Single-cell Genome-and-Transcriptome (Gtag&T) sequencing without upfront whole-genome amplification reveals cell state plasticity of melanoma subclones

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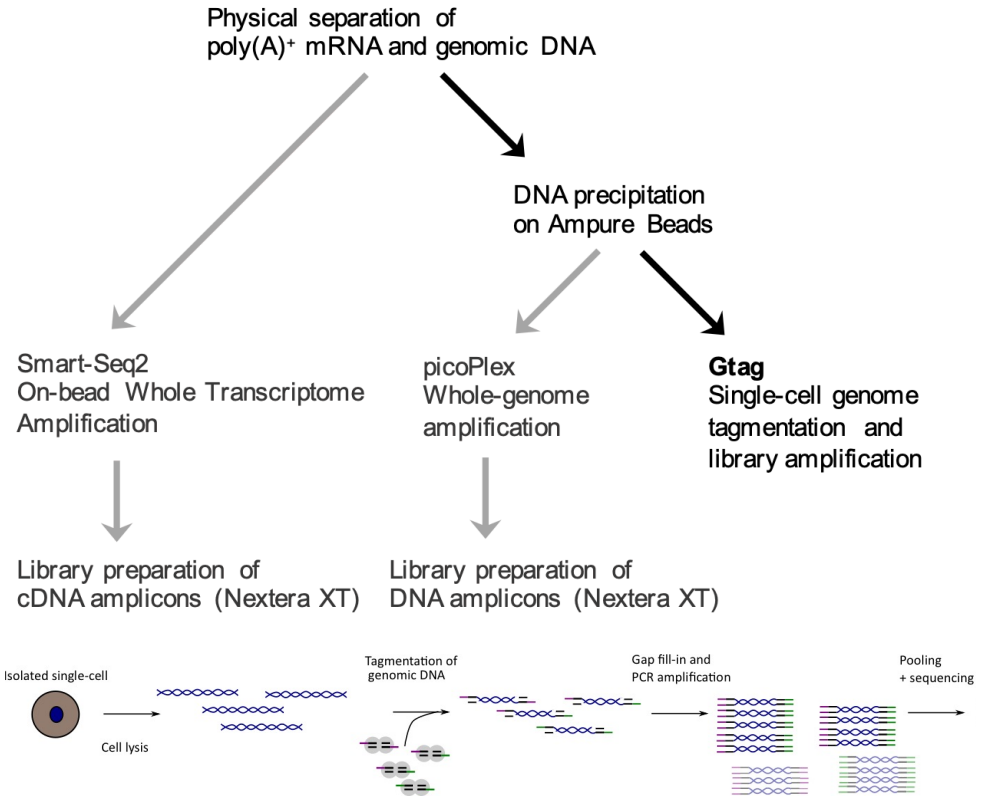
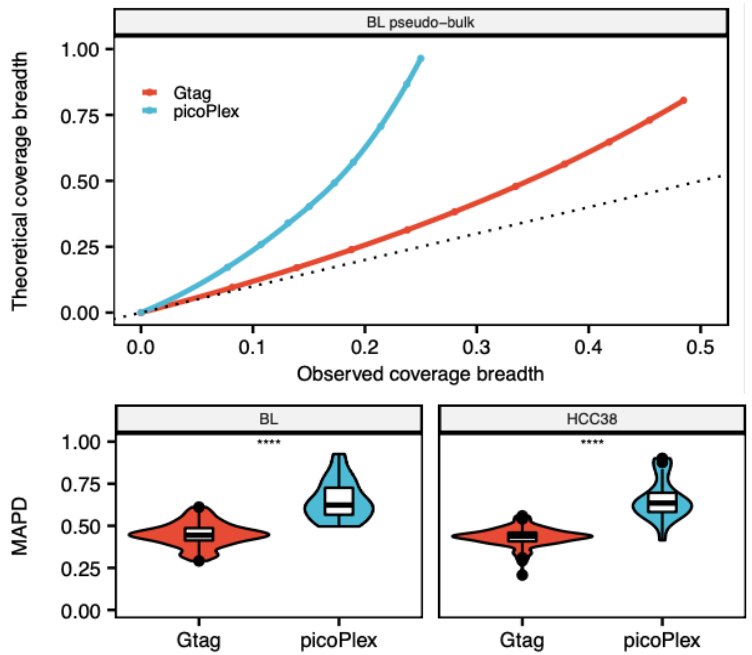
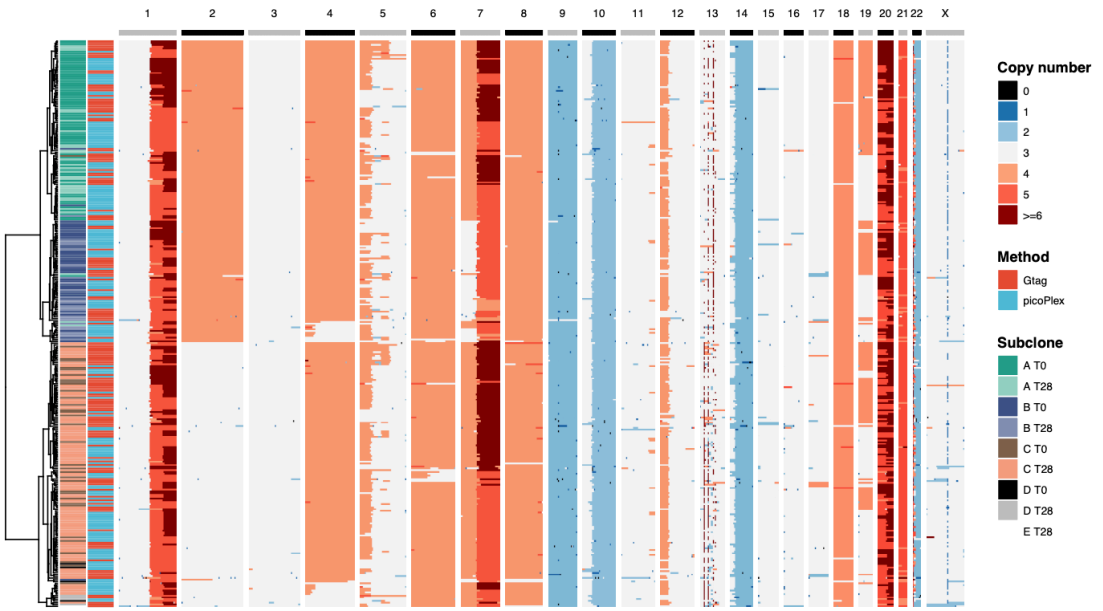
Gtag: a method for direct library preparation of single cell genomes without upfront whole-genome amplification (WGA)

