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An Educational Method to Bridge a Gap Between Histology and Gross Anatomy in Learning Microvascular Structure

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The combined method injecting silicon rubber into vessels and magnifying them on a TV monitor with a video macroscope can demonstrate three-dimensional vascular structure of organs. This study compared it with a corrosion casting method observing renal glomeruli, and implied that as it is easy to prepare samples, magnifying 200 times on the TV monitor and operating the machine, students can treat it in the dissection course and in a limited schedule. It leads students to be interested in histology as well as gross anatomy, and is helpful for them to perceive human structures. © 1994 Wiley-Liss, Inc.

Key words: anatomical education, vascular structure, macroscope

INTRODUCTION

Classical studies of anatomy divide the subject into gross anatomy and histology. The former is defined by the naked eye and the latter by microscopic study of sectioned tissues. While gross anatomy displays depth and relationships, it is limited by the resolution of the eye. Conversely, microscopy can resolve submicron detail but it is limited essentially to a two dimensional display. Much of anatomical structure lies at the borderline between these disciplines. Although many examples can be given, this paper will focus on the microvascular circulation. The knowledge of arterioles, capillaries, and venules of organs is indispensable to understanding histologic function and pathogenesis. Understanding the spatial intricacies of the renal glomerulus, hepatic sinusoid, or villous capillary network of intestinal mucosa is difficult when their structures are examined in thin section. If students have a means of visualizing anatomy in this transitional borderline they will have a better understanding of the microvasculature of each organ during dissection, as well as being led through a logical progression from gross to cellular anatomy. It also provides an interesting theme for instructors in linking these classical subdisciplines of anatomy.

Corrosion casting observed by a scanning electron microscope (SEM) is well known as a method to demonstrate microvasculature (Murakami, T. 1971). However, it is not a technique readily mastered by the novice student, and has various disadvantages in a practical course: complicated preparation for beginners, loss of the tissue except that protected by the case (even if the microvasculature is well demonstrated, the surrounding tissues are lost, so that the student has an incomplete view), necessity for expensive instruments and their maintenance, and loss of time due to operating the instruments and limitation of the number of students observing the samples. Therefore, an ideal macroscopic method must fulfill the following conditions: 1) simple enough to practice in the dissection course, 2) no maceration of tissues, 3) a technique to differentiate clearly between tissues and vessels, 4) high magnification (at least 200 times), and 5) an ability to thin section the gross specimen after macro-

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scopic preparation. These conditions are satisfied with the combination of a silicon rubber compound (Microfil, Cantor Bio-Medical Products, U.S.A.) injection method and a video macroscope (Nihon Koden, Japan).

MATERIALS AND METHODS

Silicon rubber compound (Microfil MV-112 white, yellow, red), diluted with MV-diluent equal to the compound and mixed with a stiffener (MV curing agent, 5% volume of diluted compound), hardens about 5 minutes after injection into the vessels of the animal (the rat used here). It is far softer than corrosion cast specimens and is sectioned easily by microtomy. Injected tissues are fixed in 10% formalin for 2 or 3 days, dehydrated in ethanol, and, lastly, immersed in methyl salicylate to render the specimen translucent. The vessels filled with colored silicon rubber compounds (yellow, red, white) stand out in the translucent tissue. The usual optical stereoscope does not provide a quality image because of its low magnification (<70 times) and requires that each student have access to a microscope. A video macroscope is not limited by these disadvantages (Fig. 1a). Consisting of a probe and charge coupling device (CCD) camera, light fiber, and TV monitor (Fig. 1b), the video macroscope was developed for studying the living microvascular circulation

