

3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

## **ProductInformation**

## **Ampholine**

Product Number A 5174 Storage Temperature 2-8 °C

Product Description
CAS Number: 37348-94-0

This product is a sterile filled, 40.0% (w/v) solution. It has been tested to be suitable for use as an isoelectric focusing reagent over the pH range 3.5 - 10.0. The recommend concentration for use is 2%.

Ampholytes are low molecular weight synthetic compounds which are used to generate a pH gradient under application of an electric field. At their respective isoelectric points (pl), the compounds have high buffering capacity. Often referred to as carrier ampholytes, these compounds are commonly used in a defined pH range to give selective resolution to mixtures of amphoteric materials such as proteins. Ampholytes may be used as free reagents or in isoelectric focusing (IEF) strips, whereby the carrier ampholytes are immobilized in a gel matrix.<sup>1</sup>

Application of voltage across the ampholyte pH gradient causes the amphotheric molecules of interest to migrate to the gradient positions where their net charges are zero. Materials with net positive charge move toward the cathode, and materials with net negative charge migrate toward the anode. The materials become less charged as they traverse the pH gradient until they reach their respective pl values. Substances which diffuse away from their pl immediately acquire charge and return to their isoelectric position. This focuses the materials at their pl values, separated with very small charge differences. The electric field strength determines the degree of resolution, and thus IEF is performed at high voltages (> 1,000 V). When the materials have reached their final isoelectric positions, the system has very little ionic movement, resulting in a low final current (< 1 mA).

Ampholine® IEF carrier ampholytes are mixtures of 600-700 different homologues within a defined isoelectric point range. Ampholine products are prepared by the reaction of aliphatic oligoamines with acrylic acids. Their molecular weights are generally <1 kDa. Because of their hydrophilic nature, they will not bind to proteins.

Several reports on the use of carrier ampholytes in protein electrophoresis have been published. 2,3,4,5

## **Precautions and Disclaimer**

For Laboratory Use Only. Not for drug, household or other uses.

## References

- 1. Righetti, P. G., et al., Isoelectric focusing in immobilized pH gradients. Methods Enzymol., **270**, 235-255 (1996).
- Gorg, A., et al., Recent developments in twodimensional gel electrophoresis with immobilized pH gradients: wide pH gradients up to pH 12, longer separation distances and simplified procedures. Electrophoresis, 20(4-5), 712-717 (1999).
- Gorg, A., et al., The current state of twodimensional electrophoresis with immobilized pH gradients. Electrophoresis, 21(6), 1037-1053 (2000).
- Zuo, X., and Speicher, D. W., Quantitative evaluation of protein recoveries in two-dimensional electrophoresis with immobilized pH gradients. Electrophoresis, 21(14), 3035-3047 (2000).
- 5. Chartogne, A., et al., Capillary electrophoretic separations of proteins using carrier ampholytes. J. Chromatogr. A, **959(1-2)**, 289-298 (2002).

Ampholine is a registered trademark of Amersham Biosciences.

GCY/RXR 6/03