In-gel trypsin digestion reagents and protocol

* Prepare reagents as per Shevchenko, et al. (2006) Nature Protocols 1: 2856-2860.
* 50 mL 0.1M ammonium bicarbonate (79 g/mol): 0.40 g / 50 mL
* 1 mL of 10 mM DTT ( 1.54 mg/mL 0.1M amm bicarb)

55mM iodoacetamide (10.2 mg /mL 0.1M amm bicarb)

* Trypsin - need <=50 uL solution per gel slice
* Stock = Promega V5113, 0.5 mg/mL
* Trypsin working solution per sample:
* 50 uL of 10 ng/uL trypsin in 10mM amm bicarb / 10% acetonitrile

Add 1 uL stock trypsin to 50 uL working solution and cover gel slices.

* Incubate 37 C for 1 hour to overnight.
* Remove solution from gel slices by pipettor and store at -20 C.
* Zip tip treatment not usually necessary, but may be done to increase peptide concentration.