Development of novel spatial genomics approaches to visualize mutant clones in normal human tissues

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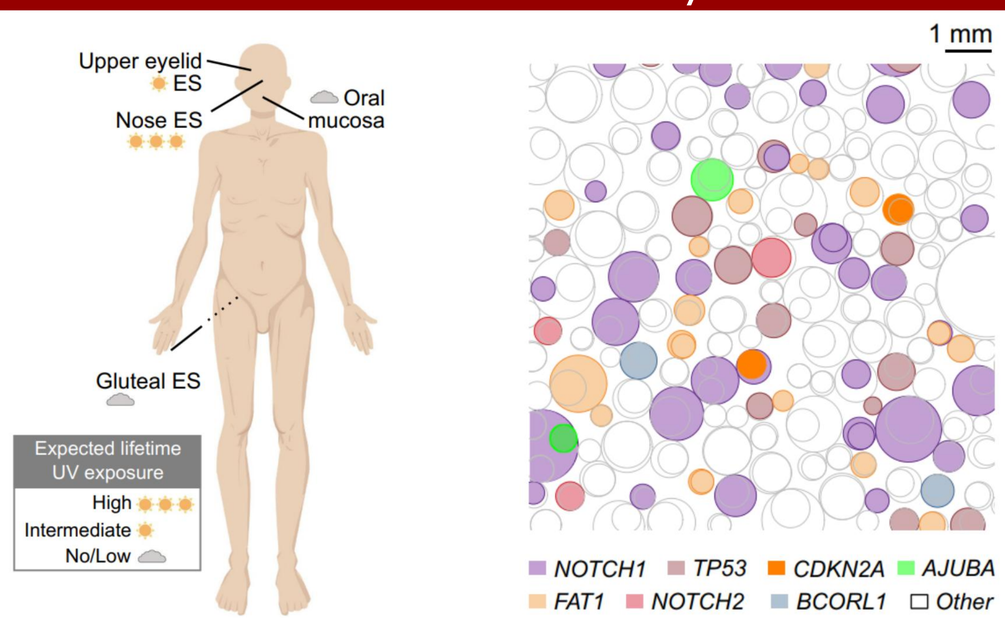
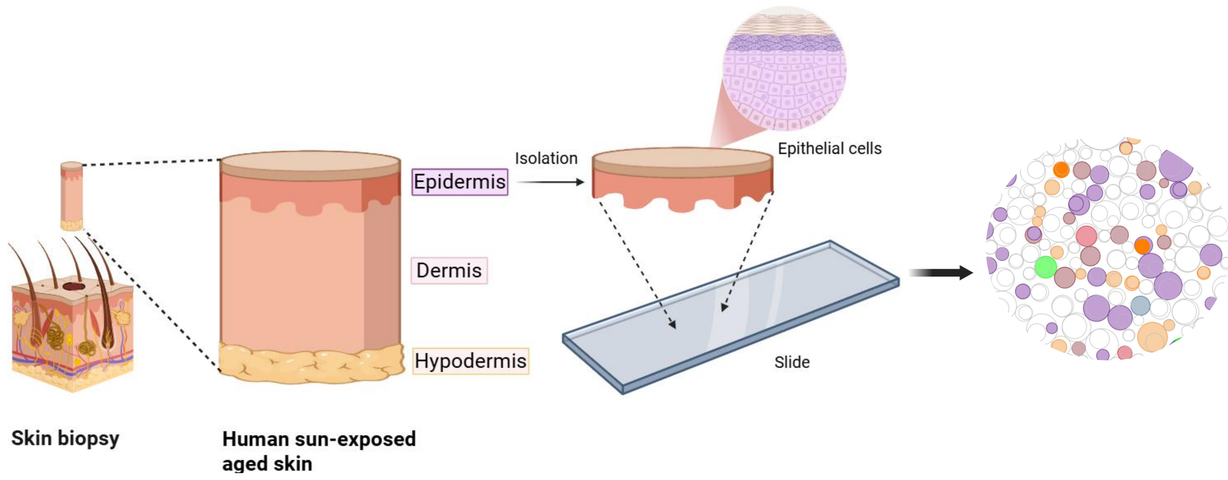
Mutant clones in healthy tissue



