

macroMS: Image-Guided Analysis of Random Objects by Matrix-Assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry

Background/objective

Previously, high-throughput chemical characterization approaches were created to enable the rapid screening of thousands of microbial colonies and liquid droplet arrays for desired products using matrix-assisted laser desorption/ionization (MALDI)-time-of-flight (ToF)-mass spectrometry (MS). Here, we demonstrated an approach that acquires mass spectra only from desired locations using a web platform (macroMS), which enables the use of commonly available imaging tools for optically guided mass spectrometry analysis of large numbers of arbitrary located macroscopic samples.

Approach

- ❖ Created an image-guided MALDI-ToF-MS workflow for sampling of either ordered or randomly located >300 μm objects located on a surface.
- ❖ Demonstrated the method using a standard office flatbed scanner and cell phone camera to obtain images and used new optical correction tools to resolve optical defects inherent to common imaging devices, including correction of moderate distortion and imperfect perspective.
- ❖ Validated the approach by screening medium-chain fatty acid (MCFA)-producing variants from a site saturation mutant (SSM) library for site Gly 1250 in the ketoacyl synthase domain of fatty acid synthase (FASII) of *Saccharomyces cerevisiae*.

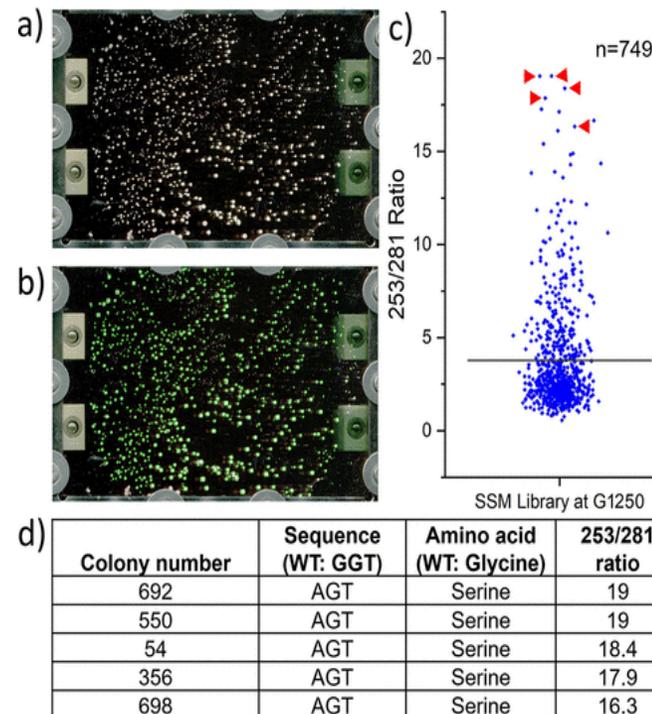
Results

- ❖ Nanoliter droplet samples of bacterial cultures (300 μm width) were targeted reliably, revealing a consistent ratio between two peaks for identifying fatty acyl fragments, and the upper limit for the number of targets is about 10,000.
- ❖ Provides data analysis followed by data visualization tools through a web platform.

Significance

macroMS-based screening of enzyme mutant libraries is effective when testing imprinted microbial colonies and provides a high-throughput pipeline for completing MS screens to search for specific compounds, a promising method for testing colonies in biofuel research and microbial identification.

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Yeast colony screening for G1250 site saturation mutagenesis library. (a) A scanned image of a 50 x 75 mm ITO slide with imprinted *S. cerevisiae* colonies. (b) Result of target finding by macroMS. Green marks are the found targets. (c) Scatter plot of the per colony ratio of C16:1 fatty acid over C18:1 fatty acid. The red arrows indicate colonies selected for sequencing. (d) DNA sequencing showed all colonies had the expected G1250S genotype.