136. Development of a High Throughput Platform for Screening GH Enzymes and Methyl Ketones based on Oxime-NIMS (Nanostructure-Initiator Mass Spectrometry)

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Project Goal: Our goal is to develop a high throughput platform to study the specificity and activity of large-scale GH enzymes and biofuel strains.

Abstract: Cost effective conversion of biomass into sugars for biofuel production requires high-performance low-cost enzyme cocktails that are active under the process conditions and biofuel producing strains with high-titers of the desired products.

Development of the requisite enzymes and strains requires large-scale studies of specificity and activity. However, the existing high throughput methods (e.g. DNS assays) lack the specificity needed to understand enzyme kinetics, which is crucial to identify bottle-neck step during the biomass deconstruction and to develop the required enzyme cocktails.

Here we describe a high throughput screening platform for glycosyl hydrolysases (GHs) activities and fatty acid methyl ketones that is based on bioconjugation chemistry and nanostructure-initiator mass spectrometry. The bioconjugation involves the oxime formation between a unique chemical probe and aldehyde (glycans) and ketone groups. The quantitative analysis is enabled by incorporation of C13 labeled internal standards. The key to the high throughput analysis is the novel integration of nanoliter-scale acoustic sample deposition with nanostructure-initiator mass spectrometry analysis (R&D100). This allows thousands of samples to be printed on a single mass spectrometry chip for rapid laser-desorption based analysis.

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