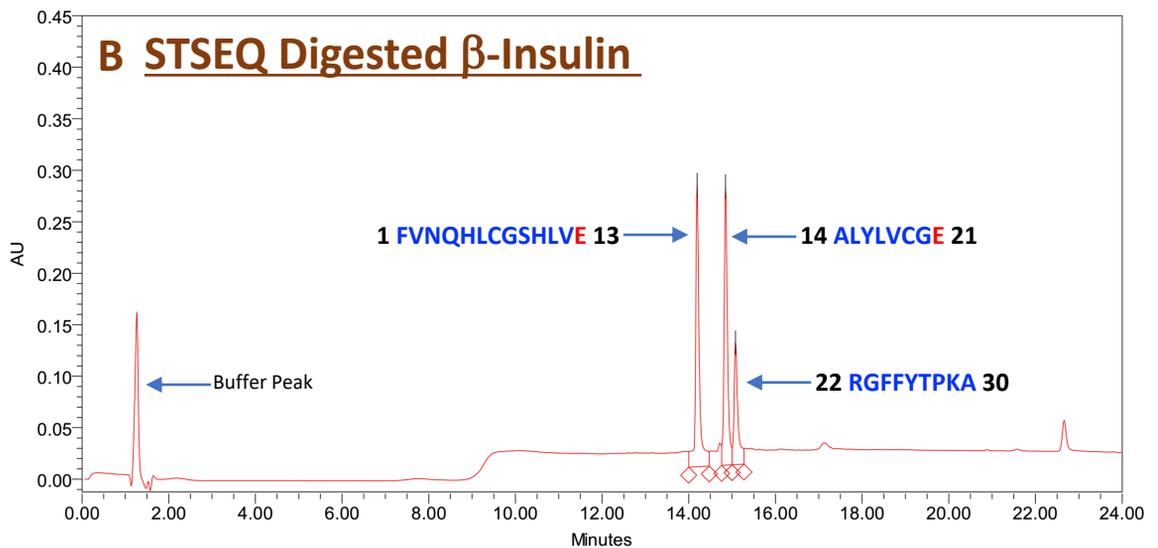
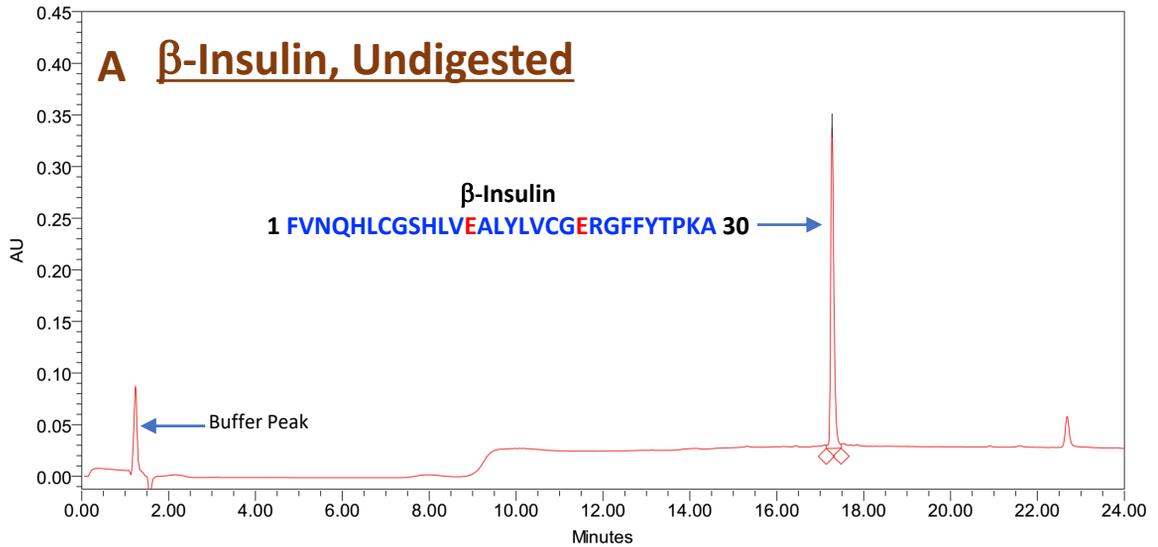


Reverse Phase HPLC of Undigested and Endoproteinase Glu-C (STSEQ) Digested β -Insulin



50 μ g β -insulin was incubated with buffer (Panel A) or 2.5 μ gP STSEQ (~4.5 units) (Panel B) in a total volume of 152.5 μ l containing 50 mM potassium phosphate, pH 8.0 for 2 hours at 37°C and resolved by reverse phase HPLC on a C18 column.

Endoproteinase Glu-C from *Staphylococcus aureus* specifically cleaves peptide bonds on the COOH-terminal side of either aspartate (D) or glutamate (E) residues in polypeptides.

β -Insulin is a 30 amino acid peptide containing two glutamate residues at positions 13 and 21 and elutes as a single peak at 17.2 minutes (Panel A). Digestion with STSEQ for 2 hours results in complete digestion at E13 and E21 generating three smaller peptides, 13, 8 and 9 amino acids in length, that elute at 14.2, 14.8 and 15.0 minutes, respectively (Panel B).