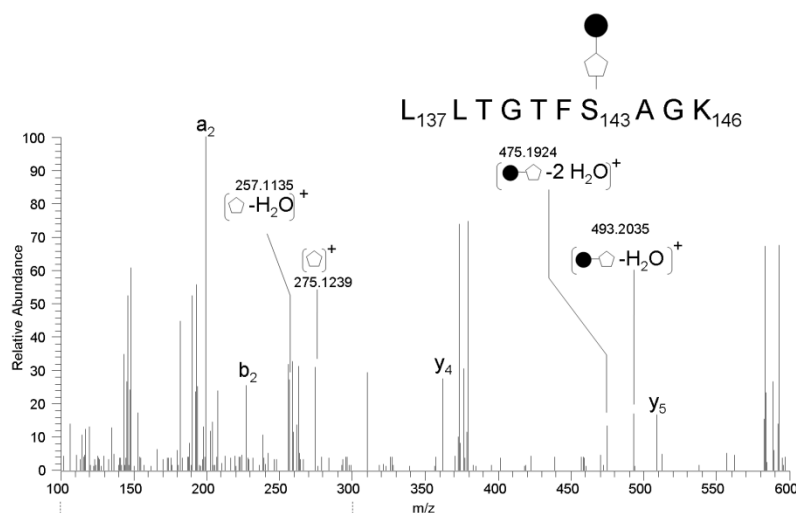
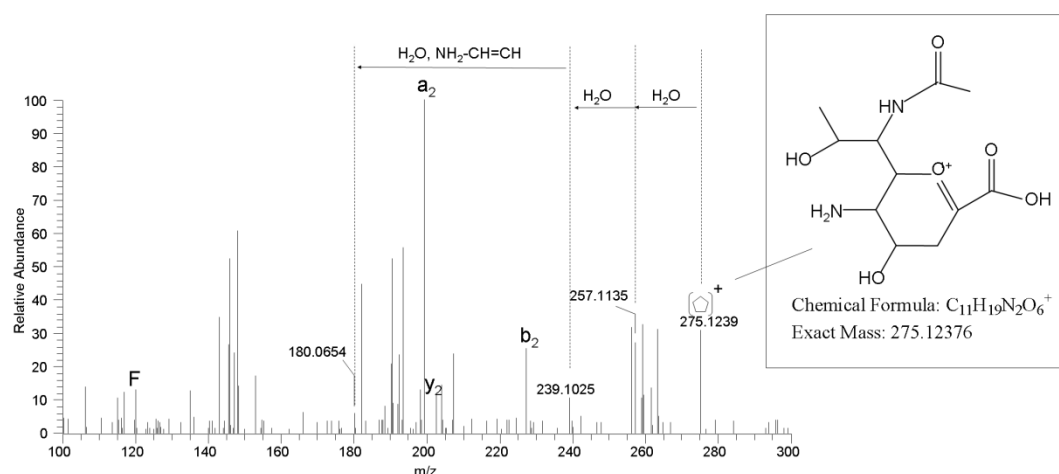


A HCD-MS/MS of m/z 752.89, +2



B



Supplemental Figure 4: HCD-MS/MS spectrum from L₁₃₇LTGTFS@AGK₁₄₆ plus a 510 Da modification (@represents the site of modification). The HCD-MS/MS spectrum showed the accurate mass of glycan-related ions corresponding to dehydrated ions of the 510 Da modification, and an oxonium ion from the 274 Da moiety of the 510 Da modification (A). Figure 3 B represents the zoomed HCD spectrum between m/z 100 to 300. It should be noted that the signals observed from the peptide with 510 Da were very low compared to those with 524 Da or 538 Da modifications, due to low abundance of the 510-Da modification occurring in the flagellin. However, we detected main fragment ions from 274-Da modification. An inset in the spectrum B shows a proposed structure of the 274-Da moiety (observed m/z of 274 Da moiety as an oxonium ion is m/z 275). The pentagon cartoon and black-colored circle cartoons in the figure represent the 274 Da- and 236 modifications, respectively.