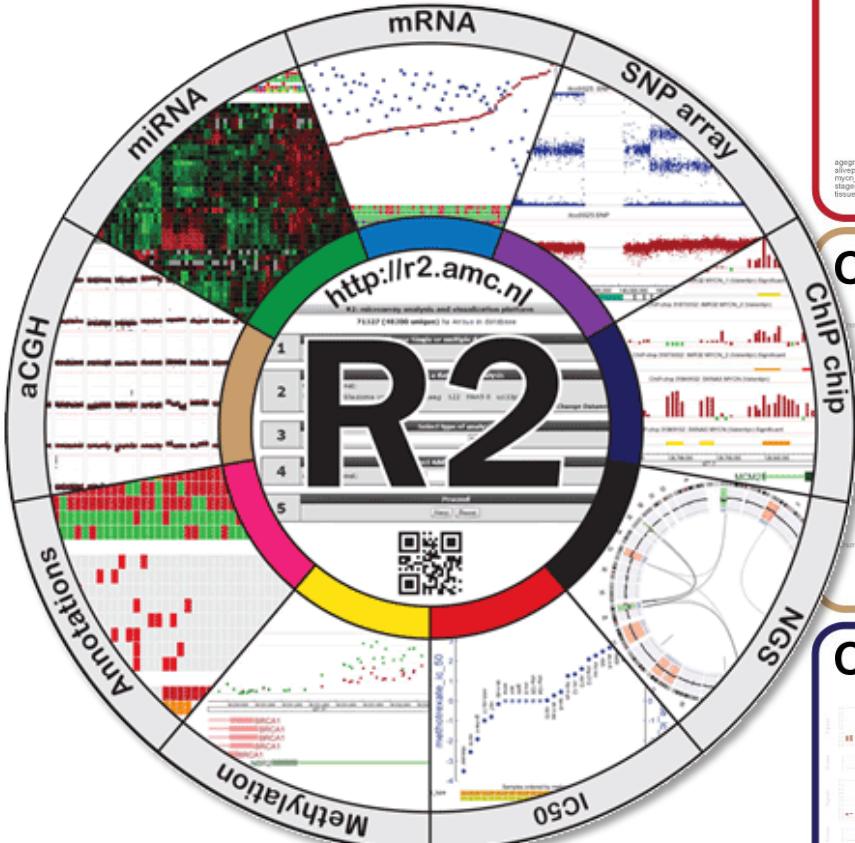
# The R2 platform



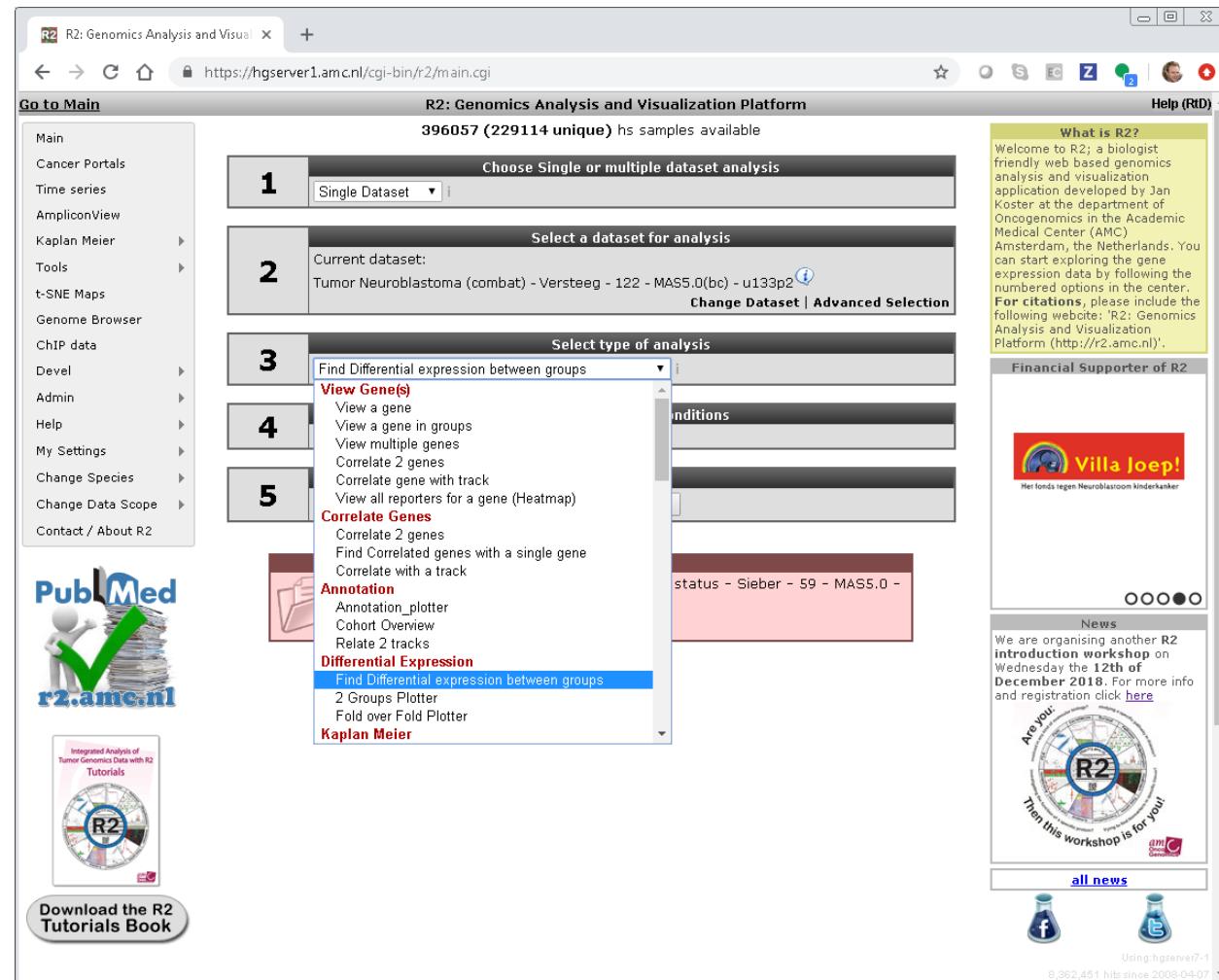
# The R2 platform



# Different types of Omics data available in the R2 Platform



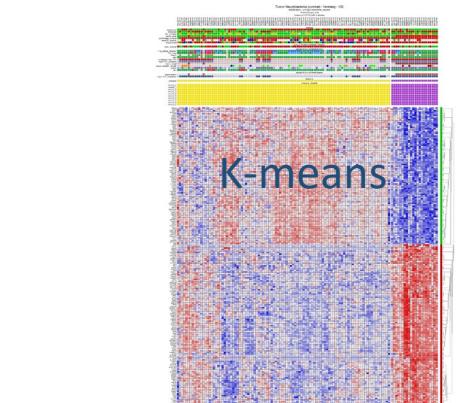
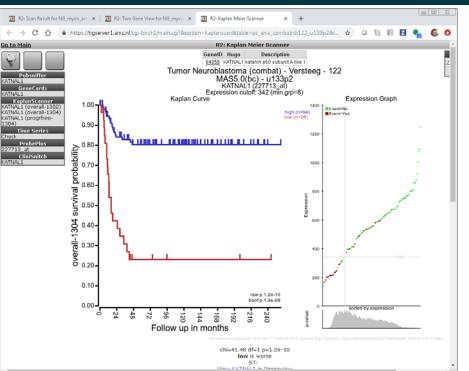
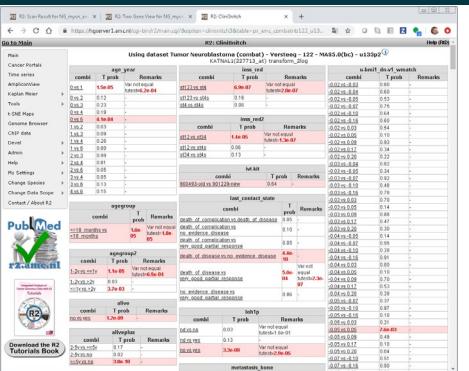
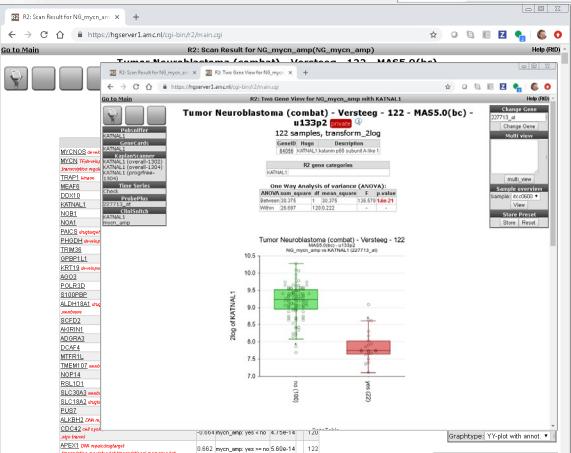
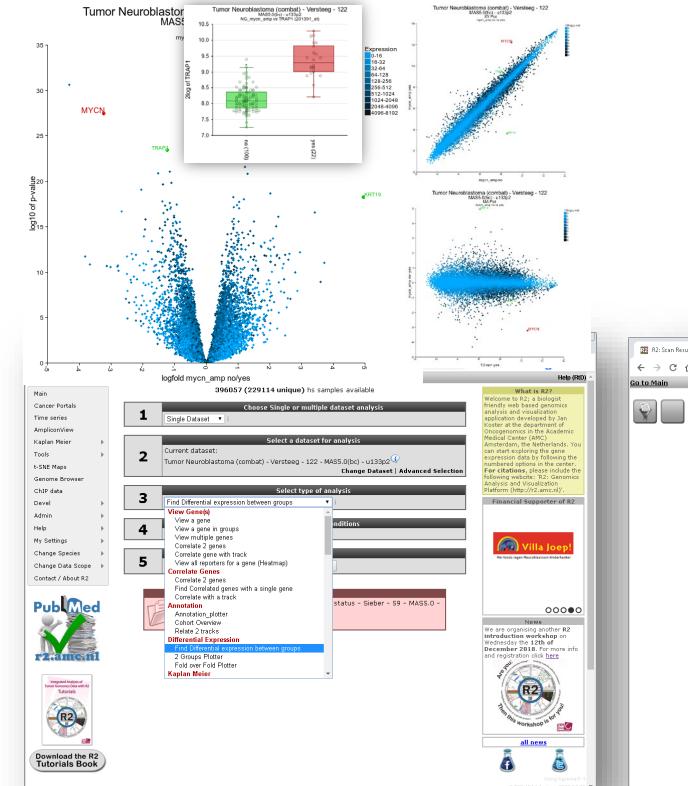
# Example: Differential Expression Analysis



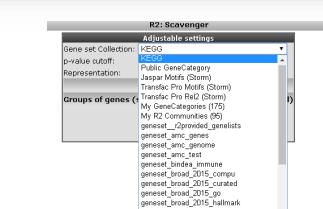
The screenshot shows the R2: Genomics Analysis and Visualization Platform interface. The main window is titled "R2: Genomics Analysis and Visualization Platform" and displays "396057 (229114 unique) hs samples available". The interface is divided into several sections:

- Left Sidebar:** "Go to Main" menu with options like Main, Cancer Portals, Time series, AmpliconView, Kaplan Meier, Tools, t-SNE Maps, Genome Browser, ChIP data, Devel, Admin, Help, My Settings, Change Species, Change Data Scope, and Contact / About R2.
- Top Bar:** "Choose Single or multiple dataset analysis" dropdown set to "Single Dataset".
- Dataset Selection:** "Select a dataset for analysis" section showing "Current dataset: Tumor Neuroblastoma (combat) - Versteeg - 122 - MASS5.0(bc) - u133p2". Buttons for "Change Dataset" and "Advanced Selection".
- Analysis Options:** "Select type of analysis" dropdown menu:
  - View Gene(s):** View a gene, View a gene in groups, View multiple genes, Correlate 2 genes, Correlate gene with track, View all reporters for a gene (Heatmap)
  - Correlate Genes:** Correlate 2 genes, Find Correlated genes with a single gene, Correlate with a track
  - Annotation:** Annotation\_plotter, Cohort Overview, Relate 2 tracks
  - Differential Expression:** Find Differential expression between groups, 2 Groups Plotter, Fold over Fold Plotter
  - Kaplan Meier:**
- Right Sidebar:**
  - What is R2?**: A yellow box with text about the platform, mentioning it's a biologist friendly web-based genomics analysis and visualization application developed by Jan Koster at the department of Oncogenomics in the Academic Medical Center (AMC) Amsterdam, the Netherlands. It includes a link for citations: "For citations, please include the following website: 'R2: Genomics Analysis and Visualization Platform (<http://r2.amc.nl>)'".
  - Financial Supporter of R2:** Shows a logo for "Villa Joep!" and text: "Met fonds tegen Neuroblastoom kinderanker".
  - News:** A box with text: "We are organising another R2 introduction workshop on Wednesday the 12th of December 2018. For more info and registration click [here](#)". It also features a circular graphic with the text "Are you: ... Then this workshop is for you! R2".
  - Social Media:** Icons for Facebook and Twitter.
  - Footer:** "Using hgserver7-1", "8,362,451 hits since 2008-04-07", and a "all news" link.

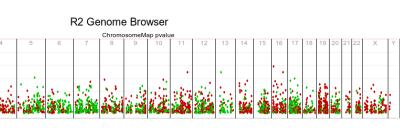
# Example: Differential Expression Analysis



## Gene Set Analyses



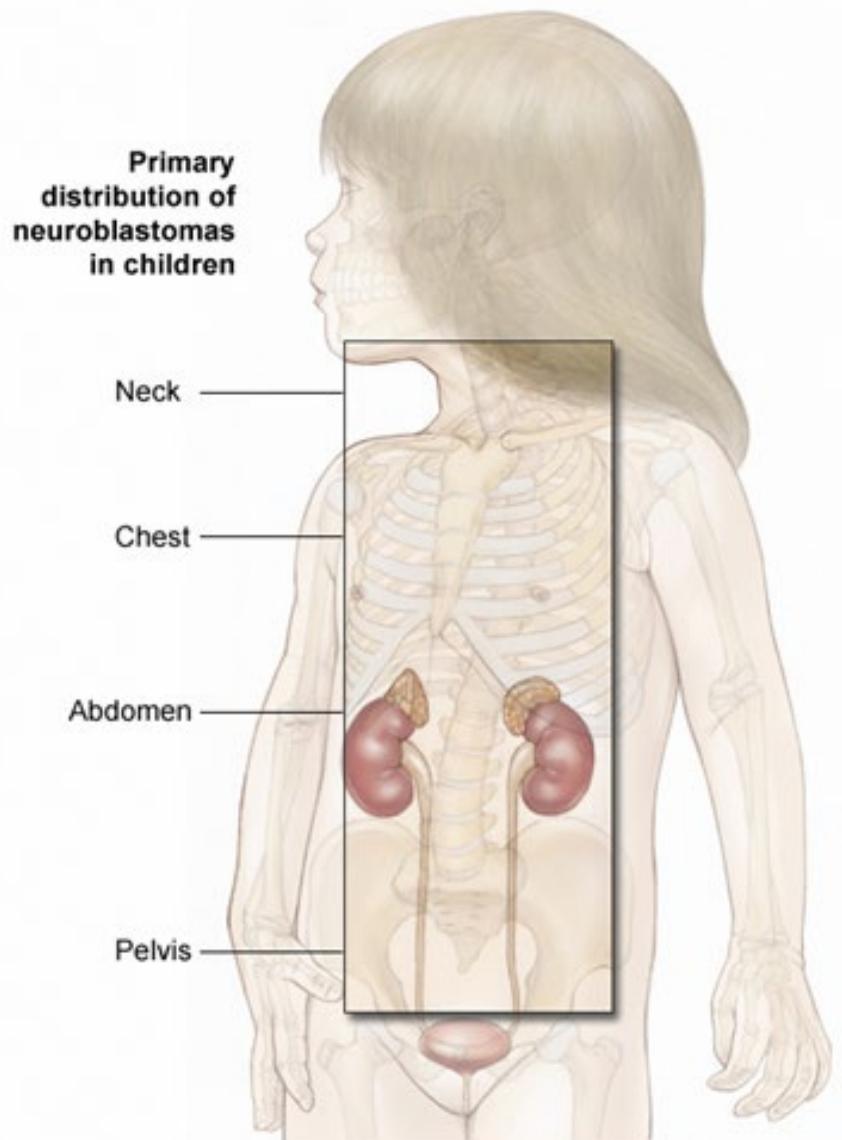
## Chromosome Map



OncoGenomics

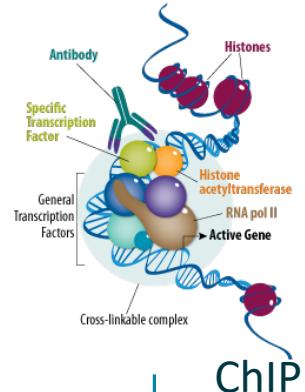
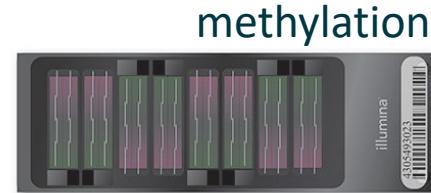
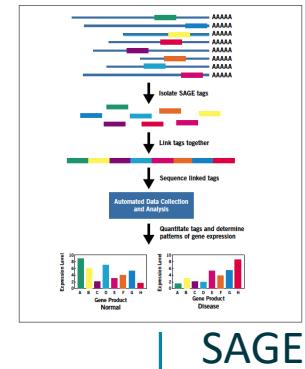
# Neuroblastoma

