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TISSUE INCUBATOR FOR REDUCED AUTOFLUORESCENCE SIGNALS

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PROBLEM & SOLUTION

NanoString's spatial molecular imaging (SMI) technology uses fluorescent probes to identify and quantify tissue transcriptomes[1]. The current output signal is obstructed by tissue autofluorescence in formalin-fixed, paraffin-embedded (FFPE) tissue. To increase signal-to-noise ratio and SMI efficacy, **we created a device to photobleach tissue samples** prior to fluorescent readout. By enabling improved SMI processing, we aim to gain deeper insights into disease progression and promote biomedical innovation.

BACKGROUND

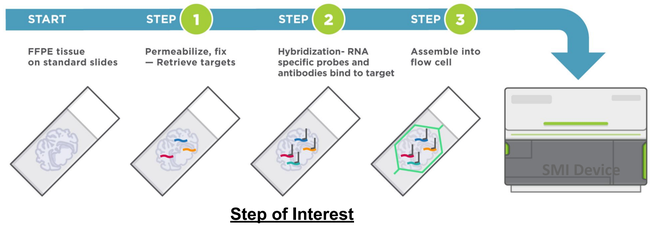


Figure 1. SMI Workflow[2]. Tissue bleaching will occur during the in-situ hybridization probe step.

DEVICE DESIGN

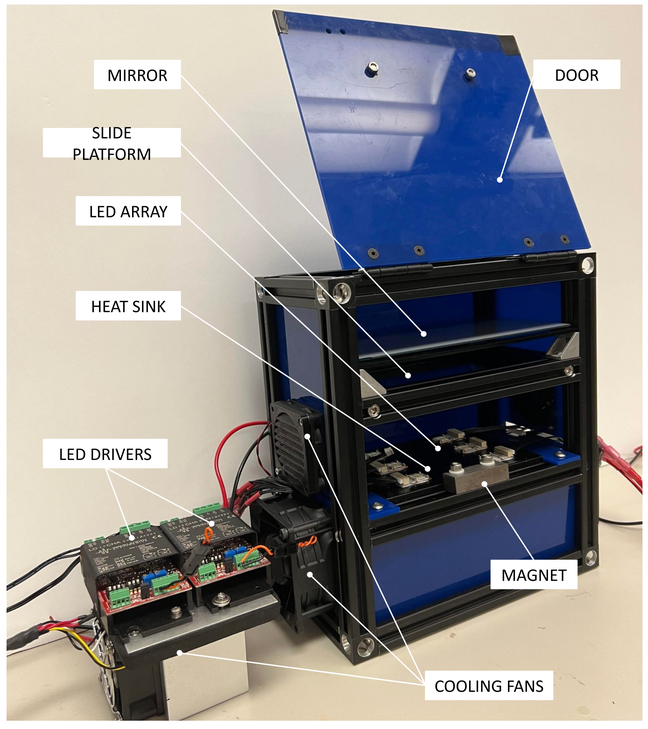


Figure 2. Device module and key components. The main components include an LED array for illumination, 4 fans for cooling, a heat sink, and LED drivers for electrical safety.

