Biomatik
Tel: (519) 489-7195, (800) 836-8089
Fax: (519) 231-0140, (877) 221-3515
Email: info@biomatik.com
http://www.biomatik.com

Peptide Handling Guideline

Version 6.4, Revision 2017-07-12

Reconstitution

Proper peptide handling and solubilization is the starting point of a successful bioassay project. If reconstitution becomes a big concern to you, please request a solubility test at the time of ordering.

1. Use a small aliquot of peptide to test the reconstitution conditions:
   Begin by testing a small sample of your peptide in the desired solvent. Once satisfied, apply to the larger aliquot as needed.
   Please note: You may request your peptide to be delivered in several aliquots at the time of ordering.

2. The initial solvent used should be the most appropriate one:
   For example, for a very hydrophobic peptide, it is recommended to dissolve it first in a small volume of organic solvent (such as DMSO or acetonitrile) before applying the aqueous solution. Adding organic solvent to a suspension of hydrophobic peptide in aqueous solution is not likely to help in dissolving. In other words, if an aqueous buffer is being added first to a hydrophobic peptide powder and then the organic solvent afterwards, it is not likely help much in dissolving the peptide properly.

3. Determine the initial solvent:
   For a short peptide which is 5aa or less - try sterile distilled water and it is likely to dissolve.
   For a longer peptide (>5aa), the overall charge of the peptide will determine which initial solvent to use:
   a) Assign a value of -1 to acidic residues: D, E, and the C-terminal free acid(-COOH);
   b) Assign a value of +1 to basic residues: R, K, H, and the N-terminal free amine($-NH_2$);
   c) Hydrophobic uncharged residues: F, I, L, M, V, W, and Y;
   d) Uncharged residues: G, A, S, T, C, N, Q, P, acetyl, and amide.

   Then, calculate the overall charge of the entire peptide:
   a) If the overall charge of the peptide is positive (a basic peptide), try to dissolve the peptide in sterile distilled water first. If water fails, add ~20% acetic acid solution. If the peptide still does not dissolve, add drops of TFA (< 50ul), or use 0.1%TFA/H_2O to solubilize the peptide. Then dilute the peptide solution to the desired concentration.
   b) If the overall charge of the peptide is negative (an acidic peptide), try to dissolve the peptide in sterile distilled water first. If the peptide persists as visible particles, sonication can be tried. If water fails, add NH_4OH (<50ul) or 0.1%NH_4OH drop-wise. Then dilute the peptide solution to the desired concentration. If the peptide contains Cys, do NOT use basic solutions (NH_4OH); use DMF instead.
   c) Peptide whose overall charge is 0 (a neutral peptide). It usually dissolves in organic solvents, such as acetonitrile, methanol, or isopropanol. If this does not dissolve completely:
      1) For peptides that tend to aggregate (due to the hydrophobic interaction), the addition of denaturants, such as 8M urea or 6M guanidine-HCl, may also be required.
      2) For very hydrophobic peptides (containing more than 75% hydrophobic residues), add DMSO drop-wise (use DMF instead for Cys containing peptides), and then dilute the solution with water to the desired concentration.

Storage

Most lyophilized peptides shall be stable at room temperature for a couple of weeks. For long term storage, it is strongly recommended that the peptide is stored in powder form at -20°C, away from strong light, and under dry conditions. Repeated freeze-thaw cycles should be avoided. A peptide solution once prepared should be used as soon as possible. If storage in solution is unavoidable, use sterile buffers at pH 4-6 and store in aliquots at -20°C or lower to prolong the storage life.

The shelf life of peptide solutions is limited, especially for peptides containing cysteine(C), methionine(M), tryptophan(W), asparginine(N), glutamine(Q), or N-terminal glutamic acid(E). For example, a Cys-containing peptide is easily oxidised, especially in basic conditions; some residues are easy to racemise, such as Proline. Avoid DMSO if the peptide contains Met, Cys or Trp, due to sulfoxide or disulfide formation. Peptide stability becomes worse when in a solution, especially at the higher pH (pH>8). We therefore recommend keeping solutions in the range of pH 4-6. It is recommended that peptides containing methionine, cysteine, or tryptophan residues be stored in oxygen-free atmosphere to avoid oxidation. The presence of dithiothreitol (DTT) can be useful in preventing oxidation.