- Childhood neuro endocrine tumor
  - Incidence Netherlands ~25-40 / year
- Peripheral nervous system
  - Neural crest
  - Neuroblast should normally form neuronal cells
- Young children (mostly 0-3 years)
- Variable clinical outcome
  - Low INSS stages
    - Good prognosis
    - Numeric changes of chromosomal copy numbers
  - High INSS stages
    - Poor prognosis
    - Structural chromosomal defects (**MYCN amp** / **LOH1p** / **11q** etc.)
- Dept. OncoGenomics collected cohort of patient samples with decades of omics data generation to help understand the disease



# R2 organic development closely link with wetlab department

- R2 started as a department resource where the introduction of new technologies resulted in extensions for the platform



Complete genomics

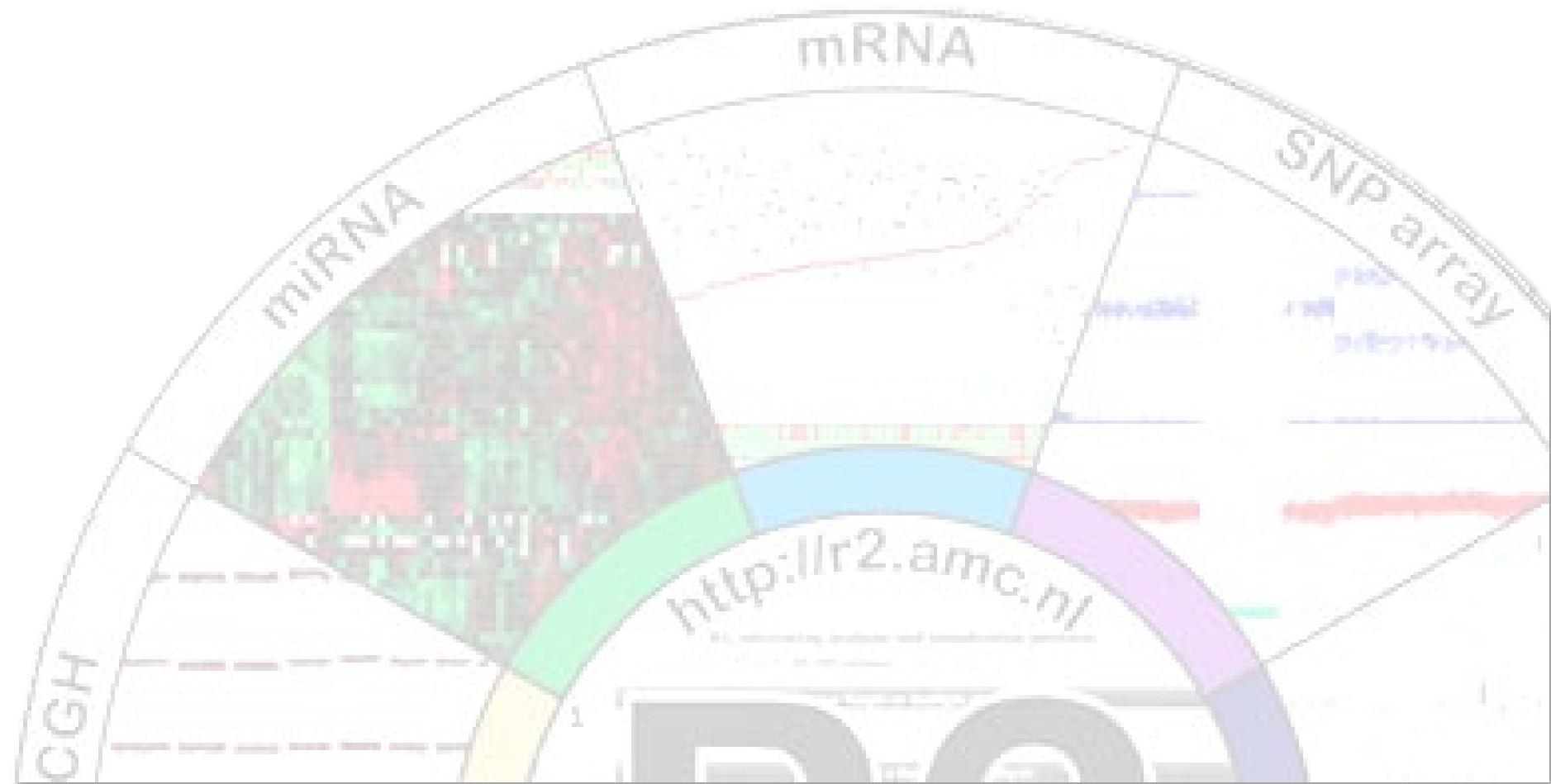
WGS

10X GENOMICS  
Single cell RNA

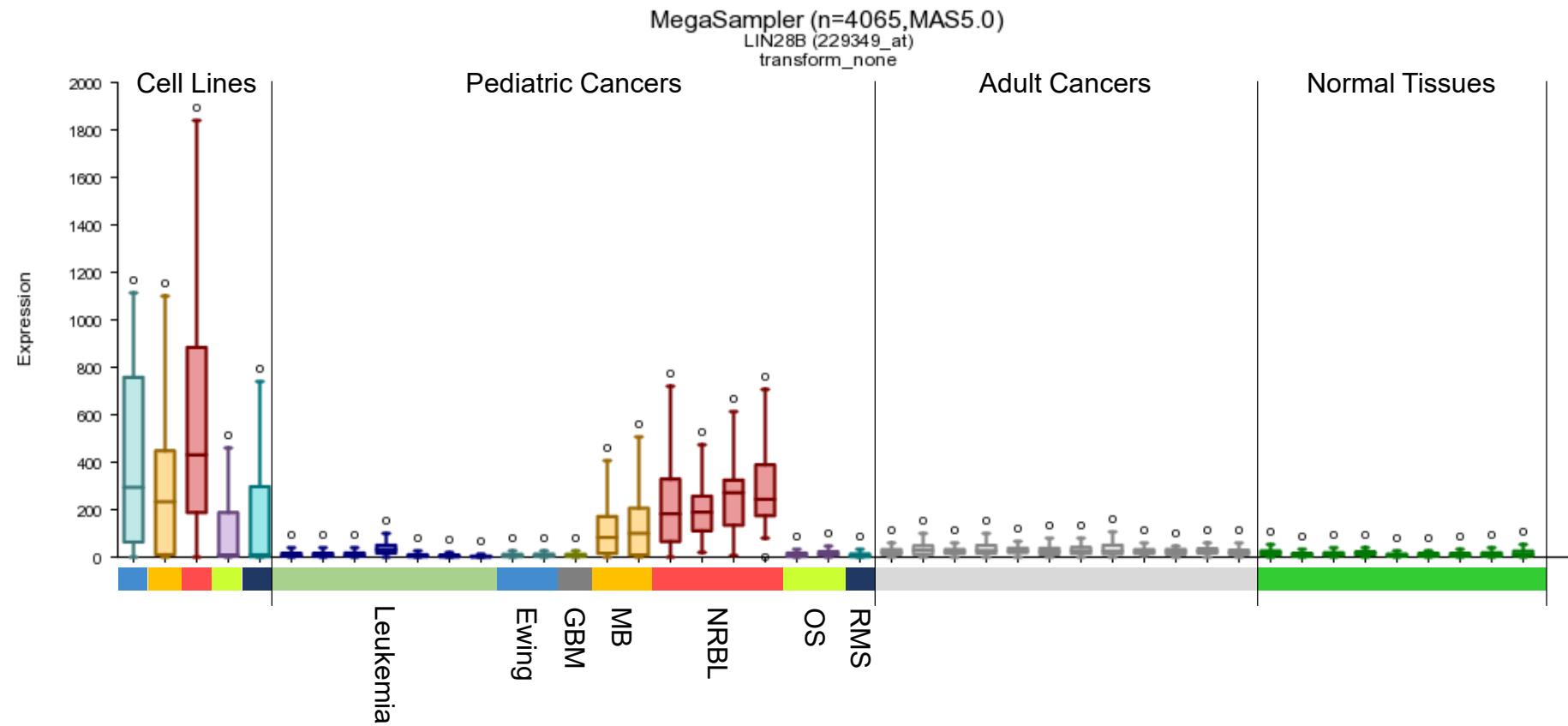
OncoGenomics

# 'Multi'Omics

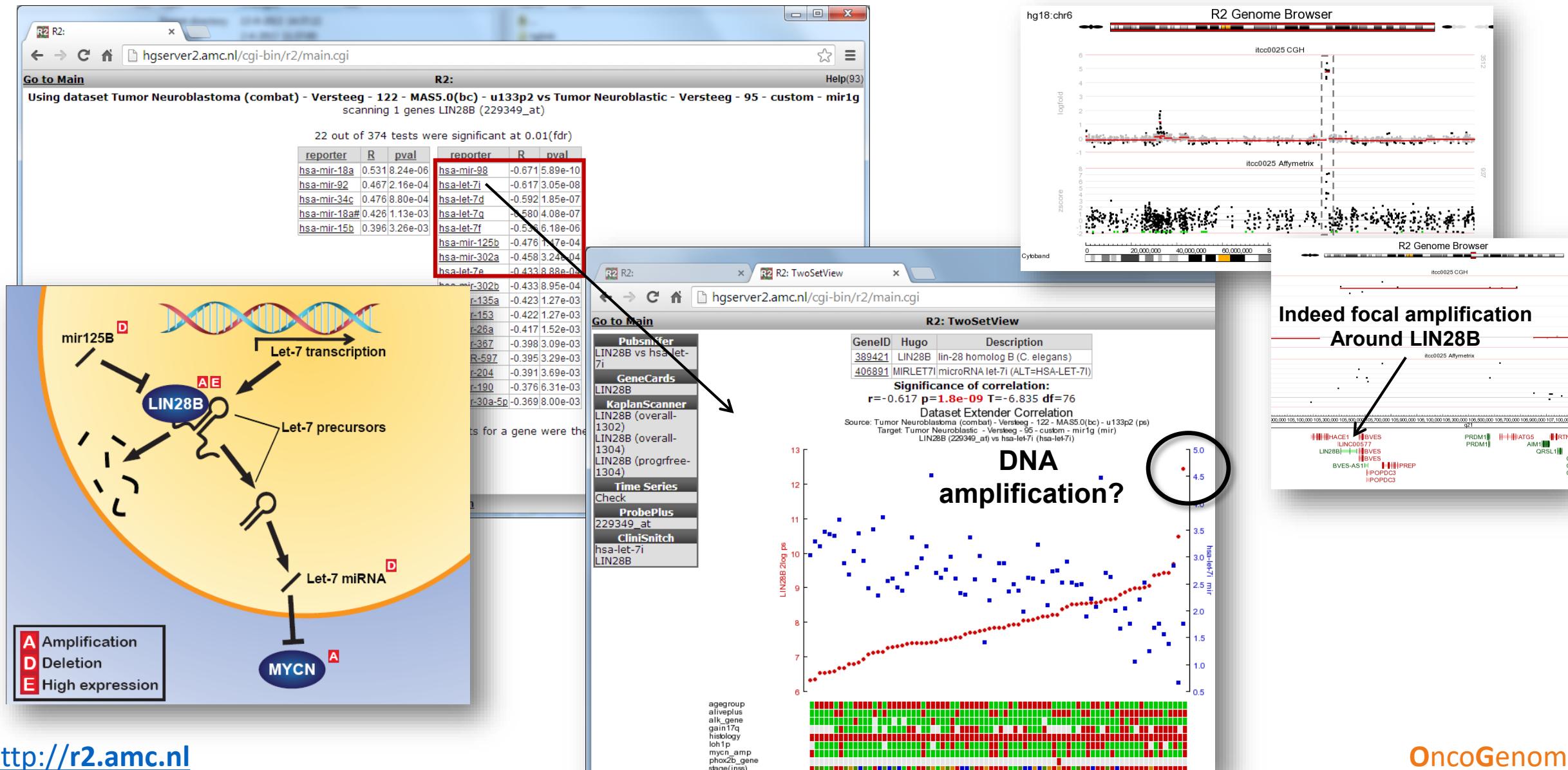
- R2 designed to assist biomedical researchers on their data mining quests
  - Data integrations are relatively straight-forward associations between 2 types of data.
- Will show examples on findings where R2 was an important component over the last decade
- Nearly all visualizations are coming from R2



