HistoProbe: A Dual-Mode Thermally Assisted Microfluidic Platform for Direct Mass Spectrometry Analysis of FFPE Tissues

Formalin-fixed, paraffin-embedded (FFPE) tissue sections are widely used in biomedical research and diagnostics, but traditional methods for removing paraffin are slow, solvent-intensive, and can lead to loss of valuable chemical information. To address these challenges, we developed HistoProbe, a dual-mode microfluidic platform designed to streamline tissue preparation and analysis by mass spectrometry (MS) and mass spectrometry imaging (MSI).

HistoProbe integrates a modified liquid microjunction probe with a precisely controlled heating system adapted from a 3D printer. This design enables localized thermal deparaffinization directly at the point of analysis, eliminating the need for toxic solvents such as xylene. In its Online mode, HistoProbe allows rapid, direct MS analysis of tissue sections with no sample preparation, supporting high-throughput metabolic profiling (>80 samples per day). In its Offline mode, the platform performs automated localized preparation with mild solvents (e.g., ethyl acetate), reducing solvent use by more than 99% while enabling high-resolution MSI.

Both modes successfully distinguished cancerous from non-cancerous regions in melanoma tissues and revealed spatially distinct distributions of biologically important lipids and free fatty acids. Compared to conventional workflows, HistoProbe reduces preparation time by more than 90%, enhances reproducibility, and substantially lowers environmental impact.

By combining speed, sensitivity, and sustainability, HistoProbe demonstrates how innovative microfluidic and thermal control strategies can modernize tissue analysis for MS-based histopathology, with broad implications for both research and clinical chemistry.