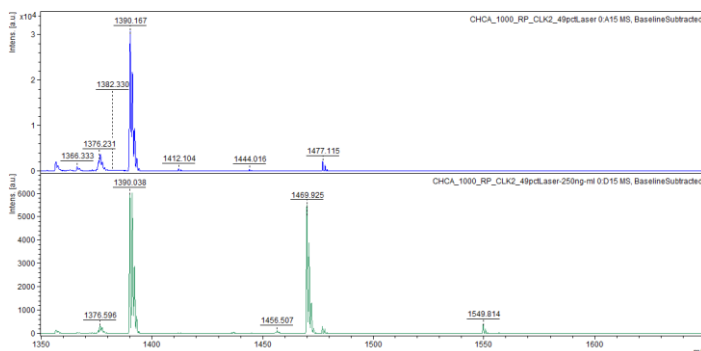


## Application Note: Label-free Biochemical screening

The use of mass spectrometry in early drug discovery for the development of label-free assays has steadily grown in the past 10 years as it minimizes data artifacts routinely observed with labeled, indirect or coupled assay systems. Additionally, recent advances in MS instruments have allowed sample throughput to approach HTS levels, ~100,000/day. **Revolution Biosciences** has developed robust label-free assays for compound screening using MALDI mass spectrometry involving protein/peptide modifications capable of Hit & Lead Optimization as well as primary screening.

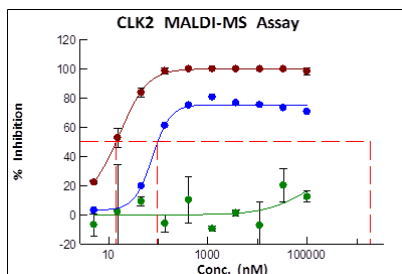
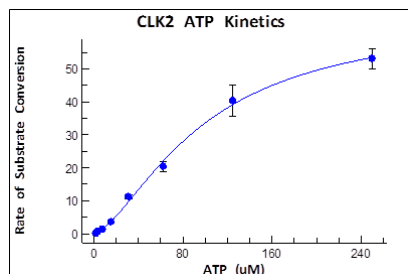


Mass Spectra for CLK2-treated SRSF1-derived peptide with & without ATP.

### CLK2 MALDI-MS Assay

As a validation of our label-free MALDI-MS approach we developed a CLK2 assay for phosphorylation of an SRSF1-derived peptide. The figure above demonstrates the assay can detect the multiple phosphorylations of the peptide consistent with the hyper-phosphorylation of SR proteins involved in alternative

splicing. The left plot demonstrates the assay obeys Michaelis-Menten kinetics while the right demonstrates dose response testing. The 384-well format allows testing from hundreds of compounds in dose response to tens of thousands in primary screening per run.



### About Us

• • •

Revolution Biosciences' collaborative R&D discovery model leverages the power of chemical biology to discover and develop transformative medicines in partnership with the life science community by enabling access to cutting edge technologies (MALDI-MS, High Content Image Screening & Differential Scanning Fluorimetry) through consulting agreements, collaborative partnerships and contract research.

We develop new bioanalytical methods to directly identify proteins, peptides and small molecules and their molecular interactions in all types of biological samples. Our small molecule drug discovery expertise and application of advanced proteomics technologies accelerates drug discovery research and our state of the art bioanalytical laboratory on the UMass Boston campus is equipped to handle bioanalytical studies from assay development through high throughput screening.