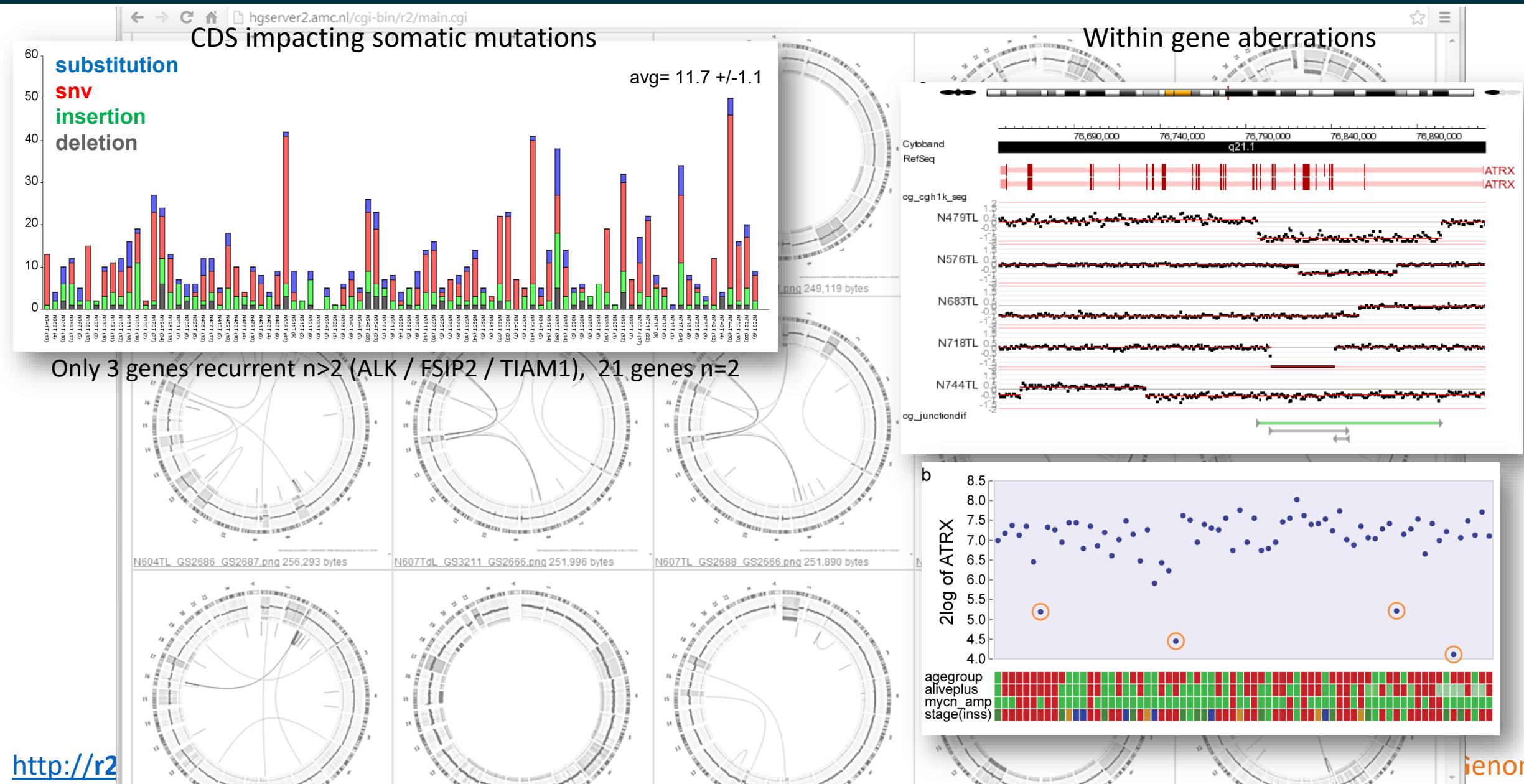
# Gex data



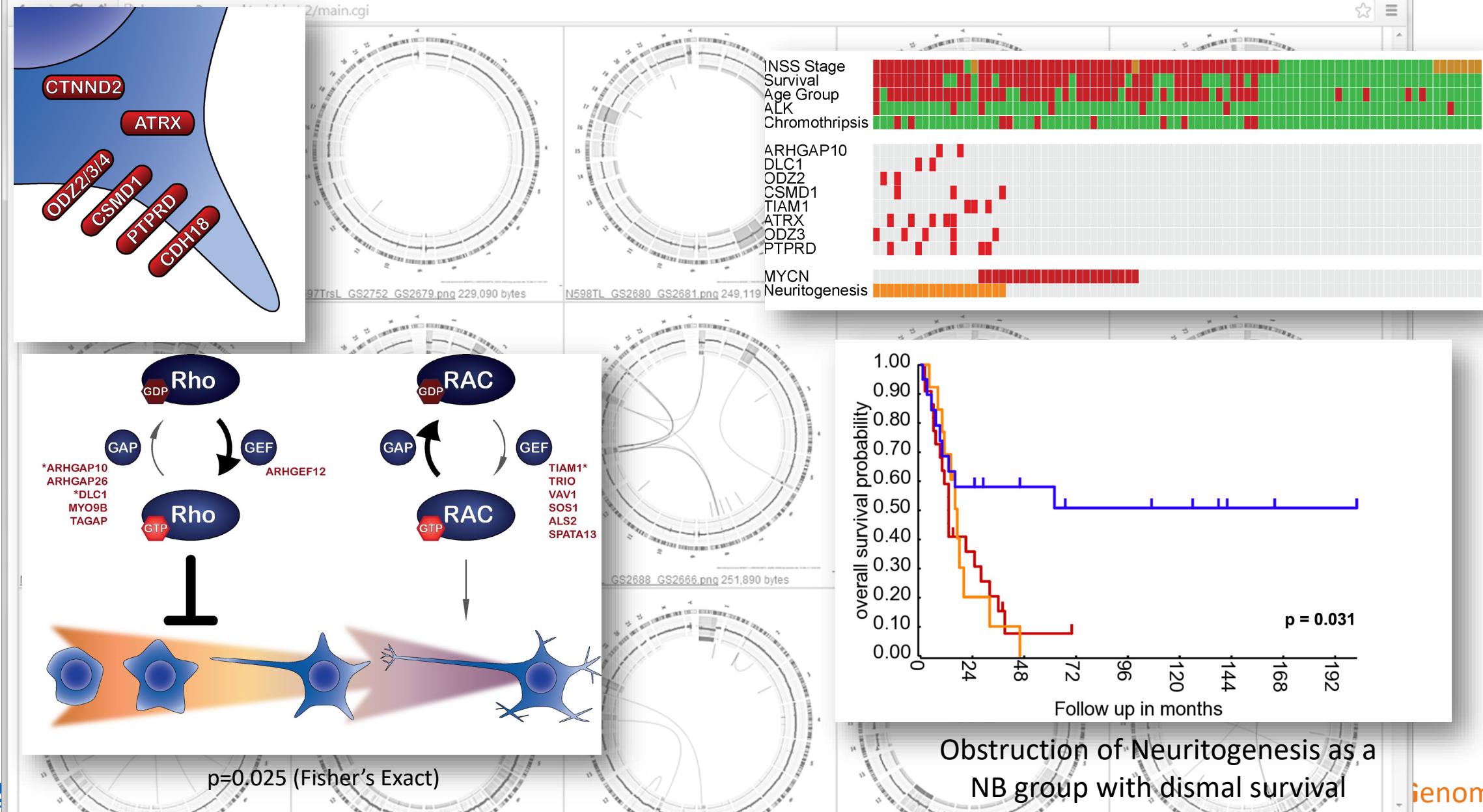
# mRNA vs qRT miRNA



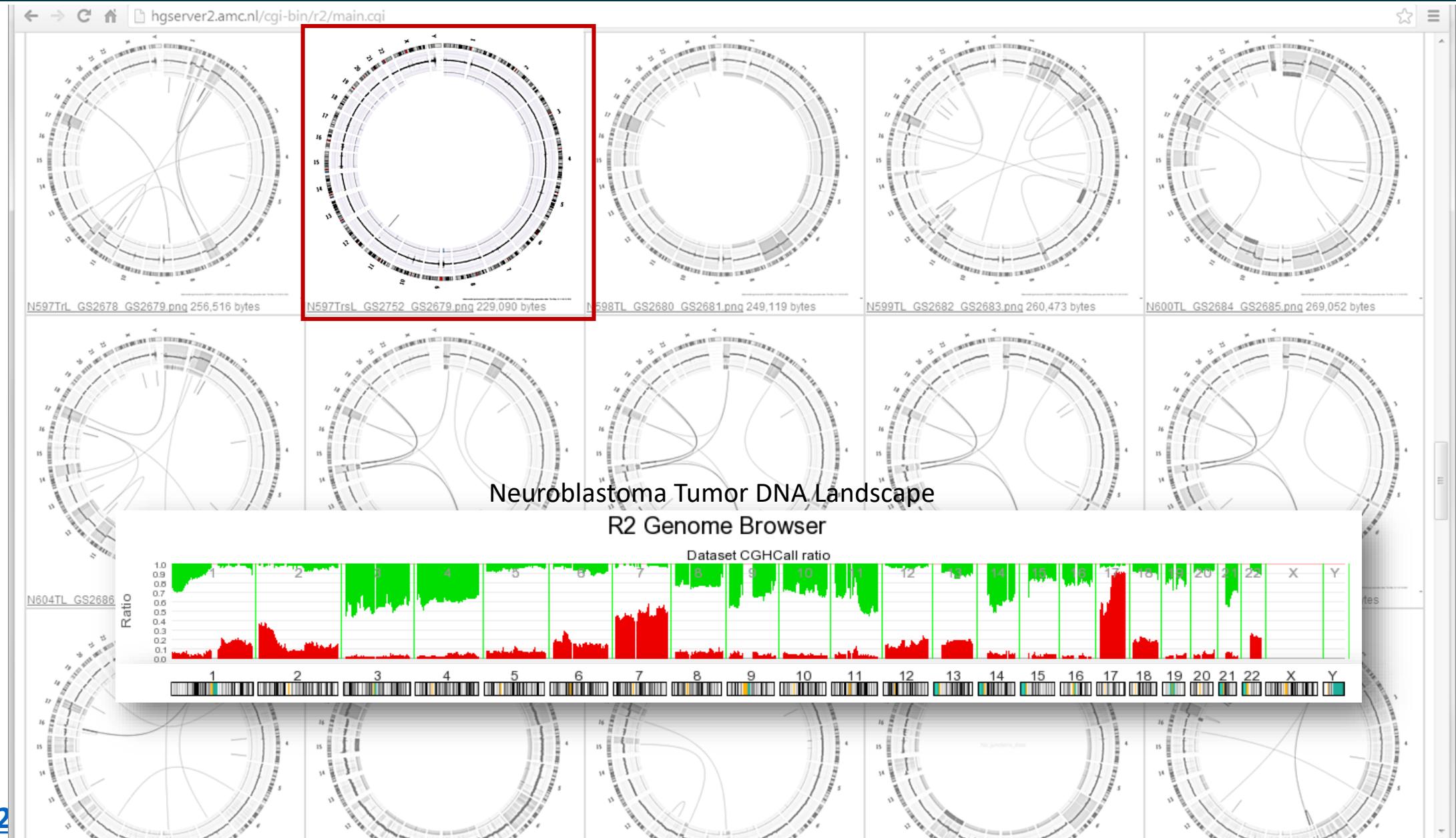
# WGS 87 Tumor / Normal pairs



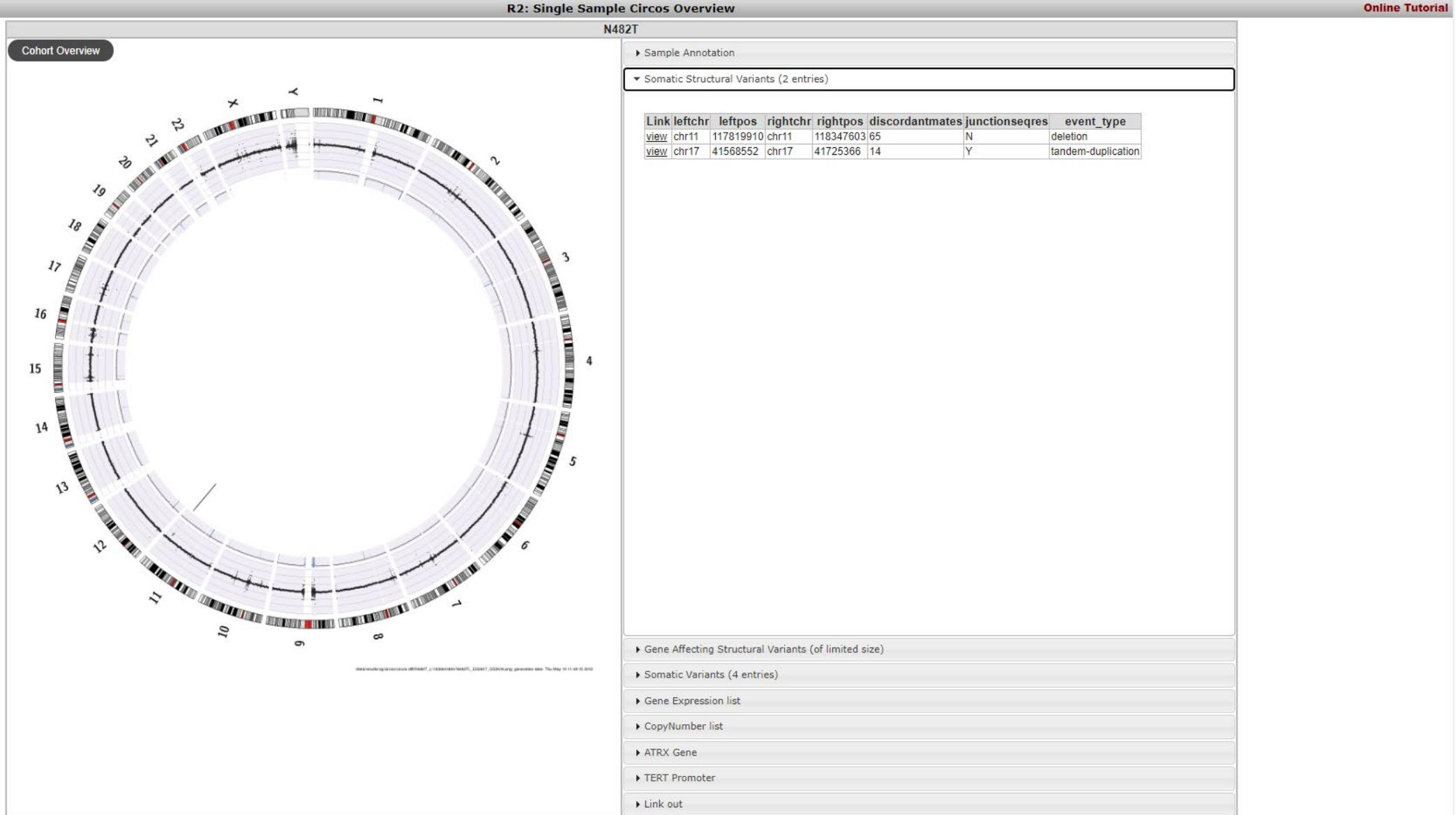
# Aberrations in Neuritogenesis pathway as high risk group



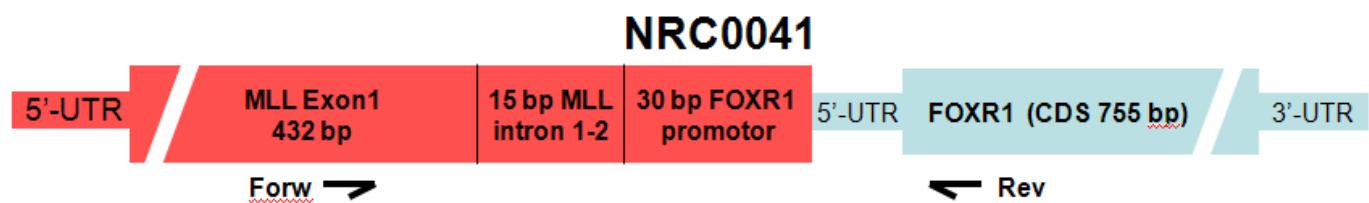
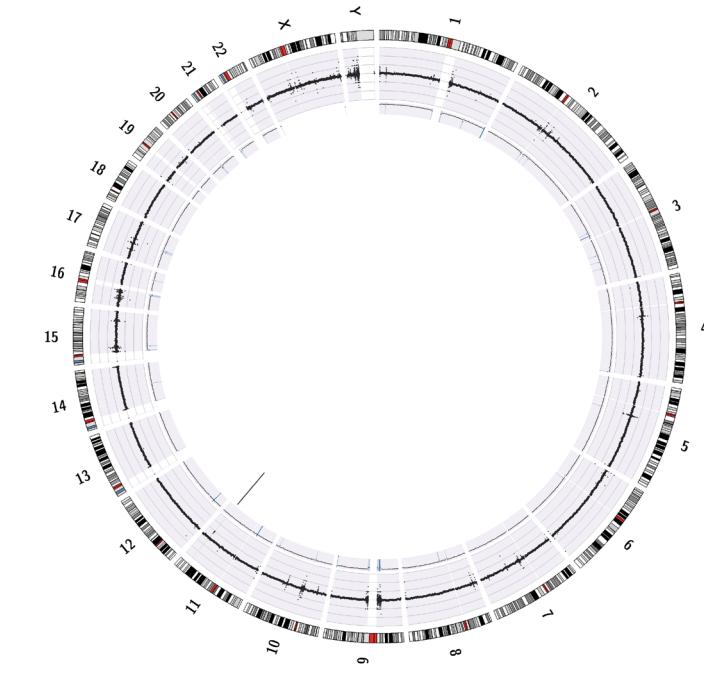
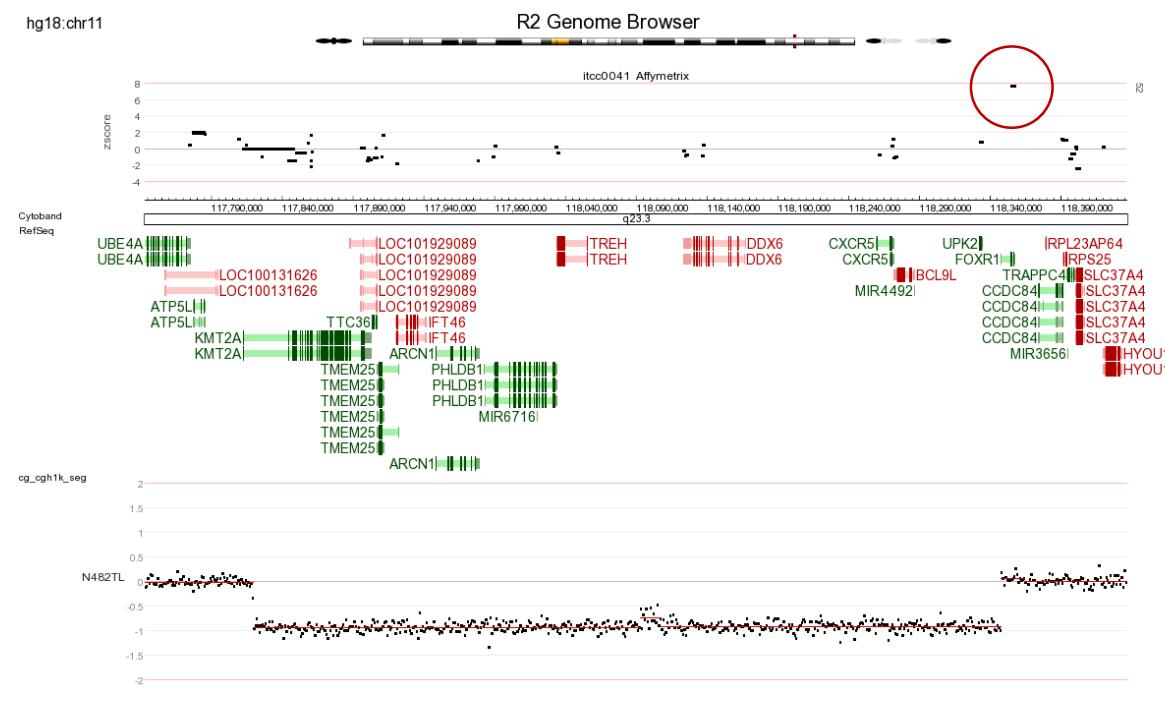
## atypical cases