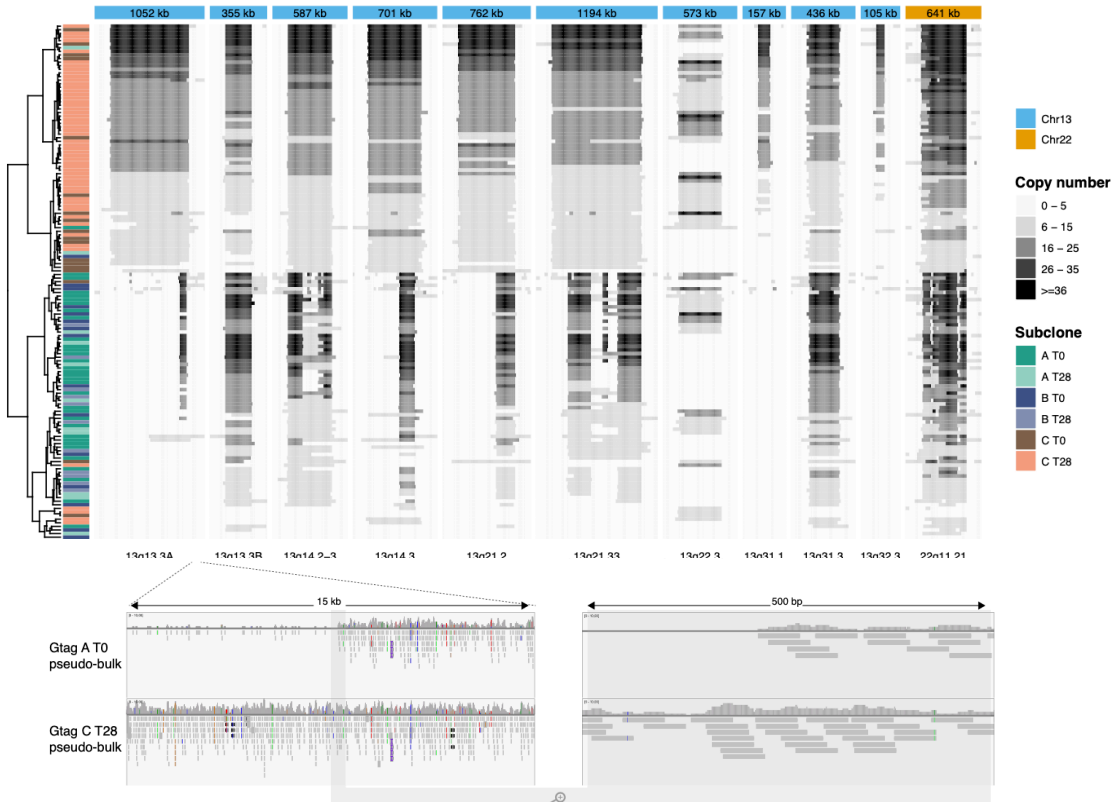


Gtag&T reduces the cost (3X) in comparison to G&T, while increasing breadth of coverage and reducing noise



Profiling of a Melanoma PDX model using G(tag)&T before treatment and at minimal residual disease reveals the presence of 3 subclones and focal amplicons at chromosome 13 and 22

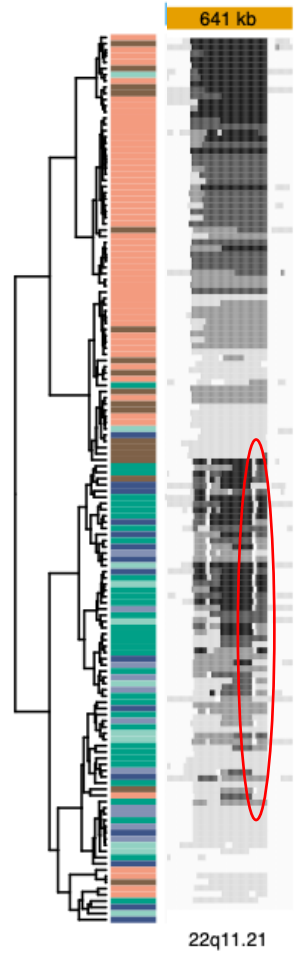




