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#21210 LX-112 KIT
Instructions for Use

For accuracy of measurement and so that the given W.P.E. of the embedding resin can be used to advantage, we recommend the gravimetric rather than the volumetric measurement for the preparation of embedding media.

The weight per epoxide equivalent (W.P.E.) Of epoxy resins can vary from batch to batch. Manufacturing specifications for LX-112 list a wide range of W.P.E. values. This variation is a possible cause of poor sectioning characteristics of a block and for irreproducible embedments. To obtain reproducible blocks for sectioning, we recommend that formulations be made on a weight basis using epoxy resins analyzed for W.P.E. and anhydrides analyzed for purity. When an anhydride contains as little as 5% free acid and/or water, resulting embedding blocks may be soft, have holes, and exhibit generally poor cutting characteristics.

RESIN FORMULATIONS

To readily use the exact W.P.E. value of the epoxy resins required for embedding on a weight basis use the following simple general equation:

$$\text{Weight of Anhydride Required} = E/WPE \times \text{Anhydride Molecular Weight} \times \text{Ratio of Anhydride to Epoxy Equivalents}$$

where E is grams of epoxy resin, WPE is the weight of epoxy resin containing one equivalent weight of epoxide and the *ratio of anhydride to epoxy equivalent* is equal to 0.7 based on Dr. John Luft's published results. (Luft, John H., 1961, "Improvements in Epoxy Resin Embedding Methods", *J. Biophys and Biochem Cytol*, **9**:409.). The molecular weights of NMA and DDSA are 178 and 266 respectively.

The first step in making a complete LX-112 embedding solution is to thoroughly mix the two resin solutions; one containing DDSA (Mixture A) and the other containing NMA (Mixture B). These solutions should be mixed in a chemical fume hood.

To make Mixture A: Assuming a WPE of 144 for the LX-112 and using 100 g of LX-112.

$$\text{Weight of DDSA} = 100/144 \times 266 \times 0.7 = 129 \text{ g DDSA}$$

- 1) Put 129 g DDSA into an 8 oz. bottle
- 2) Add 100 g LX-112
- 3) Cap and shake vigorously until thoroughly mixed

To make Mixture B: Assuming a WPE of 144 for the LX-112 and using 100 g of LX-112.

Weight of NMA = $100/144 \times 178 \times 0.7 = 87$ g NMA

- 1) Put 87 grams NMA into an 8 oz. bottle
- 2) Add 100 grams LX-112
- 3) Cap and shake vigorously until thoroughly mixed.

For your convenience Table I includes anhydride weights for various LX-112 WPE and starting with 100 g LX-112.

TABLE I

LX-112 WPE	Weight of DDSA (in g)	Weight of NMA (in g)
	(Mixture A)	(Mixture B)
140	133	89
141	132	88
142	131	88
143	130	87
144	130	87
145	128	86
146	128	85
147	127	85
148	126	84
149	125	84
150	124	83

RESIN BLOCK HARDNESS

Hardness of resin blocks for sectioning depends upon the ratio of mixtures A and B in the complete resin mix. Mixture A alone gives a soft block, whereas Mixture B alone gives a very hard block. The ratios of Mixture A to Mixture B and their hardness values were assessed by Dr. Luft in the above mentioned paper. These ratios have been converted to a weight basis which very closely approximates the volume basis, as shown in Table II.

TABLE II

Mixture A Weight in g	Mixture B Weight in g	Total Weight in g	Accelerator DMP-30	Relative Hardness
10	0	10	0.14	Softest
7	3	10	0.14	Soft
5	5	10	0.14	Medium
3	7	10	0.14	Hard
0	10	10	0.14	Hardest

For new workers in this field we recommend as a starting point mixing the two resin solutions in the proportion of 6 of A to 4 of B. This ration would produce a block of medium hardness.

COMPLETE RESIN MIX

Add 0.14 g (0.14 ml can be satisfactorily substituted) of DMP-30 to a solution of Mixture A and Mixture B as detailed in Table II. Stir or shake entire solution for approximately 5 minutes until thoroughly mixed. Incomplete mixing may produce blocks with poor sectioning characteristics.

POLYMERIZATION

The complete resin can be cured overnight at 60°C or by the following steps to enhance infiltration of the specimen.

1. Heat overnight at 45°C (18-20 hours)
2. Increase heat to 60°C for an additional 24 hours
3. Remove blocks from heat to cool
4. Upon reaching room temperature, blocks are ready for sectioning

STORAGE OF MATERIALS

Mixtures A or B six months old, or more, may be used successfully if the mixtures are kept under inert gas and continuously refrigerated. These refrigerated mixtures, however, should be warmed to room temperature before opening. This will prevent water condensation in the embedding media, which is one cause of holes in blocks and soft blocks. Furthermore, the presence of water may produce slow hydrolysis of the anhydrides to free acids which is another cause of soft, defective blocks. For added protection, bottles should be stored with a "head" of inert gas, e.g. Argon or Nitrogen, to prevent contamination by entrapped humid air.

CLEAN UP

Plastisolve (Catalog Number 13050) is an effective solvent for polymerized epoxies. Met-a-terge (Catalog Number 13045) and acetone (Catalog Number 20415) are effective solvents for epoxies that are not polymerized. All are useful in keeping working areas clean.