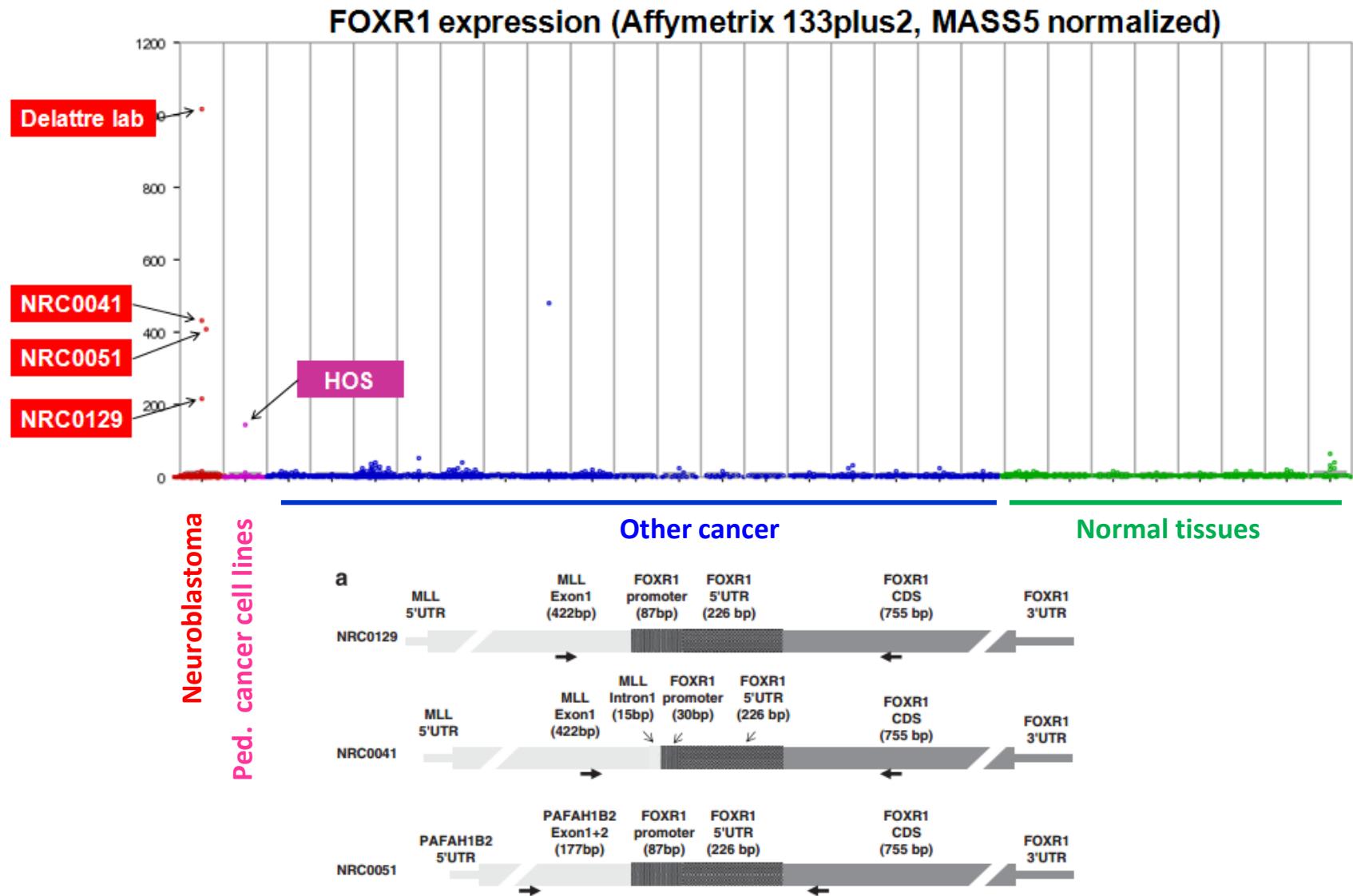
# Detailed Sample focused views

[Go to: Main](#)[Online Tutorial](#)

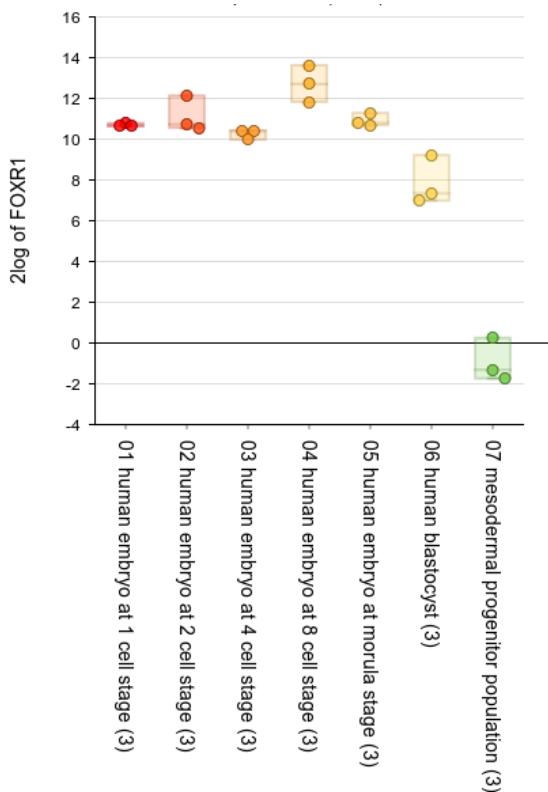
# Neuroblastoma patient with single event



# FOXR1 only expressed in combination with promoter hijacking

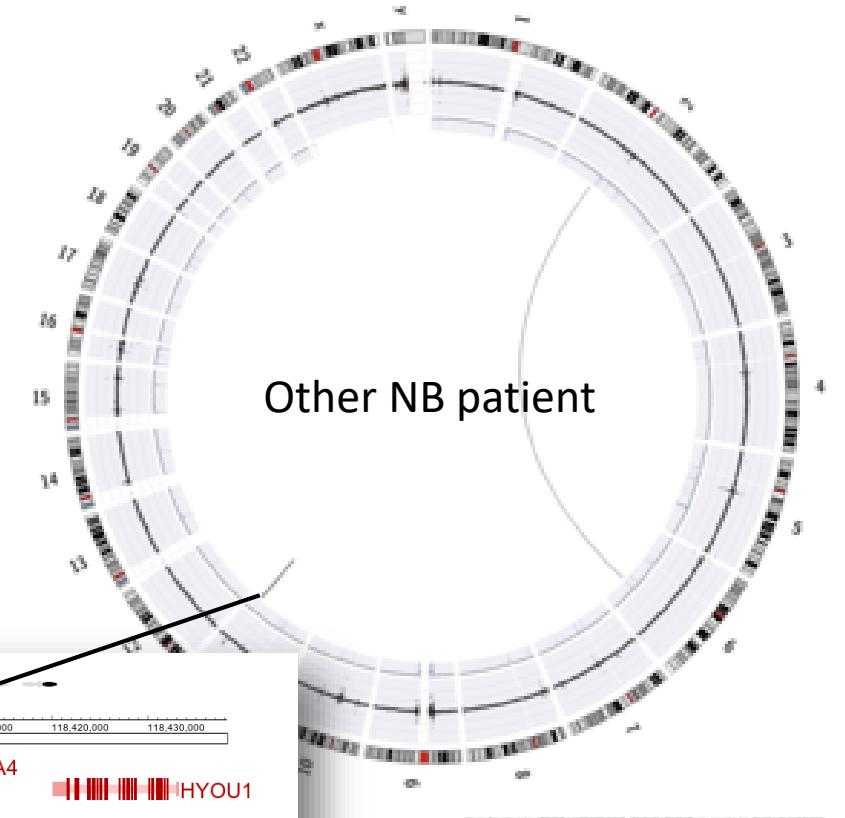
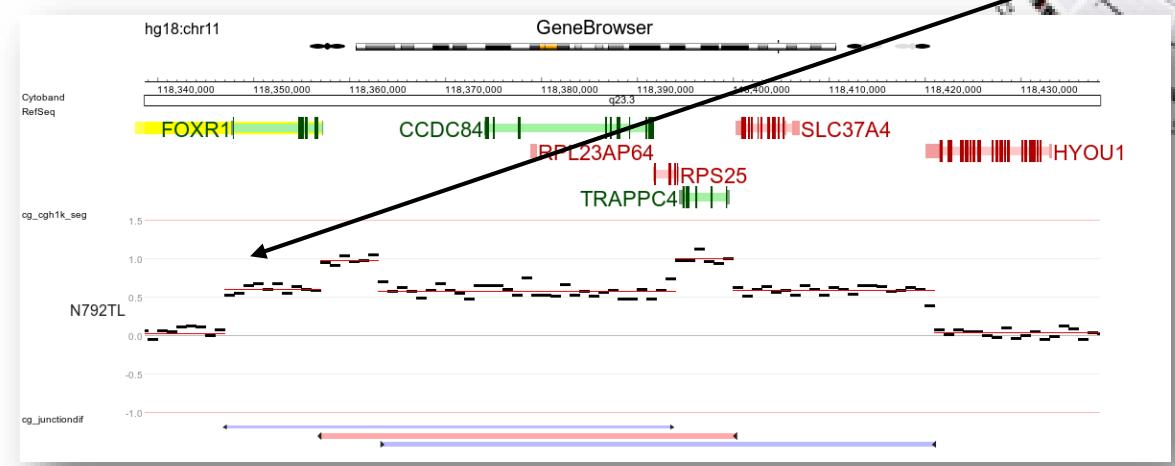
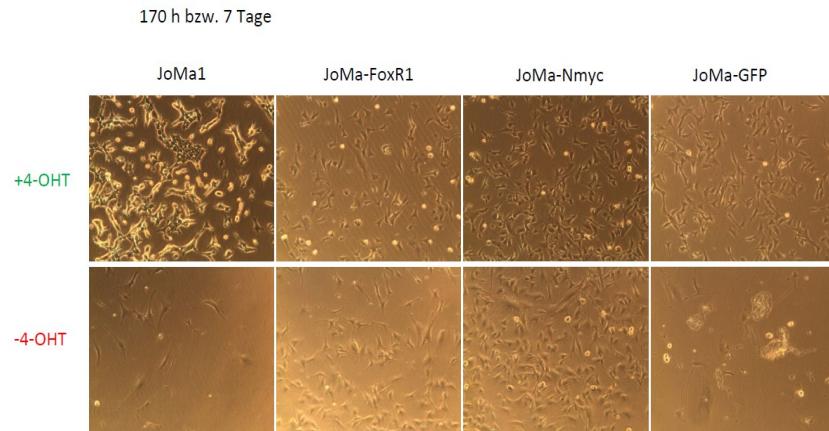


