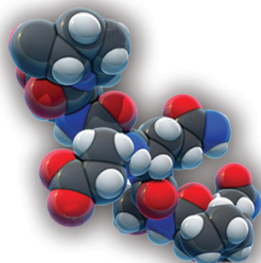


Bioinformatics and In-Silico Modelling

- Secondary Structure
- pI, Hydropathy Score

Stability Study

- Temperature
- Buffers, pH
- Concentration
- Agitation Study
- Forced Degradation



PEPTIDE

Analysis

- RP-HPLC, IEC (Purity, Potency)
- DLS, SEC: Aggregation and Partical Size Analysis

Solubility Study

- Buffers, pH Selection

Preformulation Selection

Aqueous Soluble Formulation

- Peptide Concentration
- Accelerated Stability

PREFORMULATION STUDIES:

Case Study: Peptide 1

Mol. Wt 4 kD, 41 residue all natural amino acids

pI. 4.7, GRAVY Score is -0.3

Secondary Structure: 60% Helix, 5%

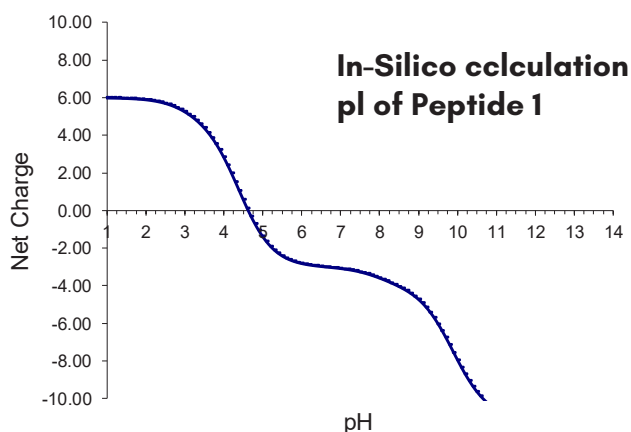
Random Coil and 35% B-Sheet

Solubility 2 mg/ml in PBS

Physical and Chemical Stability Indicators in Peptides and Proteins

- Aggregation, Gelation
- Amino Acid Stability
- Aspartic acid (D) - Shuffling, Hydrolysis near pKa
- Asn (N)-Cyclization, Deamidation at higher pH
- Asn-Glv (N-G)-Deamidation
- Met (M)-Oxidation
- Cys (C)-Aggregation, Disulphide, Oxidation
- Trp (W)-Oxidation

Calculated Net Charge vs pH

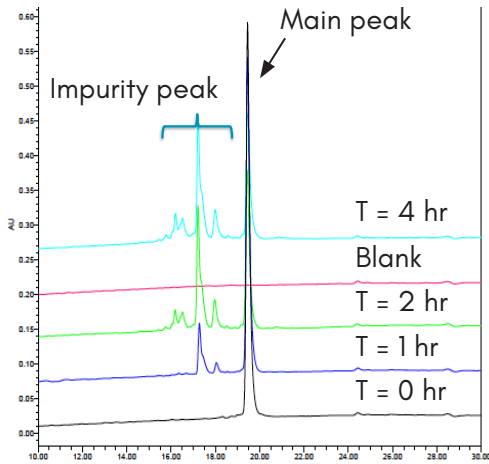


pH	Net Charge
1	5.99
2	5.89
3	5.25
4	2.83
5	-1.38
6	-2.81
7	-3.08
8	-3.59
9	-4.73
10	-7.95
11	-10.58
12	-11.45
13	-11.90

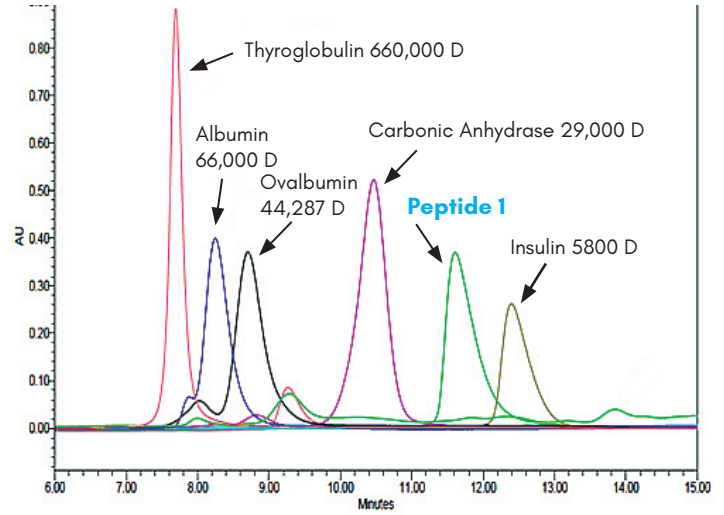
pH vs. net charges of Peptide 1

Peptide 1: Analytical Methods

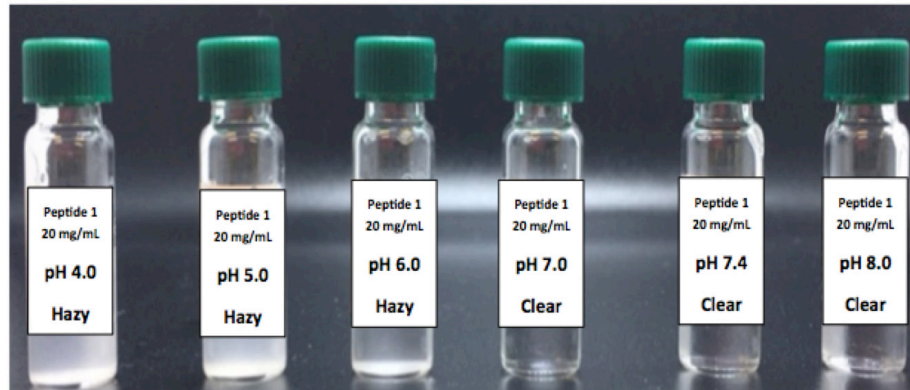
RP-HPLC Stability Indicating Method



Size Exclusion Chromatography Molecular Weight Determination



Solubility vs. pH at 20 mg/mL



Particle Size Analysis by DLS