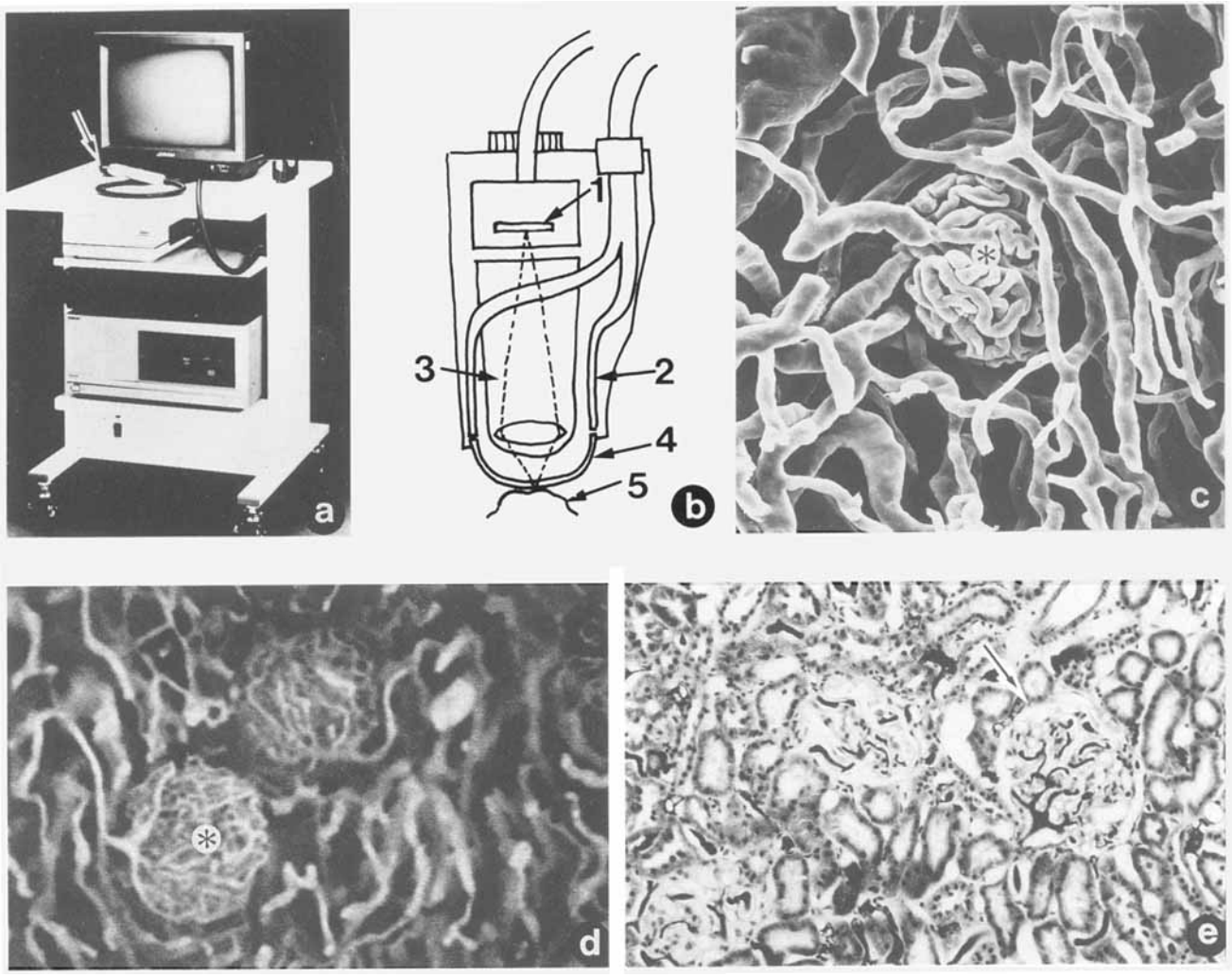

Fig. 1. **a**:A video macroscope: an arrow shows a probe. **b**: The schema of the probe. 1, CCD; 2, light guide (optic fiber); 3, lens; 4, light condensing cap; 5, object. The object lighted by the optic fiber is magnified by the objective lens and imaged on the CCD, where the images are changed into the electrical signals and sent to the monitor. After being changed into the TV signals once again there, the real time images are displayed in the TV monitor. **c**:Corrosion casts of a glomerulus (asterisk) observed by SEM. ×440. **d**: Silicon rubber injected glomeruli (asterisk) observed by the video macroscope. ×490. **e**: A thin section of silicon injected glomruli (arrow). ×320.

(Sasaki et al., 1991). However, it has other wide applications. Observing the injected and cleared specimens, manipulating the specimen gently with the probe, a student or an entire class can readily observe the colored microvasculature on the TV monitor at magnifications of 50 to 400 times. Moving and still images can be stored on videotape or on video disks for future study. After macroscopic study, the samples are embedded in a paraffin, sectioned, and stained with haematoxylin and eosin. The sections can be studied by light microscopy and compared to the video images when they are retrieved from the tapes or disks.

The results of silicon-injection into the rat kidney are here compared with the corrosion casting method, which is explained in short. Forty milliliters of resin mixture (Mercox, Oken Japan) are introduced into the abdominal aorta. The hardened samples are immersed in 18% NaOH and then rinsed in hot water for 12 h. The maceration procedure is repeated three times. After being dried in air and coated with Pt-Au at a thickness of about 40 nm with a sputter coater, the samples are observed by SEM (Hitachi S-800).

RESULTS AND DISCUSSION

The detail of glomerular struture obtained by SEM (Fig. 1c) is similar to video macroscopy (Fig. 1d) when viewed at the same magnification. The glomerular vessels appear in the dark field. Fan-like capillaries surround the glomerulus (Fig. 1d). Furthermore, when the samples are sectioned, stained, and viewed by light microscopy, the student can compare the histological image (Fig. 1e) with the three-dimensional intact specimen (Fig. 1d). In fact, students seemed to be impressed by finding biological secrets in the organ during the course. As von Lüdinghausen (1992) points out, in regard to dissection, "Especially in the anatomy laboratory, students are able to study the tissues by manipulation,..."; this is an opportunity to manipulate tissues at the macroscopic level.

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