

## Example Top-Down ECD Fragmentation Datasets (Standards)

Notes – July 2016

**Equine Myoglobin** – Uniprot code: P68082

Sequence:

GLSDGEWQQ VLNWVGKVEA DIAGHGQEV LIRLFTGHPET LEKFDKFKHL KTEAEMKASE DLKKHGTVVL  
TALGGILKKK GHHEAELKPL AQSHATKHKI PIKYLEFISD AIIHVLHSHK PGDFGADAQG AMTKALELFR  
NDIAAKYKELGFQG

- PTMs - Loss of N-terminal Met
- Selected the  $[M+18H]^{18+}$  precursor for ECD fragmentation.
- Analysed at 5uM in 50:50:0.1 Water:MeOH:Formic Acid.

**Bovine cytochrome c** – Uniprot code: P62894

Sequence:

GDVEKGGKIFVQKCAQCHTVEKGGKHKHTGPNLHGLFGRKTGQAPGFSYTDANKNKGITWGEETLMEYLENPKKYI  
PGTKMIFAGIKKKGEREDLIAYLKKATNE

- PTMs - N-terminal acetylation (increase in monoisotopic mass of 42.01057)
  - Covalently bound to a single heme (increase in monoisotopic mass of 616.177307) at C14 and C17
- Selected the  $[M+12H]^{12+}$  precursor for ECD fragmentation.
- Analysed at 5uM in 50:50:0.1 Water:MeOH:Formic Acid.

**Yeast Alcohol dehydrogenase** – Uniprot code: P00330

Sequence:

SIPETQKGV IFYESHGKLE YKDIPVPPK ANELLINVKY SGVCHTDLHA WHGDWPLPVK LPLVGGHEGA  
GVVVGMGENV KGWKIGDYAG IKWLNAGSCMA CEYCELGNES NCPHADLSGY THDGSFQQYA TADAVQAAHI  
PQGTDLAQVA PILCAGITVY KALKSANLMA GHWVAISGAA GGLGSLAVQY AKAMGYRVLG IDGGEGKEEL  
FRSIGGEVFI DFTKEKDIVG AVLKATDGGG HGVINVSSE AAIEASTRYV RANGTTVLVG MPAGAKCCSD  
VFNQVVKIS IVGSYVGNRA DTREALDFFA RGLVKSPIKV VGLSTLPEIY EKMEKGQIVG RYVVDTSK

- Analysed under native MS conditions (100 mM ammonium acetate buffer) of yeast alcohol dehydrogenase 1, which forms a tetramer with a mass of ~147 kDa.
- The N-terminus methionine (M1) has been removed, and the serine at residue 2 (S2) has been acetylated. A zinc ion may be non-covalently bound to residues C44 and H67. A second zinc ion may be bound to residues C98, C101, C103 and C112.