Early human embryogenesis



# FOXR1

Induction of FOXR1 expression drives growth of normal non-malignant neuroblasts

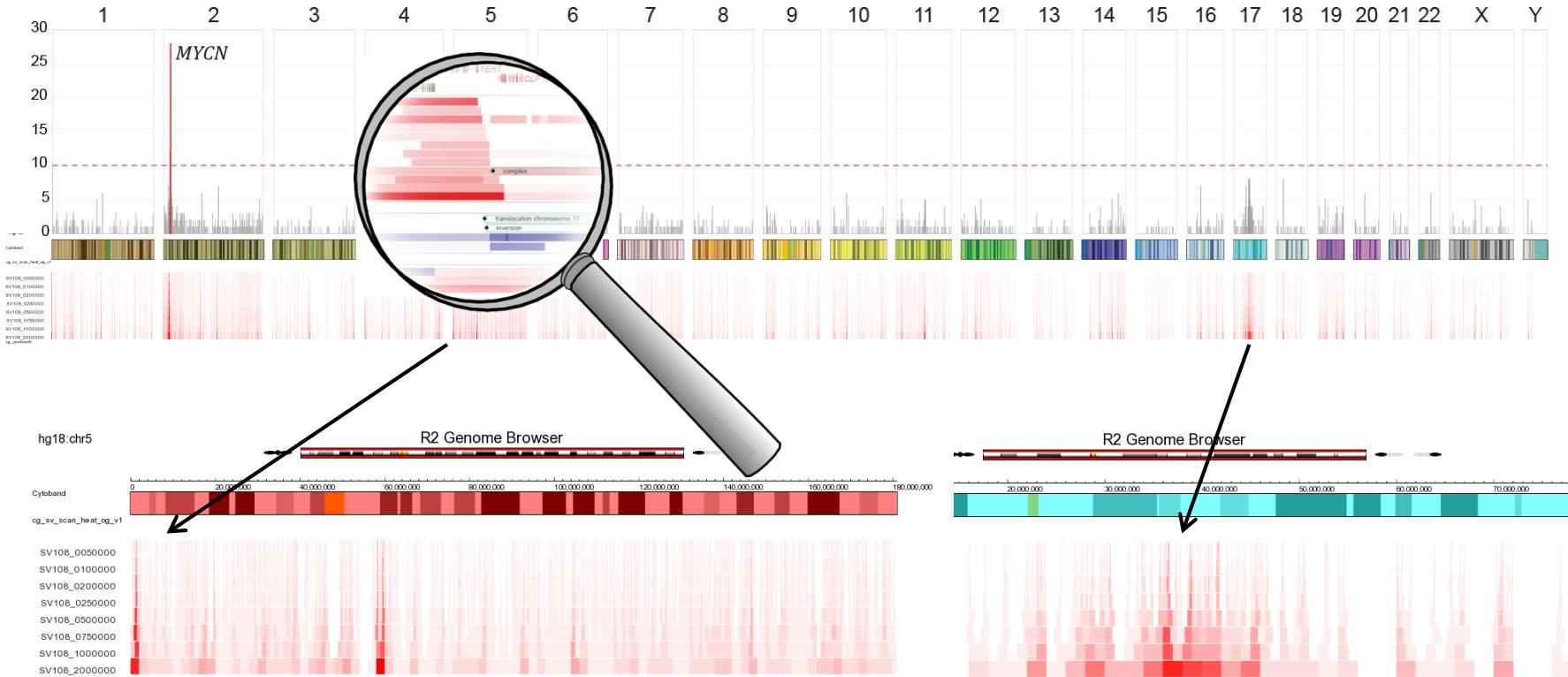


# Structural Variations

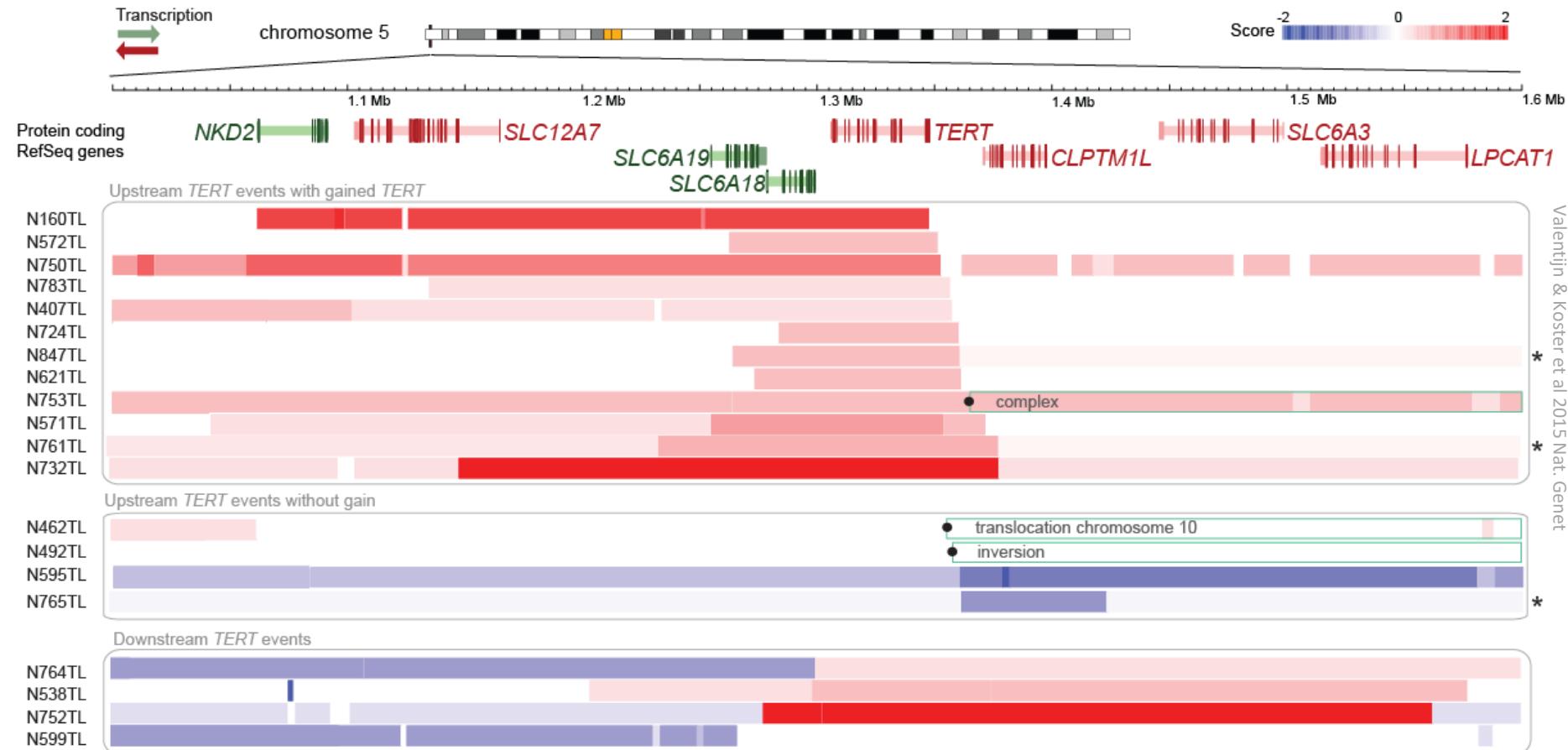


# Scan for Enrichment in SV

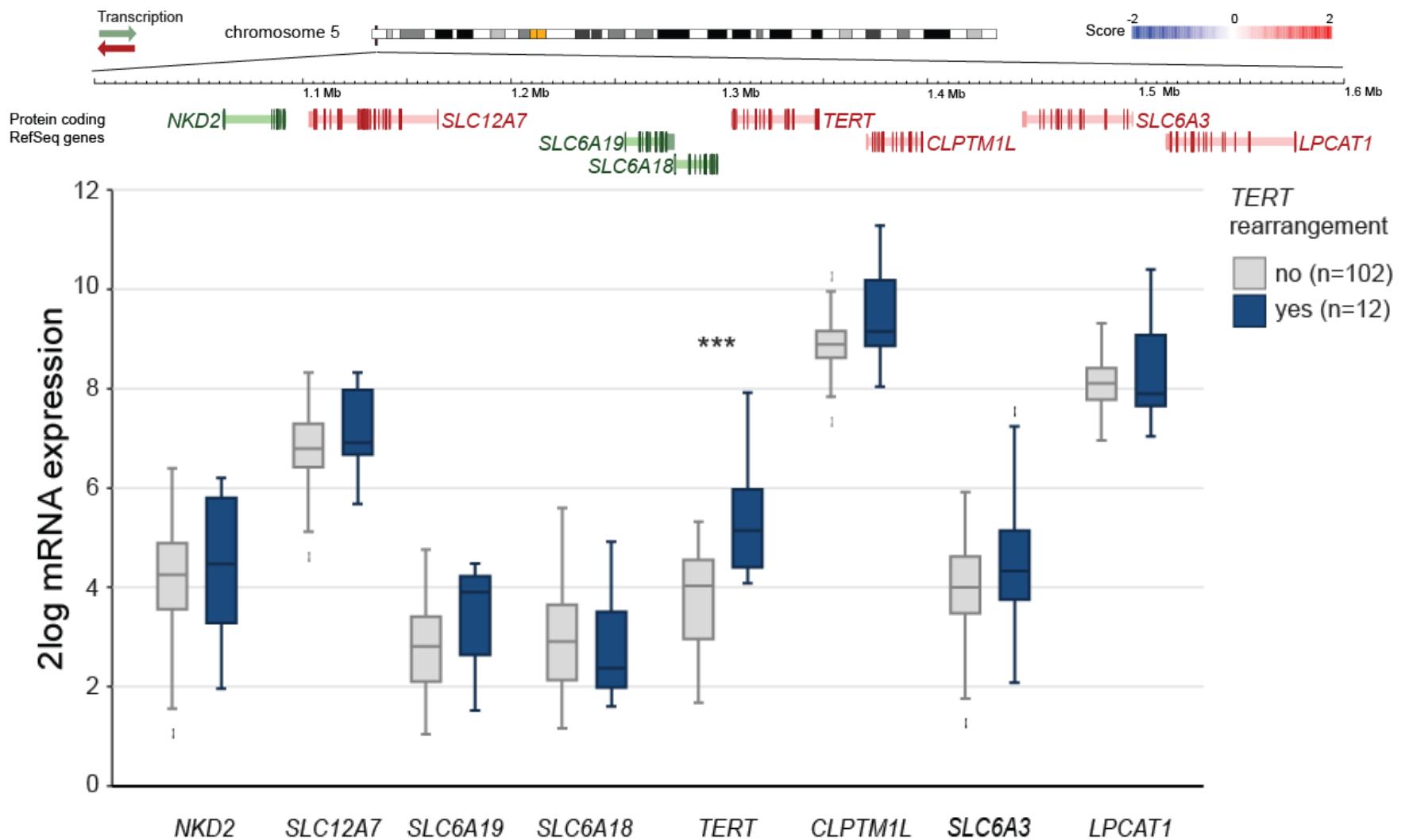
- Are there hotspots of SV in the neuroblastoma genome?



# TERT region has gains, losses and inversions in 23% of high stage NB

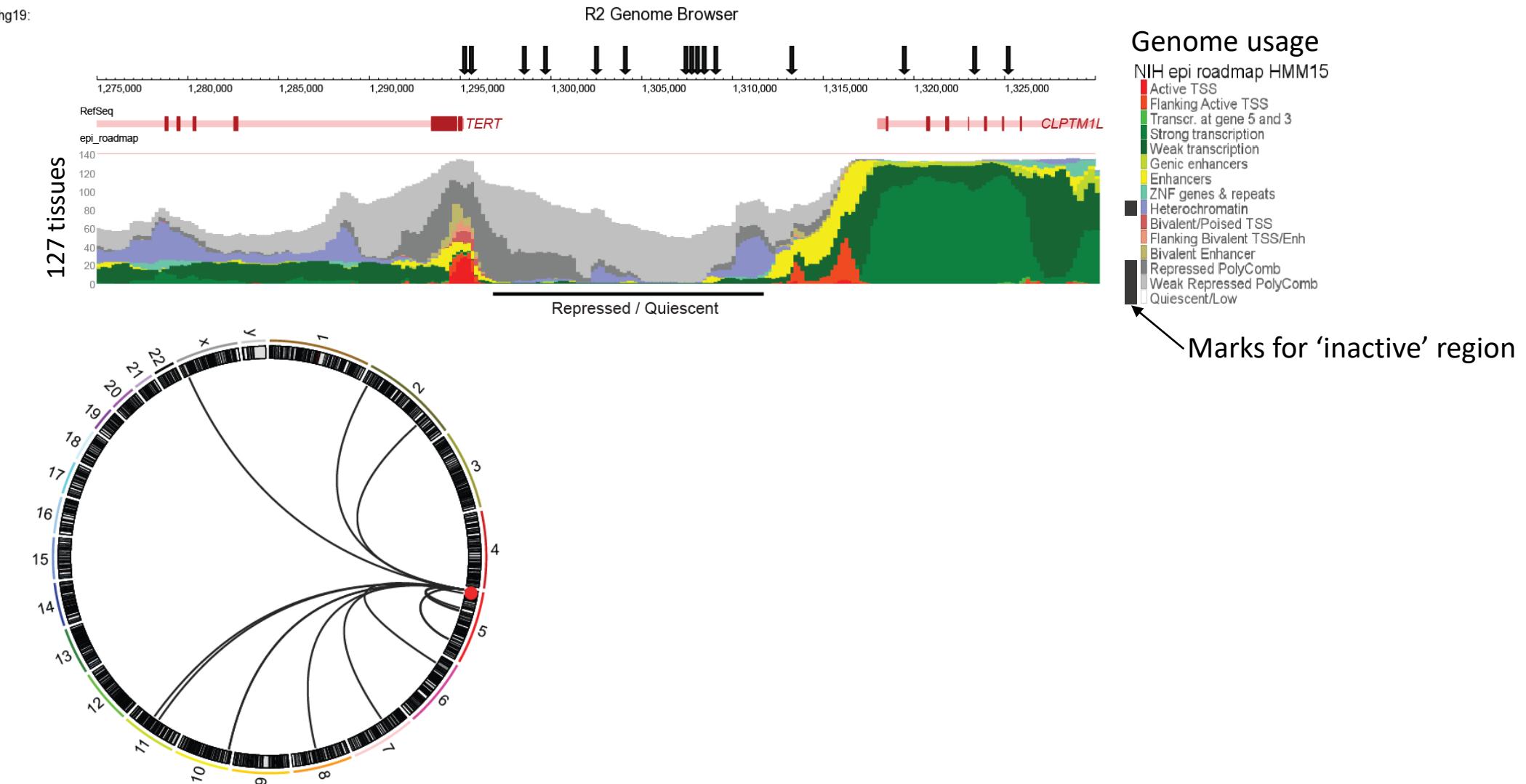


# TERT gene expression affected

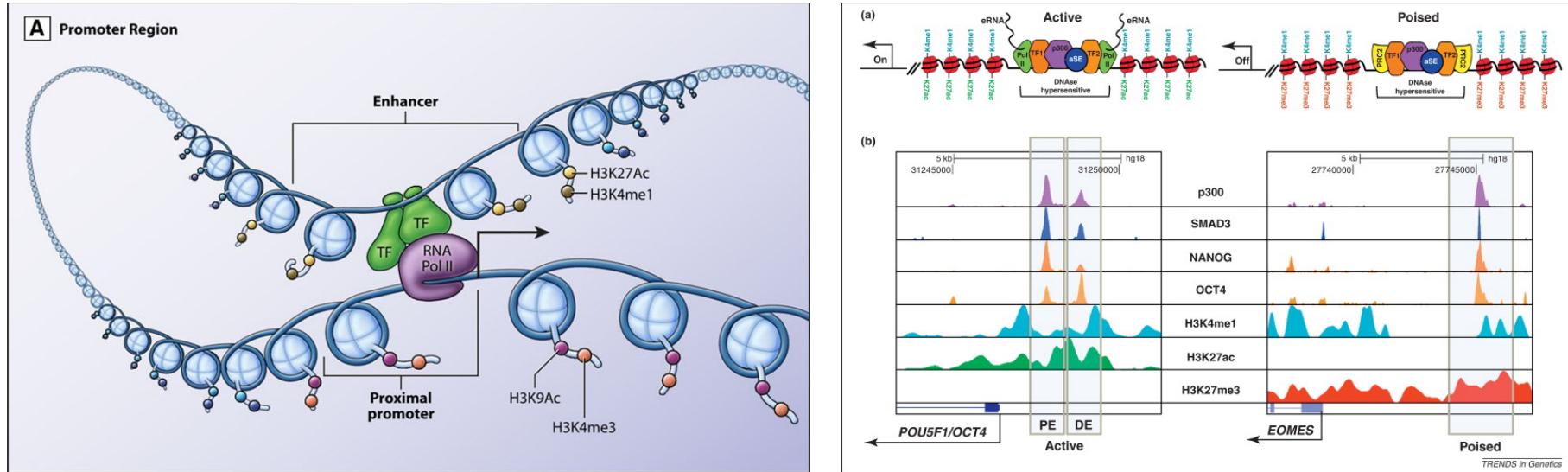


Valentijn & Koster et al 2015 Nat. Genet

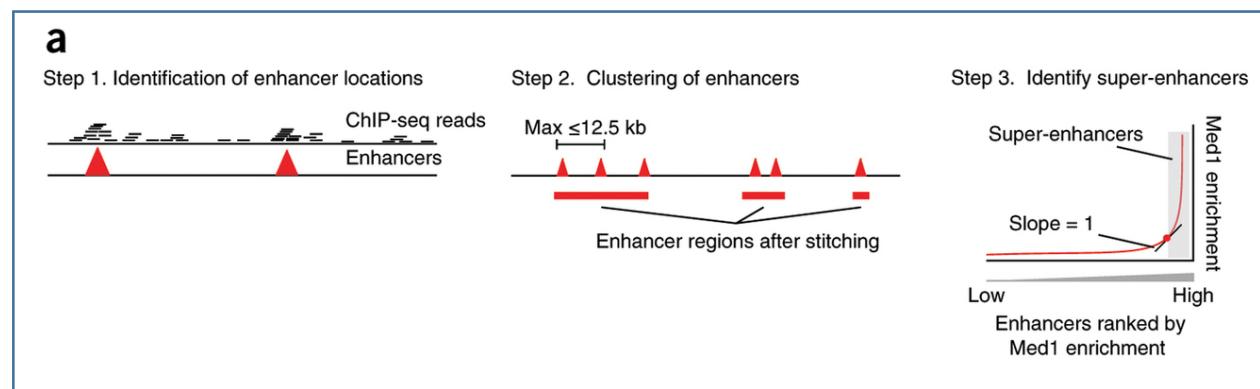
# TERT upstream region mostly quiescent / repressed



