

Modified Vascular Injection Procedure

Used in the Department of Veterinary Anatomy
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Ever since Wm. Harvey made the observation that blood circulated in vessels there has been a continuing quest for a detailed blueprint of the systems of vessels in the various animals and man. The course of the larger vessels was readily worked out in fresh specimens. As man's knowledge of the various phases of medicine increased it became evident that there was much to be learned about the anatomy of the circulatory system. Much thought and experimentation was given to devising methods by which knowledge of this system could be expanded. A colored substance was sought which, when injected into the vessels, would solidify in a short time. Time proved this to be the approach of choice. The search revolved around finding the ideal injection medium.

Latex emulsion proved to be one of the most satisfactory media as evidenced by its widespread use. The most serious limitation was the inability to inject it into vessels of capillary size in the intact animal. This was accomplished in extirpated organs and amputated appendages. The disadvantage of the latter method was the difficulty in cannulating, perfusing

and injecting every artery when several supplied the same part. The ideal medium would be one which could be injected through the capillary bed through one cannula in the intact animal. A study was undertaken in an attempt to determine the reason or reasons for the limitations of latex emulsion injection medium.

The first consideration was to remove all of the blood possible and to change that which remained in such a manner that it would not clot when coming into contact with an acid. This state was accomplished in several steps. First, the animals were put under surgical anesthesia with pentobarbital sodium via the cephalic vein, to effect. Second, †(chorozol Fast Pink B), 2 gms./gal. of normal saline, was given by gravity at the rate of 200 cc/40–60 lbs. of body weight. Third, the common carotid artery and femoral vein were cannulated. Infusion of warm saline solution via the femoral vein was begun simultaneously with exsanguination through the cannula in the carotid artery. When the heart beat began to weaken, exsanguination was stopped until the blood volume was restored by the normal saline to a point where the heart activity improved. Fourth, when the heart failed to respond to the latter procedure digitalis was given via the cephalic vein at the rate of 1/20 mg. per dose and repeated when the heart appeared to be losing the effect of the previous injection.

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† Salantine Pink 4B6, Allied Chemical and Dye Corp., 40 Rector St., New York 6, New York

A maximum of .4 mg. ***Purodigin* per 60 pounds of body weight was given. Fifth, the saline infusion was stopped when the response of the heart to digitalis began to diminish. When the heart failed to pump any fluid through the cannula, it was pumped by hand by alternate compression and decompression of the thorax. The fluid that was left in the vessels was of a low viscosity and could be moved freely in the vessels 12 hours postmortem. There was no evidence of clotting when the animals were perfused with a weak acid.

The true diameter of the lumen of the vessels was not considered of prime importance in this study. If the relative size of the vessels remains constant, there would be an advantage in a substance that would destroy the elasticity of the vessels, thereby, increasing the size of the lumen. Acetic acid (5% solution), reported at various times to be effective, was used at the rate of 1000 cc./40-60 pound dog at 120 mm. Hg.

The use of 5% acetic acid in conjunction with latex injection posed several serious problems when working with the intact animal. The problems revolved around the fact that latex, chemically basic, changed from a liquid to a solid instantly when it came into contact with an acid. The tissue breaks down forming a perivascular hydrophilic radical. The circulatory system could not be irrigated following the use of the acid solution in the intact animal. The reason for this was that when the elasticity of the vessels was destroyed, they became very porous. The porous vessel walls together with the perivascular hydrophilic radical resulted in an edema that doomed to failure subsequent attempts to inject latex.

The perivascular hydrophilic action of the acid radical added to the difficulty in another way. It drew the H₂O from the latex emulsion increasing its viscosity thus making it more difficult to get into the smaller vessels. Another serious problem was the reaction of the latex emulsion to an acid. When one considered the fact that the slight acid reaction of the tissue

will gradually increase the viscosity and eventually turn the latex emulsion to a solid, one could readily imagine the speed with which this reaction was accelerated when 1000 cc. of a 5% acetic acid solution, very strong in comparison, was infused into the circulating system.

These problems were solved in three ways. First, ammonium hydroxide was added to the latex emulsion (6 ounces of 12% solution to one quart of latex emulsion). This reduced the original viscosity of the latex emulsion and changed it more toward the basic side thereby requiring more time for it to become acid in the circulatory system. Second, the 5% acetic acid injection was followed immediately by an injection of normal saline solution (750 cc./40-60 pounds of body weight). This acted as a buffer between the acid and the latex. Third, an incision was made in the fourth intercostal space on the left side. The pericardial sac was invaded and the aorta was clamped between the heart and the attachment of the pericardium to the aorta. The pressure on the latex emulsion was raised to 120 mm. Hg. before being released into the circulatory system. This was done to obtain the greatest possible speed of injection. Speed of injection was very important because the speed of the chemical reaction and the action of the hydrophilic radical were increased as the lumen size of the vessels decreased. The reason for this was, as the lumen size decreased, there was a proportionate increase in the amount of medium in contact with the vessel walls.

This method made it possible to inject all the minute capillary beds of the animal body including the capillaries of the retina of the dog and the goat by way of the common carotid artery.

Volume Correction

The volume number appearing on the cover of issue number one, 1961-62 was in error. XXIX (29) should read Vol. XXIV (24). Persons in possession of this issue should make the necessary correction on the cover page.

**Wyeth Laboratories crystalline digitoxin