- *0.006x coverage per single cell (= 400,000 mapping reads)*
- *Copy-number calling in bins of 500.000 mappable positions*
- *Presence of 3 subclones*
- *Focal amplicons on chromosome 13 and 22*

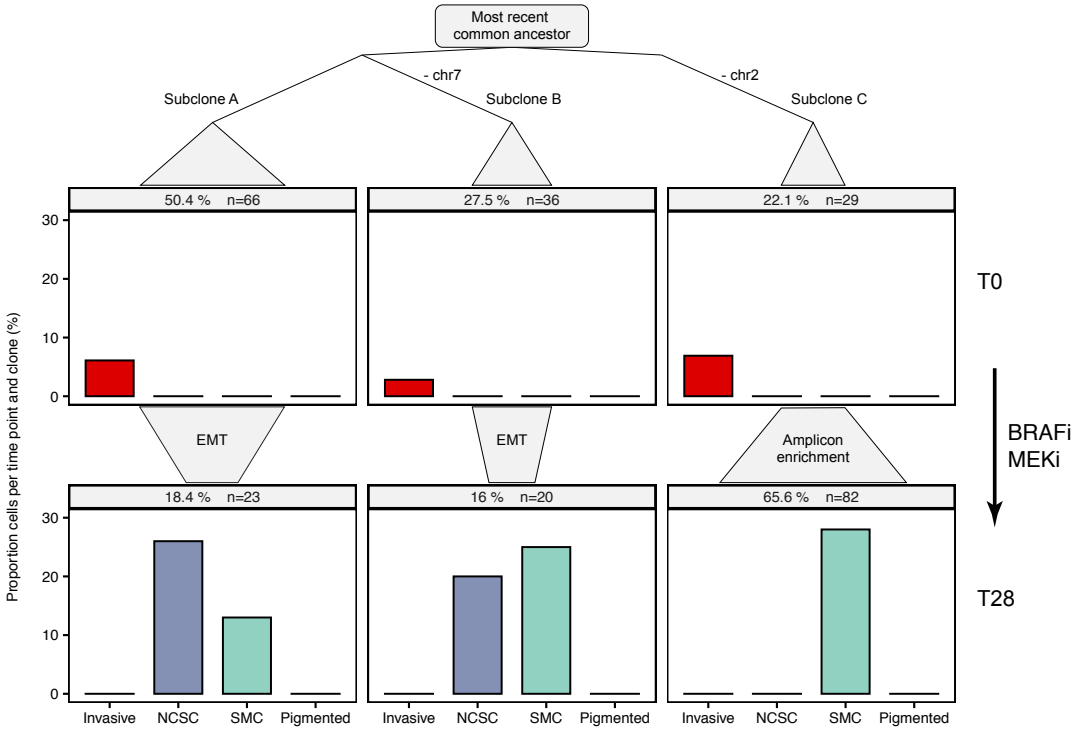
- *Focal amplicon heterogeneity (10.000 mappable positions):*
  - *Presence*
  - *Copy-number*
  - *Size*
- *Breakpoint detection at near-basepair resolution*