# Enhancers



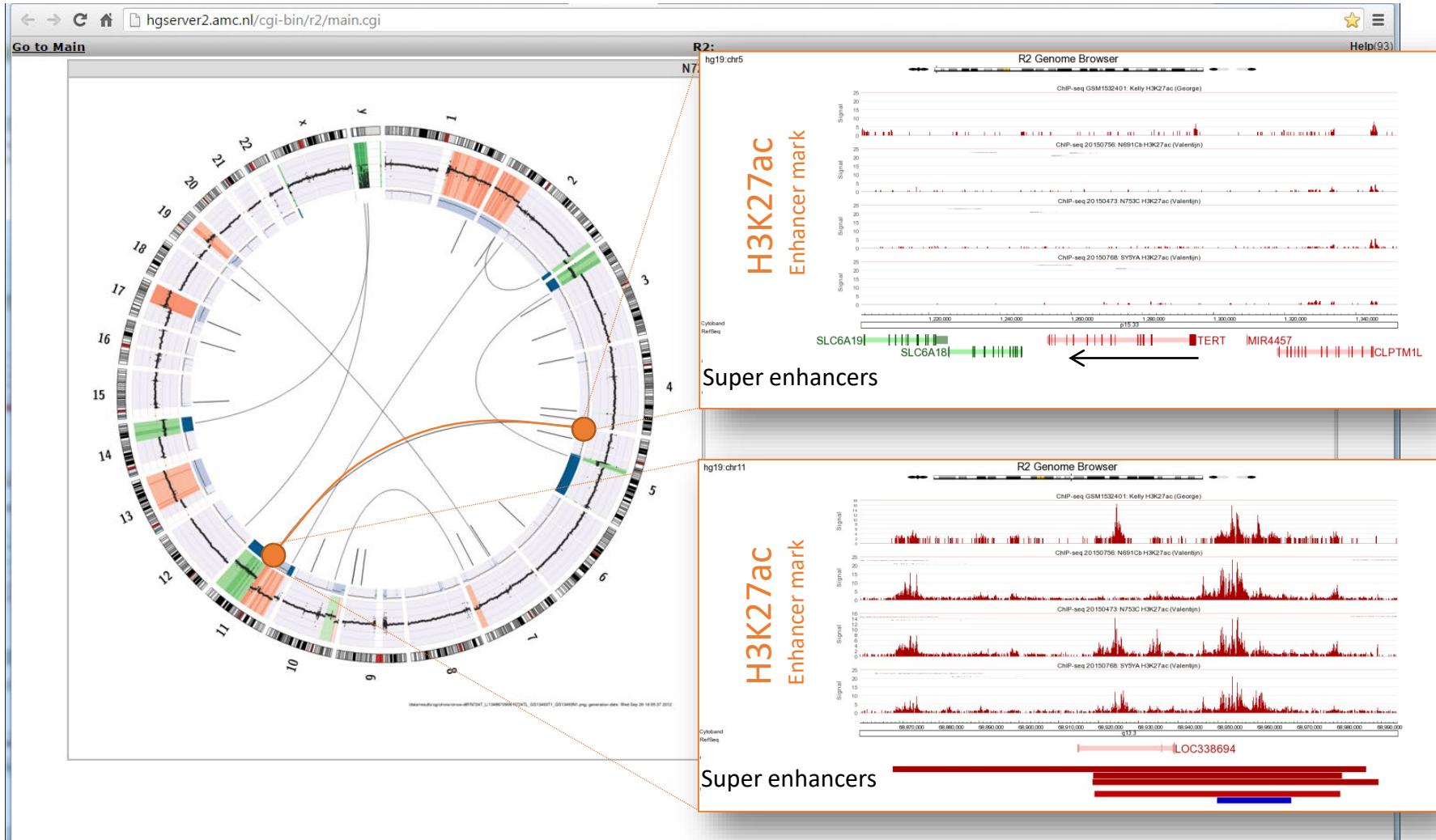
Active enhancers are marked by H3K27Ac



Super Enhancers are the most 'active / enriched' enhancers

<http://atvb.ahajournals.org/content/35/11/2297>  
<http://www.nature.com/ng/journal/v47/n1/full/ng.3167.html>  
[http://www.cell.com/trends/genetics/fulltext/S0168-9525\(12\)00033-9](http://www.cell.com/trends/genetics/fulltext/S0168-9525(12)00033-9)

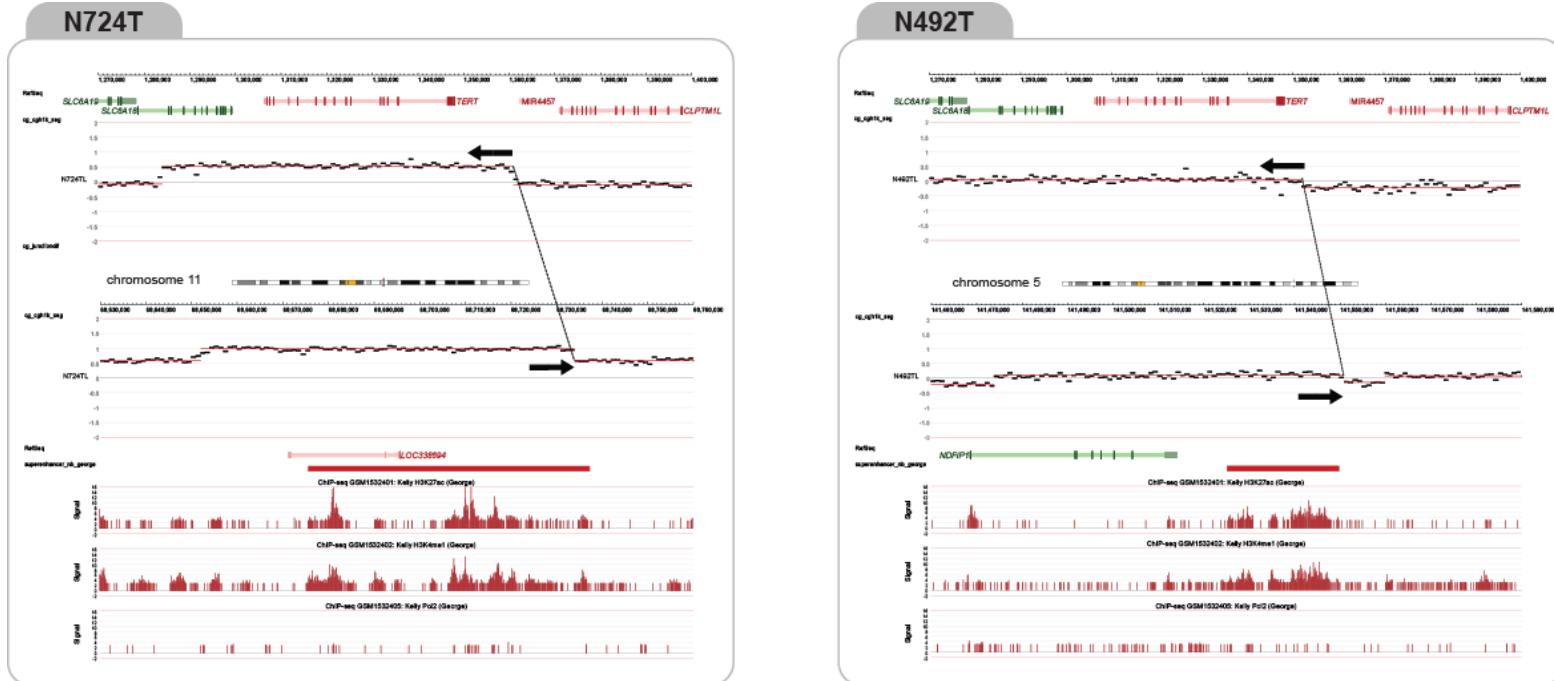
# TERT translocates to super-enhancers



# TERT translocates to super-enhancers



# TERT translocates to super-enhancers (enhancer hijacking)

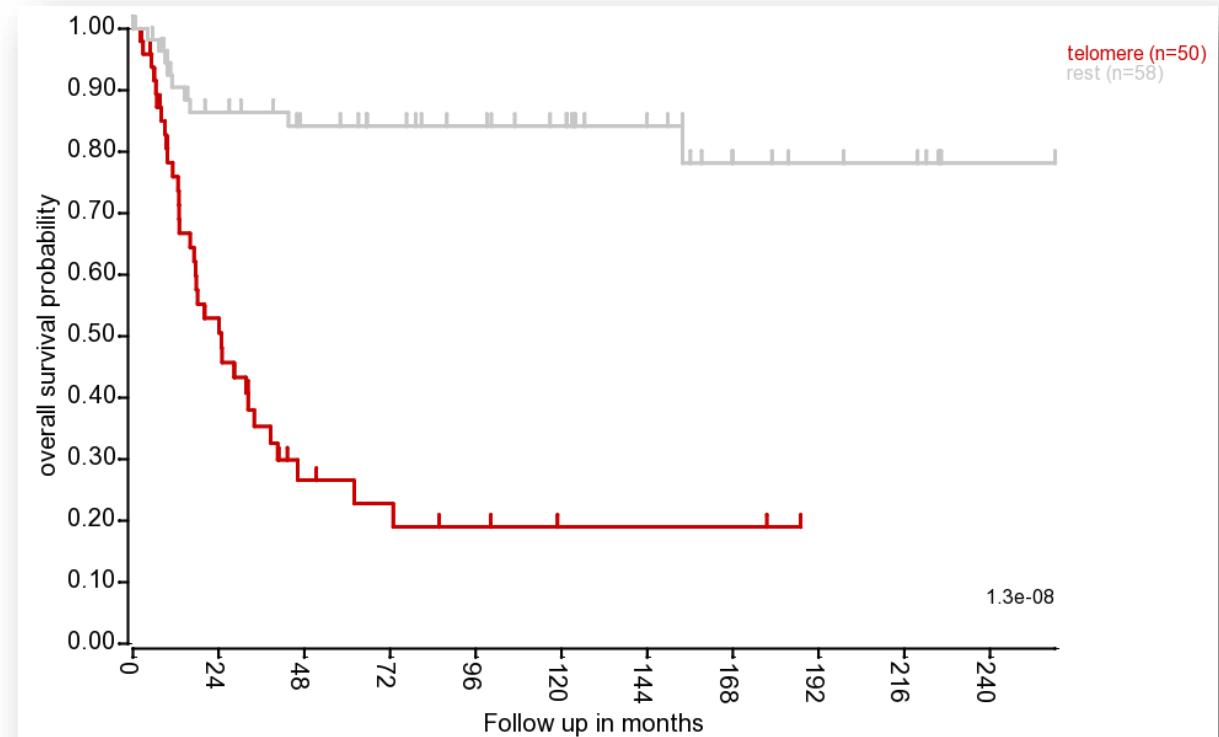
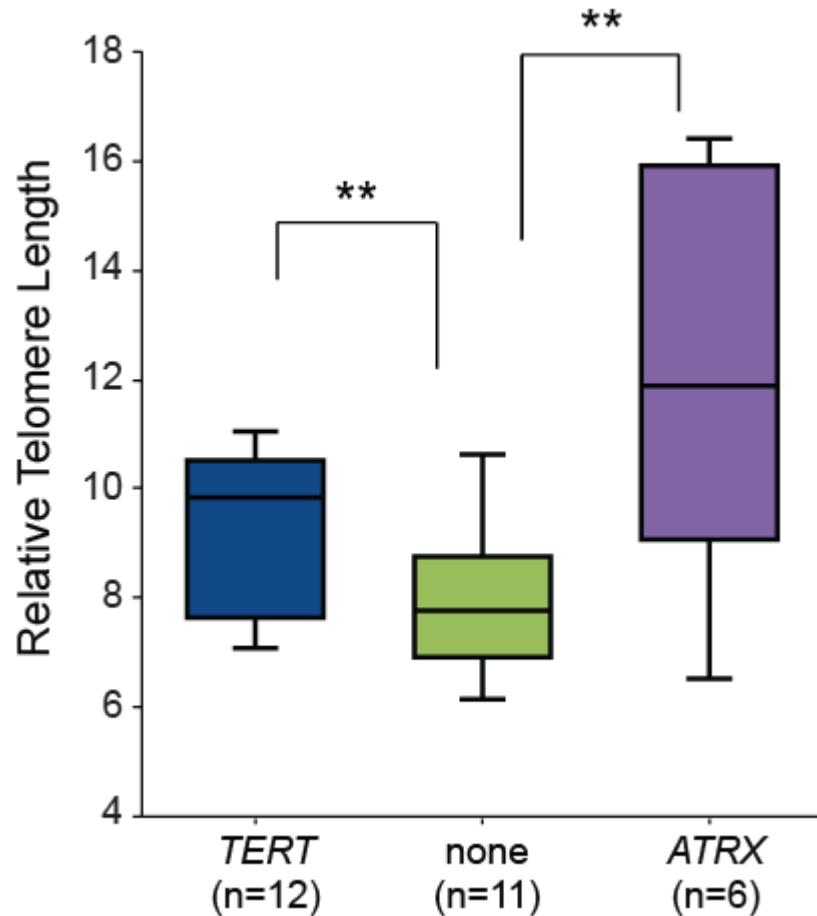


Valentijn & Koster et al 2015 Nat. Genet

	r100k	r250k	r500k	r750k	r1000k
p count>=5	0.00034				
p count>=6		0.0011			
p count>=7			0.00306		
p count>=9				0.00045	0.00234

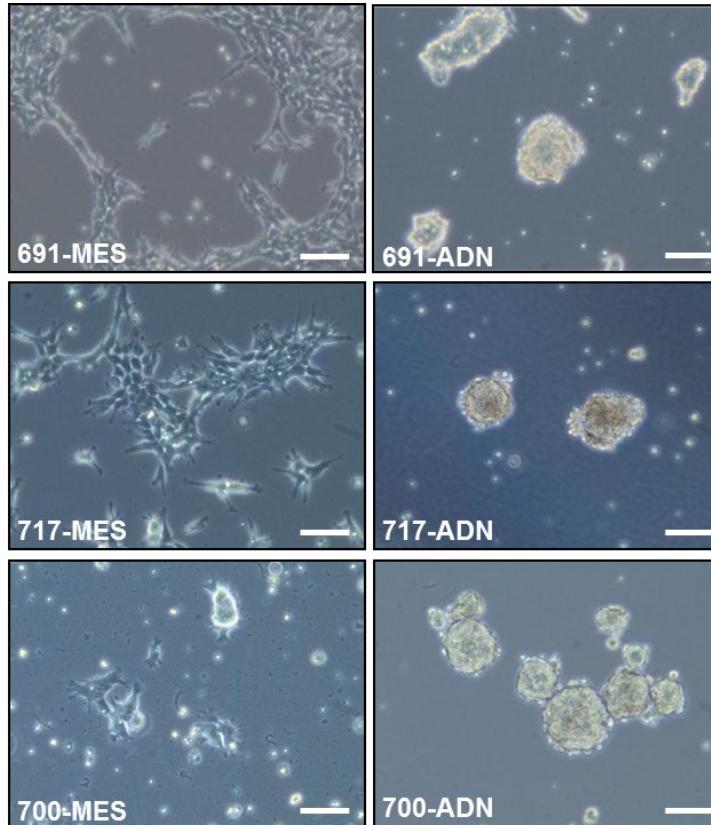
100,000 iterations of random breakpoints

# Longer telomeres, Nb is a telomere disease

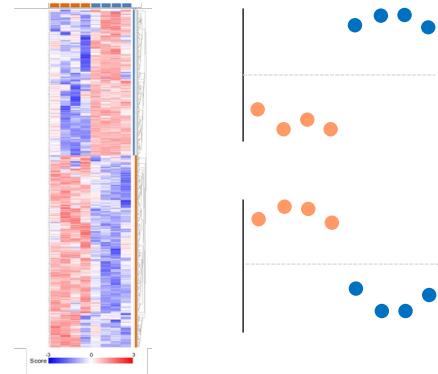


# Neuroblastoma sub-clones

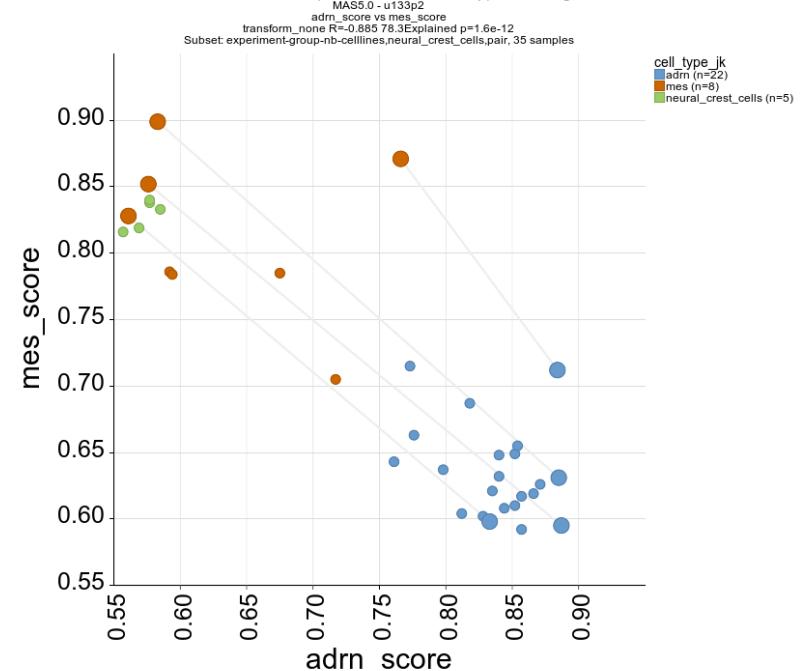
- Patient derived cell lines
  - Multiple lines from same patient
  - 2 phenotypes
    - Adrenergic (epithelial like)
    - Mesenchymal (stem cell like)
- mRNA clustering
  - By phenotype
- Experiments show
  - Types can interconvert
- Epigenetic?