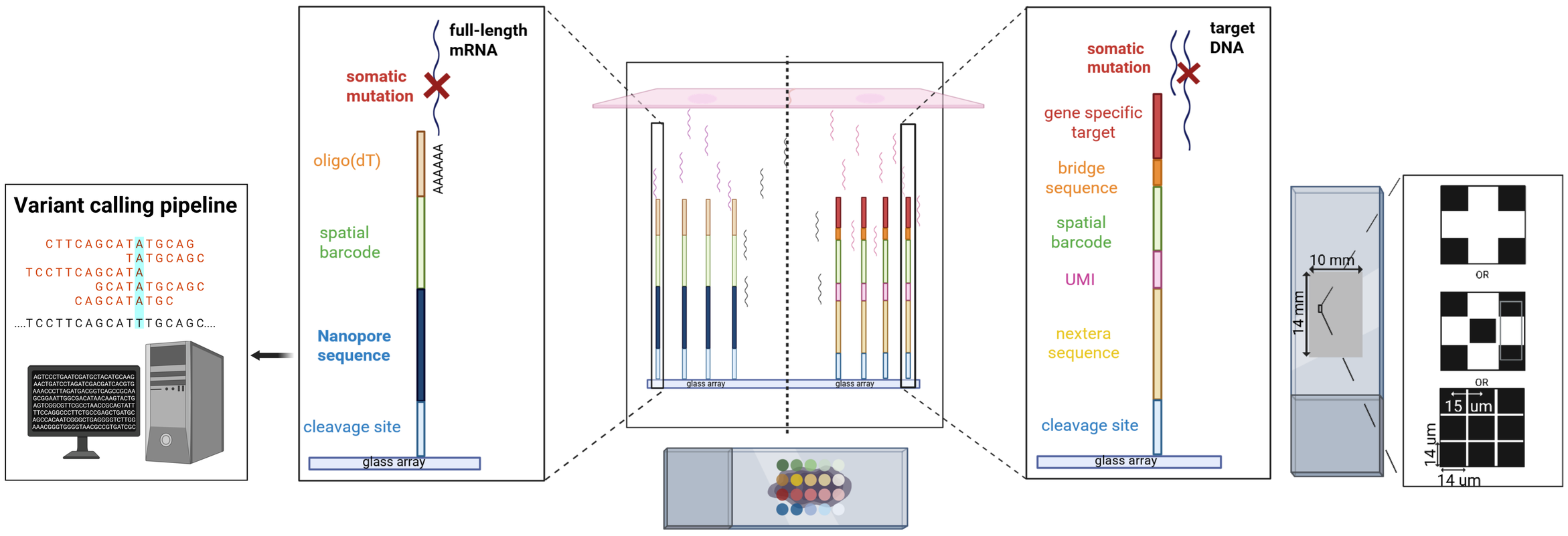
Clonal alterations in healthy UV-exposed skin: Expansion of clones is driven by somatic mutations in known cancer genes such as *TP53 and NOTCH1.*

Spatial profiling of longitudinal skin sections



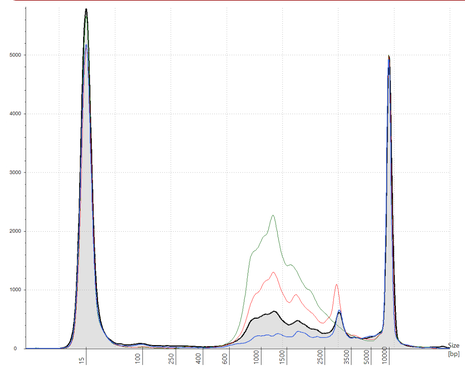
Longitudinal skin sections: Skin punch biopsies are obtained and the epidermal layer is isolated. These will then be transferred onto the in-house glass slides to visualize the mutational landscape.

