We appreciate the response to this publication feature and welcome all contributions. Contributions may be sent to José A. Mascorro, our Technical Editor, at his e-mail address: jmascor@tulane.edu. José may also be reached at the Department of Structural and Cellular Biology, Tulane University Health Sciences Center, 1430 Tulane Ave., New Orleans, LA 70112 and Ph: (504) 584-2747 Fax:

Making Your own Molds for the EM lab

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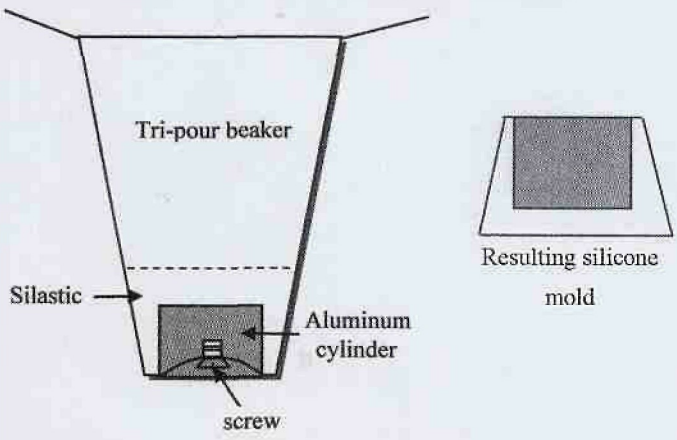
Silicone rubber can be cast into many useful shapes quite easily in the laboratory. I make my own metallurgical mounting molds for mounting samples in epoxy resin. I have patterns for making molds which are one inch in diameter by 5/8 inch high and others for making 1.25 inch and 0.75 inch diameter molds. They are all based on the disposable Tri-pour plastic beakers. You can use dixie cups or other disposable containers and you can make almost any size and shape molds that you can design a suitable reverse mold for.

For the one inch molds, I use a 50 ml. beaker which is about 1.5 inches in diameter at the bottom. There is a hole in the center of the bottom of the beaker to admit a small screw that is about 0.25 inches long. For the hollow in the mold, I have an aluminum cylinder, one inch in diameter and 5/8 inch tall. The bottom of the cylinder is machined a little concave to match the shape of the bottom of the



beaker, and tapped with a hole to receive the small screw. This allows you to screw the aluminum cylinder tightly down to the bottom of the beaker to prevent the silicone rubber from running under it.

The Silastic E RTV Silicone rubber kit (Dow Corning Corp.) is mixed according to directions. I use a 110 gram mixture to make five molds at once. The rubber base is weighed, then the catalyst is weighed, added and mixed thoroughly in a disposable 200 ml cup. The mixture is then out-gassed in a vacuum desiccator to remove as much of the air trapped in the Silastic as possible. The mixture is dispensed into the Tri-pour beaker reverse mold, until there is at least 3/8 inch of Silastic over the aluminum cylinder. Dispense slowly so that the Silastic has time to run down into the corners and minimum air is trapped. The individual beakers and the original Silastic mix are both out-gassed in a vacuum dessicator.

The Silastic will cure at room temperature over 24 hours. The screw is removed and the aluminum cylinder pushed up to remove the rubber mold from the beaker. It may help to run a thin spatula around the inside of the beaker to release the sticky rubber. Any flashing or rubber that has run under the cylinder can be trimmed with a sharp blade. I find it helps to spray the rubber molds lightly with a silicone mold release spray each time before pouring in the epoxy resin, when using the silicon rubber molds to mount samples. The molds will dry out and crack in use, but I use mine for six months to a year before replacing them.

The chemicals in the Silastic E RTV Silicone Rubber Kit can be irritant and toxic. Read all the MSDS and warnings and wear proper protective gear when mixing and dispensing the silicone rubber.

Vascular Corrosion Casting

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With the development of low viscosity resins, casts of the fine capillary beds of tissues and organs were attainable, and these could be viewed and studied in detail in the scanning electron microscope [1, 2]. The advantage of such casts is that they permitted, for the first time, viewing of the intact, three-dimensional complex vasculature of tissues and organs. It follows that once vascular casts are obtained, quantitative measurements of various types could also be made from the casts if care was exercised. Of course, in theory, it should be possible to prepare casts of the three-dimensional lumenal space of any structure, biological or other [3]. This report will concentrate only on applications of corrosion casting to the vasculature of animal species.

Several modifications of the general method of preparing vascular corrosion casts are currently used in different laboratories, as well as several types of resins [4]. Outlined here is one method. After appropriate anesthetic and anticoagulant are administered (for rat: ip injections of nembutal, 60mg/kg; heparin, 700U/kg), an artery leading to the organ of interest is located and cannulated. The cannula is attached to one port on a three-way valve [5]. Another port on the valve is attached to a saline or Ringer=s reservoir (warmed to 37° C), and the third port on the valve is attached to a syringe barrel. The entire system is joined to a manometer and pressure bulb for controlling perfusion pressure. A vein draining the organ of interest is opened as an outlet, and blood is flushed from the organ with the warm saline or Ringers=s solution at a pressure appropriate for the animal (80-100mm Hg for rats). Resin (8 ml Mercox, 2 ml methylmethacrylate monomer, and 0.3 ml catalyst; prepared just before use) is placed in the syringe barrel and infused through the same cannula until the onset of polymerization (about 8-10 min). The

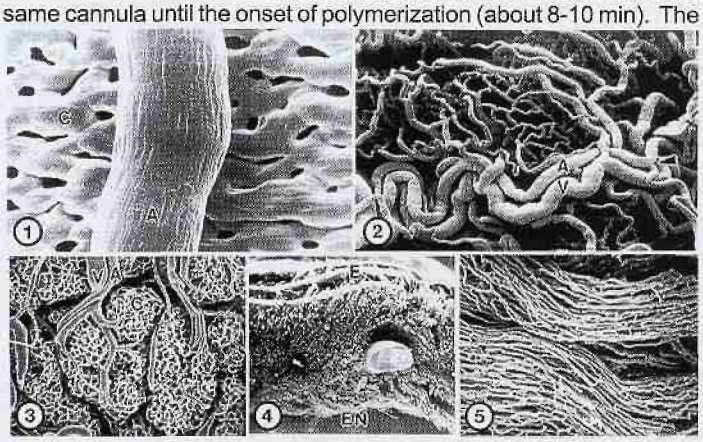

Figure 1. Vascular corrosion cast of frog retina. A, artery; C, capillary. x320
Figure 2. Vascular corrosion cast of rabbit bladder. A, artery; V, vein; arrowhead, sphincter. x132
Figure 3. Vascular cast of avian salt gland. A, artery; V, vein; C, capillary bed. x106
Figure 4. Vascular cast of right ventricular free wall of rat heart. E, epicardium; EN, endocardium. x34
Figure 5. Vascular cast of capillary beds of trabecular muscle of rat heart. x121