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IEF Markers

Catalog Numbers **A2910**, **T1021**, **L5137**, **C3666**, **C6403**, **C6653**, **M9267**, **L1277**, **and T1146**Storage Temperature –20 °C

Product Description

The use of individual protein markers enables the investigator to select a marker mixture best suited for the pH range being studied. Thus, the needless waste of proteins that would stack on the anode or cathode is eliminated. In pl determinations, individual protein markers should be used in conjunction with at least two or three other markers as part of a standard curve.

Catalog Number	Protein	Approximate pl
A2910	Amyloglucosidase from <i>Aspergillus niger</i>	3.6
T1021	Trypsin Inhibitor from Glycine max (soybean)	4.6
L5137	β-Lactoglobulin A from bovine milk	5.1
C3666	Carbonic Anhydrase Isozyme II from bovine erythrocytes	5.4
C6403	Carbonic Anhydrase Isozyme II from bovine erythrocytes	5.9
C6653	Carbonic Anhydrase Isozyme I from human erythrocytes	6.6
M9267	Myoglobin from equine heart	Minor Band 6.8 Major Band 7.2
L1277	Lectin from <i>Lens</i> culinaris (lentil)	8.2, 8.6, 8.8
T1146	Trypsinogen from bovine pancreas	9.3

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

Store the lyophilized product at -20 °C with desiccant.

After reconstitution, the product is stable for 12 months at -20 °C.

Preparation Instructions

Catalog Numbers L1277 and C3666 contain 1 mg of protein and should each be dissolved in 0.25 ml of deionized water. The remaining markers contain 2 mg of protein and should be dissolved in 0.5 ml of deionized water. The reconstituted solution contains 4 mg/ml of protein and 200 mM glycine.

Solutions may be diluted for use as individual markers or combined for use as pl standards in the desired pH ranges. In either case, the recommended final protein concentration should be 0.4–0.6 mg/ml per marker except for Catalog Number L1277, which should have a final concentration of 0.8–2.0 mg/ml.

<u>Note</u>: Lentil Lectin (Catalog Number L1277) and β-Lactoglobulin A (Catalog Number L5137) may be hazy upon reconstitution. This in no way affects the electrofocusing, and results in an insignificant loss of protein.

Procedure

Application of solutions to the gel may be accomplished in several ways. Small paper applicators are available which will apply ~15 μ l. Protein samples, usually up to 10 μ l in volume, may also be applied directly to the gel as drops, streaks, or rectangles. Basins may be made in the gel into which the sample is introduced; however, the depth of the basin should be <30% of the gel thickness. Deeper troughs may skew the pH gradient. 1

One mm thick gels (5% acrylamide and 3% crosslinking agent) should be run at 1 W/cm gel length. If paper applicators are used, they should be removed 30 minutes into a run. One additional hour should be sufficient time to complete the electrofocusing. If a constant power supply is not available, run at 25 V/cm for 30 minutes and 50 V/cm for 5.5 hours.

References

 Righetti, P.G., and Drysdale, J.W., Isoelectric Focusing Laboratory Techniques in Biochemistry and Molecular Biology (Work, T.S. and Work, E., gen. eds.) North Holland Publishing Co., Amsterdam, New York, Oxford (1976).

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