22q11.21 amplicon shows clear gene dosage effects and is reversely correlated with expression of pigmentation markers



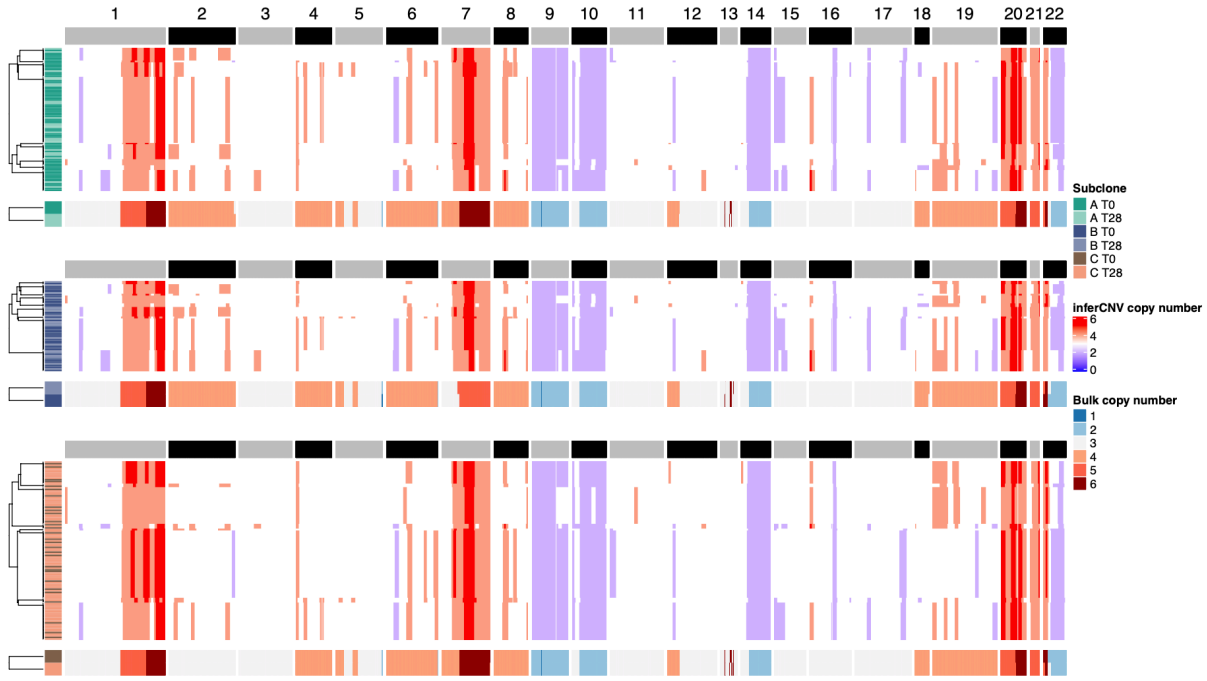
- *Chr13 amplicons: gene with (sub)clonal gene dosage effect or no effect at all*
- *22q11.21 amplicon: majority of genes have clonal gene dosage effect*
  - *LZTR1 & THAP7 are located on segment with lower copy-number in subclones A & B*
  - *PIK4A: subclonal effect due to epigenomic effect*

Phylogenetic reconstruction of tumour evolution with transcriptomic information at single-cell level with Gtag&T



- *Cell states of Rambow et. al (2018) were assigned based on the transcriptome (Invasive, NCSC, SMC & Pigmented)*
  - ✓ *Absence of NCSC state in subclone C*
  - ✓ *SMC & Invasive state present in all subclones*
- *Enrichment of subclone C at MRD*
- *Enrichment of amplicons in subclone C at MRD*

Limited resolution and accuracy of transcriptome-based DNA copy number inference methods



- *InferCNV was used to obtain CNAs based on the transcriptome information and compared with the scDNA data from G(tag)&T*
  - ✓ *Sensitivity: 48%*
  - ✓ *Specificity: 90%*