mRNA Geneset score

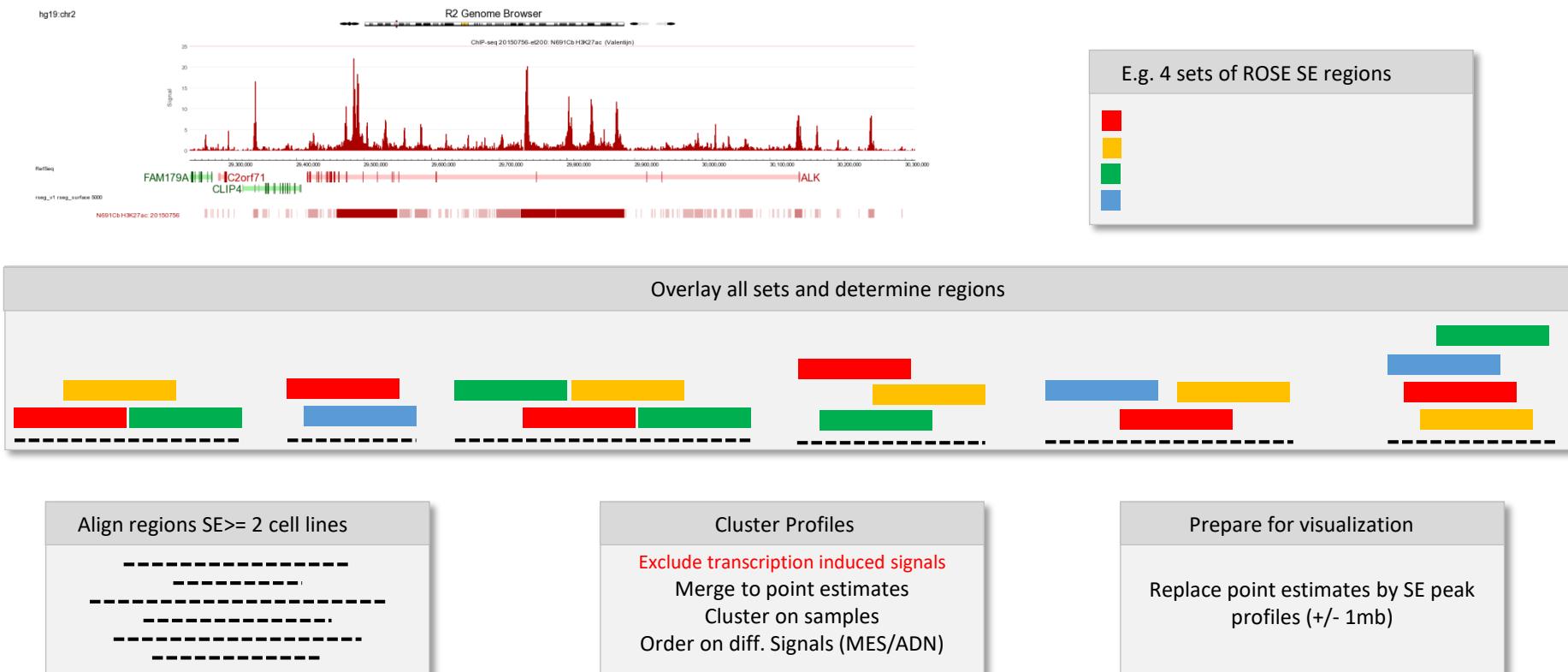


Mixed Neuroblastoma (MES-ADRN-Crest-Exp) - Versteeg - 52

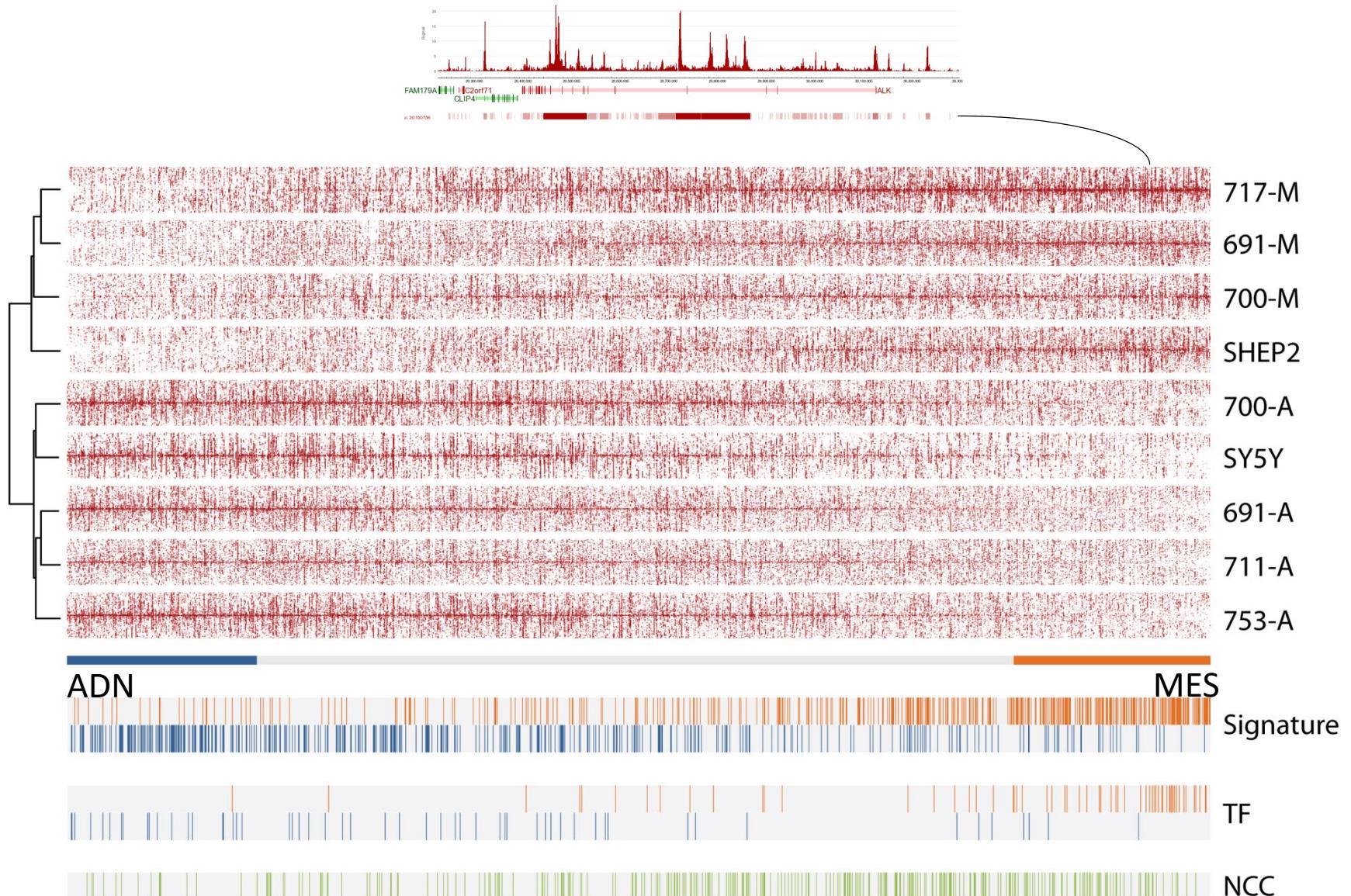


# Super Enhancer Landscape

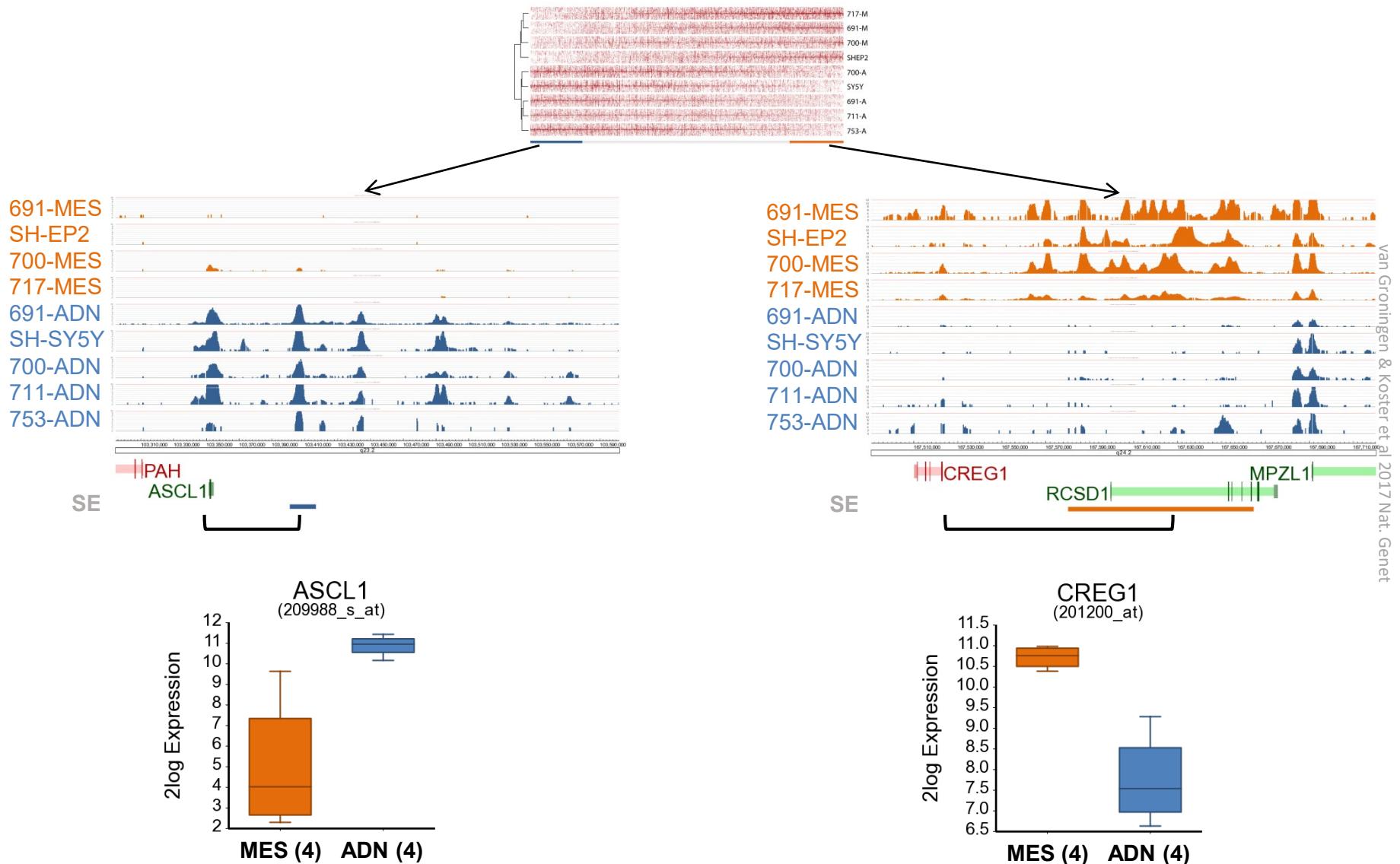
- Assessed enhancer landscape in 9 samples
  - 4 X MES
  - 5 X ADN



# SuperEnhancer Landscape MES/ADN

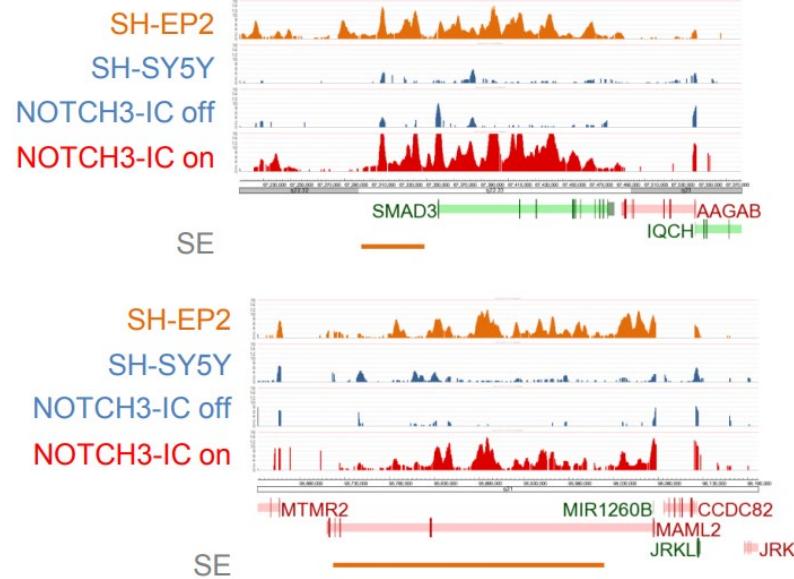
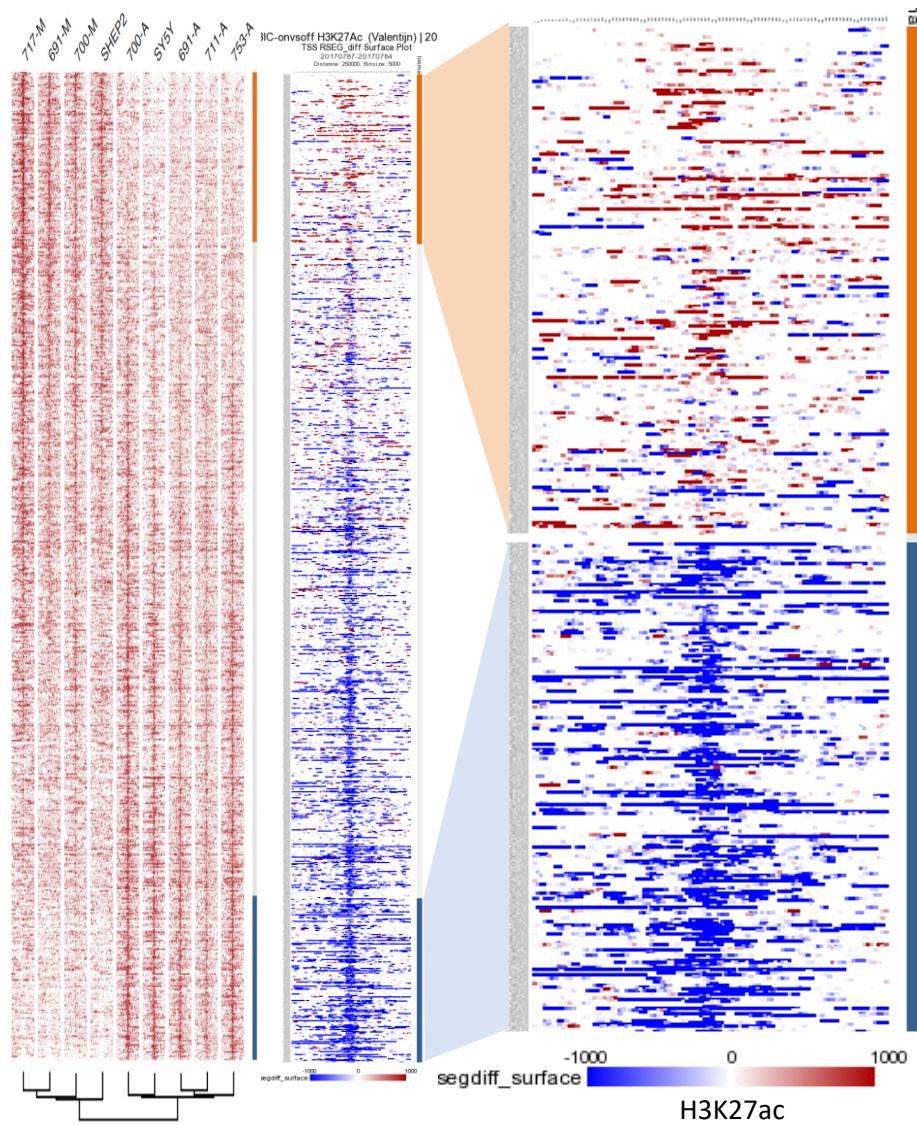
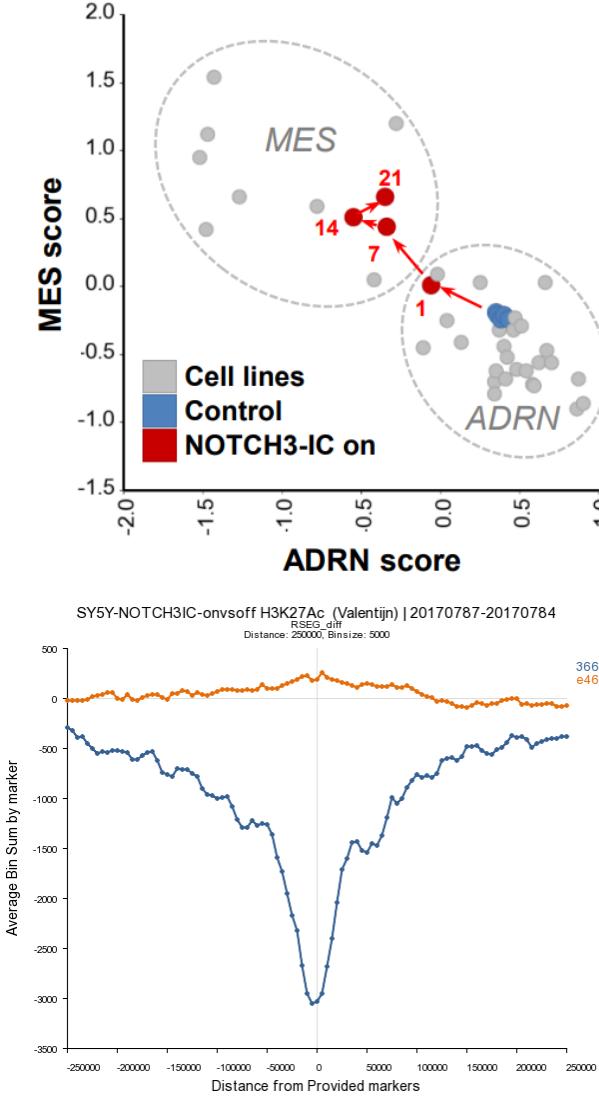