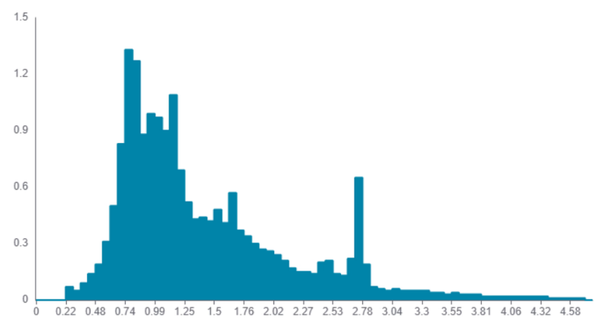
In-house spatial genomics array design strategies for mRNA and DNA targeting



Microarray design: Two array designs are explored to enable spatial genomics. In a first approach, the probes are designed to capture mRNA transcripts. Through a long-read library preparation strategy, full-length cDNA is sequenced using Oxford Nanopore Technology, followed by a variant calling pipeline (left). In a second approach, the probes are modified to target a gene region of interest and libraries are amplified using Illumina-short read technology (right). The platform features spot sizes of 14 x 14 µm, each spot containing 2-5 million capture probes. The spots can be printed in full design, 1/2 (checkerboard), or 1/4 density.

In-solution library prep – *TP53* enrichment





***TP53*-enriched long-read sequencing libraries:** libraries were generated using the cDNA-PCR barcoding kit modified with an in-house enrichment workflow for *TP53*. sequenced on ONT MinION MK1b.

Future perspectives

- **enriched library prep for other targets of interest**
- **DNA-targeted approach**
- **Optimization on-slide protocol**

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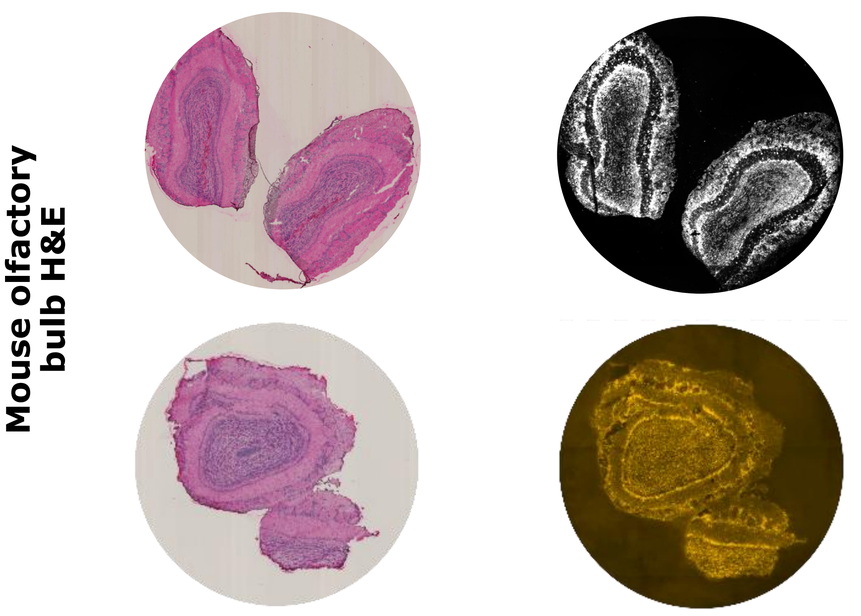
On-slide fluorescent cDNA footprinting



Fluorescent cDNA footprints: footprints from olfactory bulb tissue were generated by incorporating Cy3-dCTP into the reverse transcription (RT) reaction mix. The top image was obtained using the in-house workflow, the bottom with Visium 10X Genomics reagents.

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