#### **Stability**

All peptides are different but it can generally be said that a peptide is most stable as a dry solid stored at low temperature in the dark. However, since the peptide must be used the following guidelines may prove useful.

Dissolve the minimum amount of peptide practical for the intended use. Keep the solution cold when not in use but, as with all biological material, avoid repeated freeze-thawing cycles.

Solutions of peptide that contain methionine and cysteine are particularly prone to oxidation. Methionine will oxidize to the corresponding 'met-oxide peptide'; which elutes slightly earlier than the parent peptide under reverse-phase conditions. Cysteine will tend to combine with other cysteines to form either intra or inter molecular disulphides; these usually elute slightly later than the parent peptide under reverse-phase conditions.

# **Finally**

Thank you for sourcing your peptide from **Cambridge Research Biochemicals** At CRB we have been making peptides for over thirty year and each one is important to us. We want this peptide to make a valuable contribution to your work, for you to be pleased with it and to want to use us again the next time you require a peptide.

If you have any questions, no matter how trivial it may seem, we'd like to help you. Please e-mail us (including your email address and telephone number) and we'll have one of our experienced peptide chemists contact you straightaway.

Some services mentioned throughout may be chargeable. The information and recommendations given here are, to the best of our knowledge, information and belief, accurate at the time of publication. In all cases, it is the responsibility of the user to determine the applicability of such information or the suitability of any products for their own use.

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#### Safety

This peptide is for use as a research tool only – it must not be used in humans. Standard laboratory practices should be followed when handling this material. The toxicological properties of this material have not been investigated. Appropriate measures should be taken to avoid skin and eye contact, inhalation and ingestion.

#### **Peptide Weight**

This peptide has been supplied to you by gross weight. This means that, in addition to the peptide, part of the weight of the material you have received will be made up of residual water and counter ions. Since peptides are normally purified in aqueous acetonitrile buffers containing trifluoroacetic acid you may expect all acidic residues to be protonated and be neutral and the basic residues histidine, arginine and lysine to be protonated and positively charged with a trifluoroacetate counter ion. Peptides containing both acidic and basic residues may form internal salts rather than be present as salts of trifluoroacetic acid. Typically the residual water and counter ions account for around 20% of the weight you have been supplied.

The fraction of the gross weight that is peptide is usually known as the net peptide content. The net peptide content can be determined by elemental analysis or by UV absorption. If you need to know the net peptide content and do not wish to do the analysis yourself then you can request CRB to conduct the analysis for you.

Alternatively you can request this analysis when you place your order.

You can also, for an additional fee, request to receive a peptide based on a net weight of material (for example; if you request 10mg of net peptide and we determine the peptide content to be 80%, you will receive 12.5mg gross of peptide).

If the trifluoroacetate counter ion is undesirable, please request CRB to replace it with a different counter ion.

# **HPLC** Analysis

This peptide has been analysed by HPLC using a Vydac Peptide and Protein Technology reverse-phase column. The column has a particle size of  $5\mu$ , aiding resolution, and a 300Å pore size, particularly suited to the separation of molecules such as peptides. The wavelength used for the analysis, 230nm, is chosen because of the absorbance of the peptide amide bond in this region. Amino acids such as phenylalanine, tryptophan, tyrosine and histidine may also contribute to the absorbance at this wavelength.

## Maldi-TOF

The molecular weight of this peptide has been determined by Maldi-TOF mass spectrometry. Using positive ion ionisation the peptide is protonated and 'flies' along the path of the mass spectrometer as the molecular ion plus hydrogen, indicated as  $[M+H]^+$ . The machine used is of the reflector type that significantly increases sensitivity so that accuracies of greater than 50ppm are possible thus it is easily possible to distinguish between C-terminal carboxylic acids and carboxamides and between reduced and oxidised disulphides even at higher molecular weights. If your peptide has been despatched with a Maldi trace you may notice that the  $[M+H]^+$  peak is followed by a series of other peaks at approximately +1 units: these are the corresponding carbon-13 isotopomers of the peptide.

Occasionally, additional peaks in the spectrum corresponding to [M+23] and [M+39] may be present in addition to, or in place of, the  $[M+H]^+$  peak. These correspond to  $[M+Na]^+$  and  $[M+K]^+$  species formed during the sample preparation and ionisation processes.

## Solubility

Solubility of peptides can vary enormously. Therefore, it is essential that trial dissolution studies be carried out on a small sample of peptide to determine the optimum conditions before use. Use of incorrect dissolution conditions could result in complete loss of peptide as it may not be possible to recover undissolved peptide from the buffer salts. Many peptides will dissolve in water and this should be the first solvent to try.

As a general guide, basic peptides, those containing a preponderance of Lys and Arg residues, should dissolve in acidic buffers. Conversely, acidic peptides containing an excess of Glu and Asp residues should dissolve in alkaline buffers. Hydrophobic peptides may only dissolve in organic solvents such as DMSO. If the peptide is required to be used in PBS for testing, dissolution of the peptide first in a suitable buffer or solvent in which it is readily soluble before careful dilution with PBS may be necessary for achieving dissolution.