

TECHNICAL NOTES

LOW VISCOSITY/SPURR MINI KIT

Formulated according to Spurr Prod. No. 18108

Kit contains the following chemicals:

NSA (Noneyl Succinic Anhydride) 55ml (in 120ml jar)

ERL 4221 20ml

DER 736 (Dow Epoxy Resin 736) 12ml

DMAE (Dimethylaminoethanol) 5ml

Additional Supplies included in this kit:

- 3 x 10ml syringes
 - 1 x 5ml syringe
- 2 wooden mixers

How to prepare the embedding media:

- Open the large 120ml jar that contains the NSA and add entire contents of ERL (20ml) and DER 736 (12ml)
- to it. Drain them completely.
- Using the 5ml syringe, withdraw 1ml of DMAE and add to the mixture, pushing the plunger rapidly to insure that all the contents are released into the mix.
- Mix thoroughly, minimizing exposure to atmospheric moisture.
- Draw the mixture into the 10ml syringes to dispense the mixture for infiltration and embedding in molds or capsules.

Infiltration:

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- Tissue is fixed as usual and dehydrated through absolute alcohol and brought through two changes of propylene oxide. Tissue is infiltrated in three steps of 30 minutes each.
 - 1. Equal volumes of propylene oxide and embedding media: 1:1.
 - 2. One part propylene oxide to two parts embedding media: 1:2.
- 3. Embedding media, only.
- Transfer the tissue to embedding capsules and fill with fresh embedding media. Polymerization is at 70°C for 24 hours.
- NOTE: Remaining media can be stored in syringes at 4°C for 2 days. This mixture is not miscible with ethanol.

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Microwave-assisted Resin Polymerization. Resin polymerization is best done under water in the microwave. This method insures reproducibility, good cutting properties and beam stability. (Giberson et al., J. Vet. Diag. Invest. 9:61-67)

Figure 3A. Capsule Preparation for Resin Polymerization

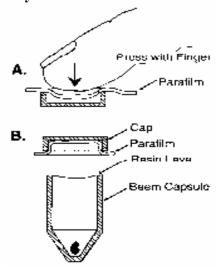
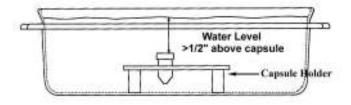


Figure 3B. Resin Polymerization Underwater



Polyethylene embedding capsules are used for microwave-assisted polymerization. Figure 3A describes the best method devised to insure that the capsule will not leak when under water. The cap is removed from the capsule and Parafilm placed in the lid as shown in Step 1. After the sample is in the capsule, fill to the top as shown in Step 2 and then place the cap with Parafilm liner on the capsule. In Figure 3B the Product No. 36131 Capsule Holder or 36131-2 Capsule Holder with Lid is used to keep the embedding capsules oriented underwater during polymerization. Place the plastic dish, (4-cup) with holder and capsules, on top of the PELCO ColdSpot®, Product No. 36116-10 or 36116-20. Fill with water as shown in Fig. 3B. The wattage should be set to approximately 300W for 30 min. and then change the wattage to approximately 750W for the remaining 45 min. The new BioWave Pro has its own programmed polymerization cycle and the above is not needed. The polymerization requires 1 hour and 15 minutes. Cover but do not seal the dish to avoid loss of water volume due to evaporation and to allow the water temperature to reach 100°C. Covering the water with a closed cell foam or a Styrofoam sheet works well. If the water boils too vigorously reduce the high wattage down to below 500W.

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