# SuperEnhancer Landscape MES/ADN

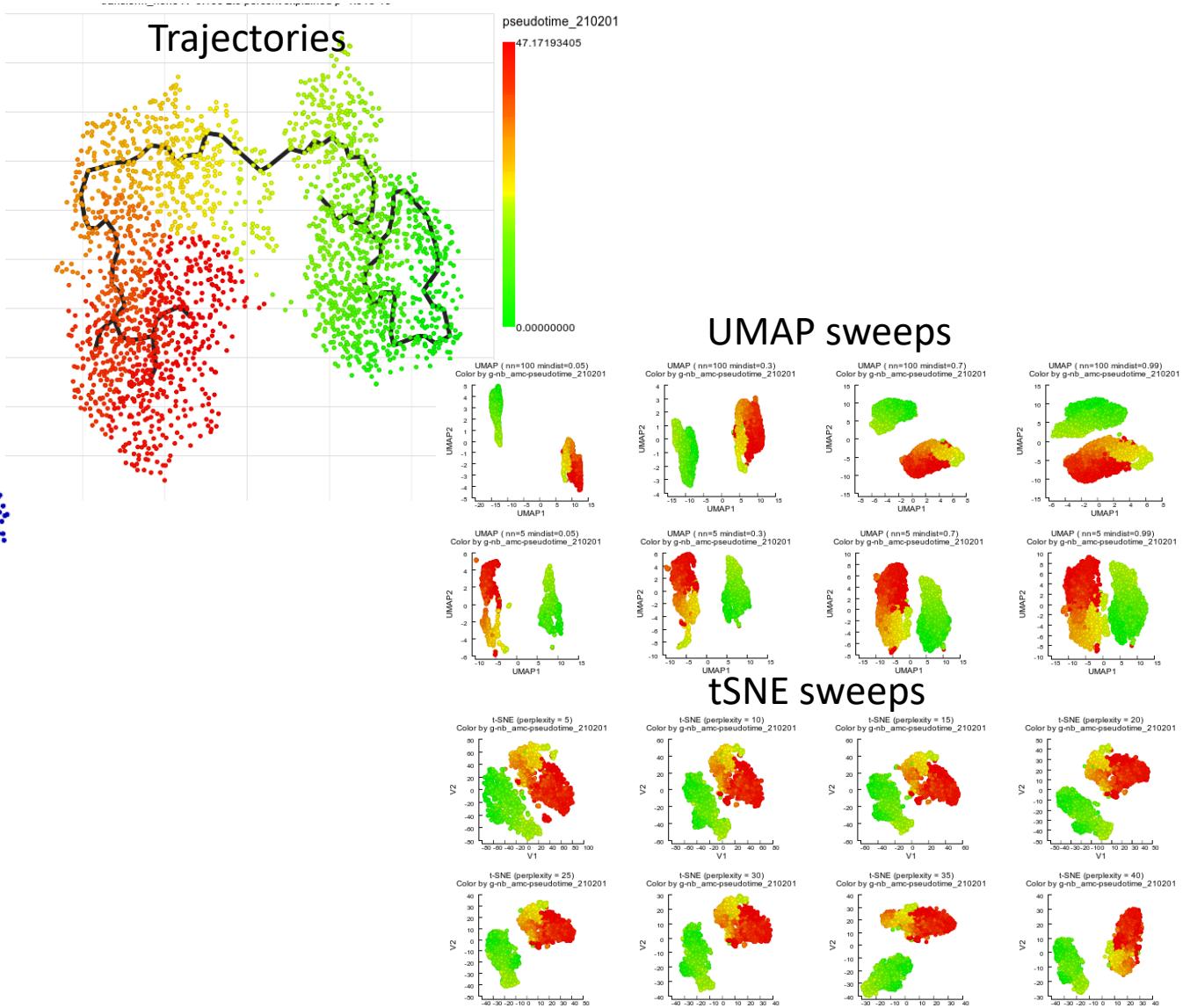
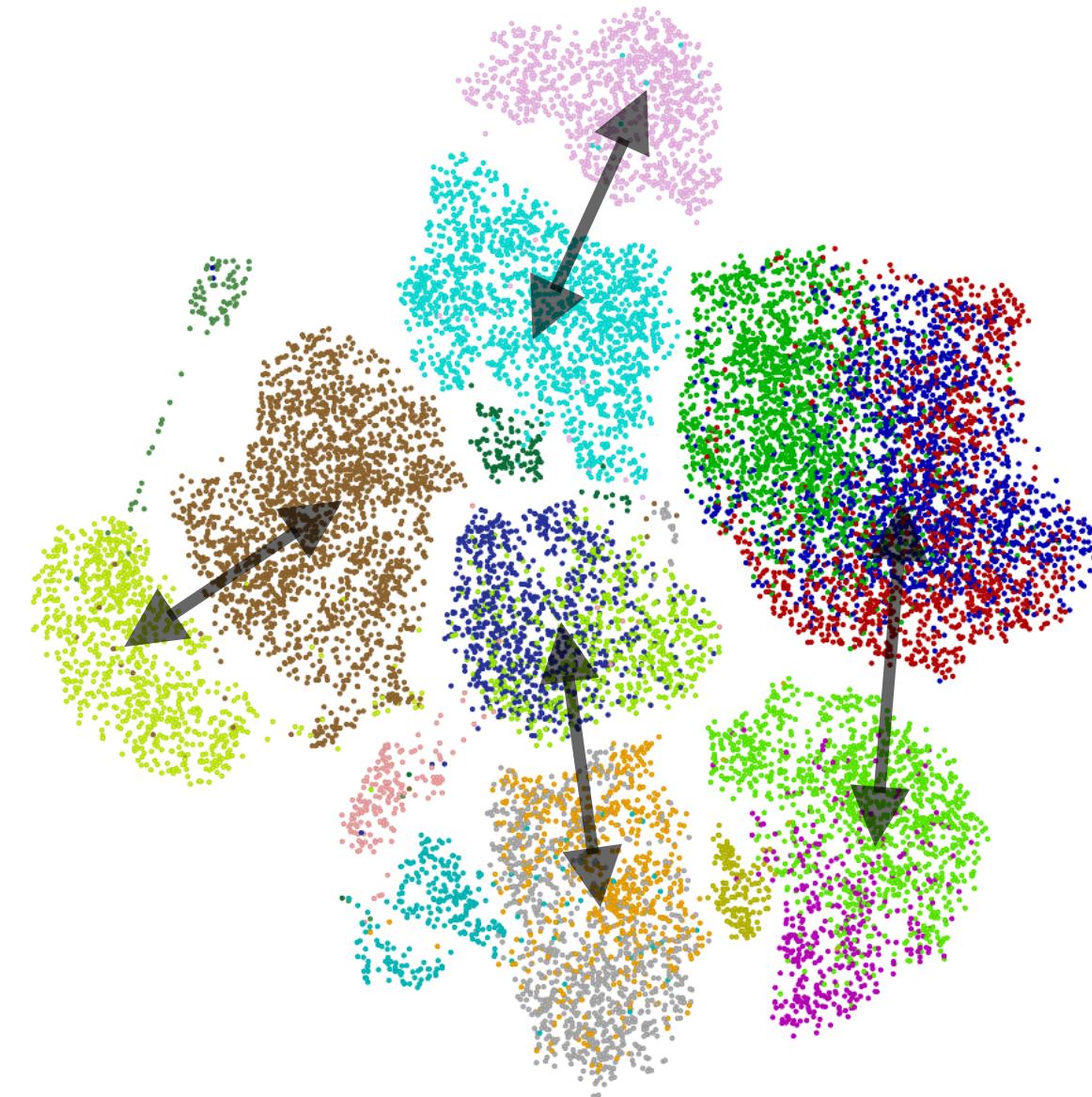


# NOTCH3-IC forces ADRN cell line towards MES state (Gex and SE)

a



# Single cell analysis to understand the 2 cell types



# Acknowledgements

## R2 Development/Concepts/Support

Romeo Willinge Prins

Danny Zwijnenburg

Richard Volckmann

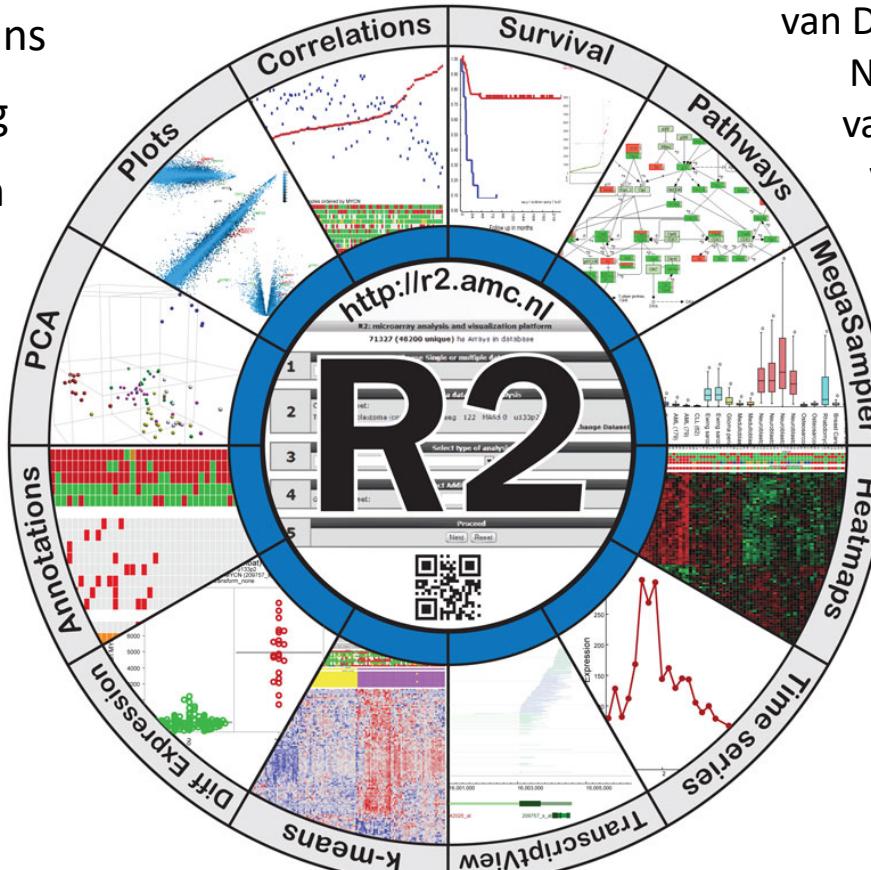
Christian Griffioen

Kris von Stedingk

Piet Molenaar

Lieke Hoyng

**Jan Koster**



## Wetlab & Collaborators

van Dijk-Kerkhoven Lianne  
Nowakowska Natalia E.  
van Kampen Antoine H.  
van Noesel Carel J. M.  
Broekmans Marloes  
Haneveld Franciska  
van Groningen Tim  
Valentijn Linda J  
Akogul Nurdan  
Hasselt Nancy E  
Bras Johannes  
Jongejan Aldo

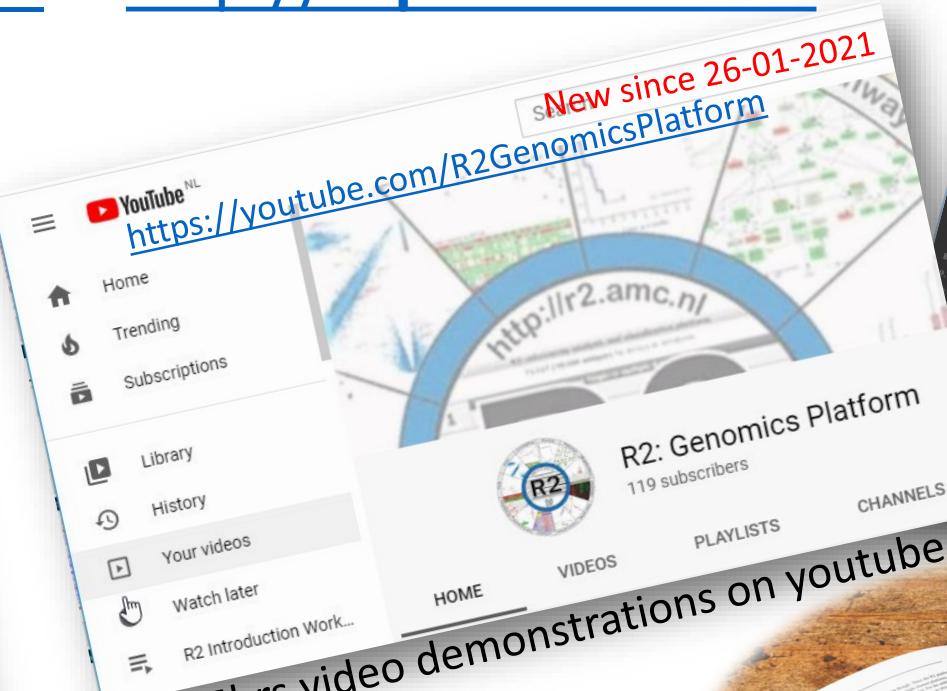
Westerhout Ellen M.  
Hamdi Mohamed  
van Sluis Peter G.  
Ebus Marli E.  
Stroeken Peter  
Koster Linda  
Baas Frank  
Santo Evan  
Schild Linda  
Chan Alvin  
Ora Ingrid  
van der Ploeg, Ida

Huizer-Smit Margriet  
Lecca Maria C.  
Lakeman Arjan  
Van Arkel Jennemiek  
Eleveld Thomas  
Molenaar Jan J.  
Caron Huib N.  
Tytgat Godelieve A.  
Westerman Bart A.  
van Nes Johan  
**Versteeg Rogier**



# Interested in R2?

- Visit us via <http://r2.amc.nl> or <http://r2platform.com>



[R2-support@amc.nl](mailto:R2-support@amc.nl)