RESULTS

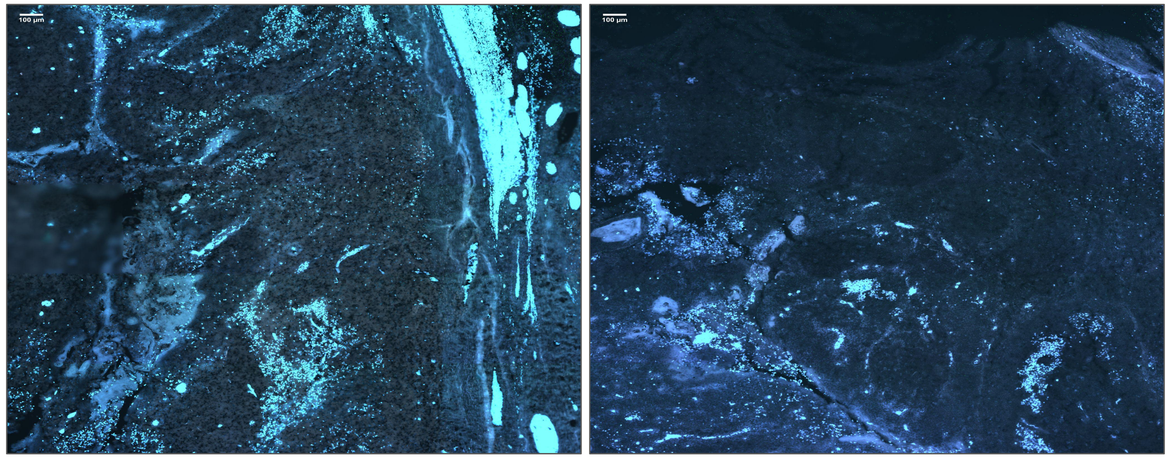


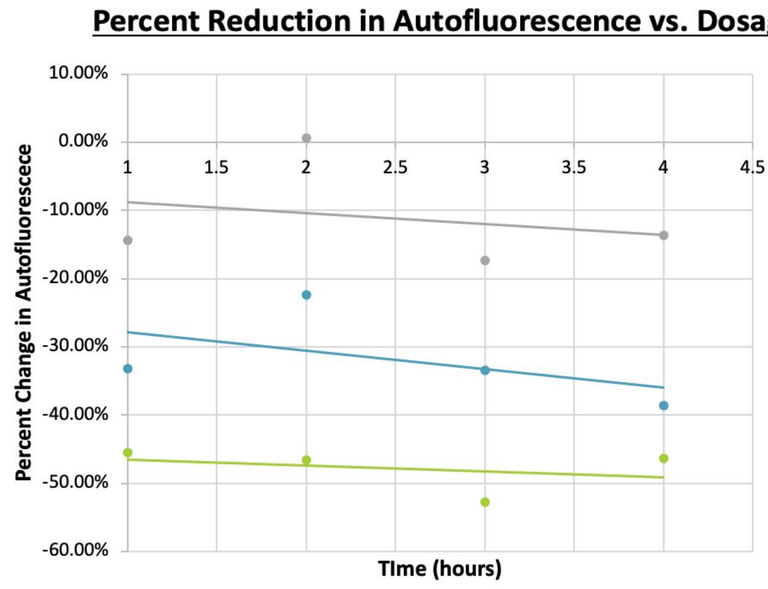
Figure 3. Tonsil tissues before and after photobleaching.

- The tissue was imaged (left) then subjected to **1 hour** of photobleaching before being imaged again (right).
- **The mean intensity of light after bleaching decreased by 67%, 40%, and 15%** in the AF532, FITC, and DAPI channels, demonstrating a notable reduction of autofluorescence.

Figure 4. Reduction of tissue autofluorescence with light dosage.

- Light dosage was determined by exposure time (1-4 hours). Each tissue sample was imaged in three channels of light - AF532, FITC, and DAPI.
- As the duration increased, the dosage of light delivered increased and the percent change in intensity from the tissues became more negative.

There is a direct relationship between autofluorescence reduction and light dosage.



Time (Hours)	Dose (kJ/cm²)
1	0.43
2	0.86
3*	2.26
4*	3

*Included mirror which increases dose by 1.75 x

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REFERENCES

1. He, Shanshan, et al. (2022). High-Plex Multiomic Analysis in FFPE Tissue at Single-Cellular and Subcellular Resolution by Spatial Molecular Imaging. *biorxiv*. https://doi.org/10.1101/2021.11.03.467020

2. NanoString. (2022, April 8). CosMx SMI overview - single-cell imaging. NanoString. Retrieved May 9, 2022, from https://nanostring.com/products/cosmx-spatial-molecular-imager/overview/