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Murakami T. Application of the scanning electron microscope to the study of the fine distribution of the blood vessels. *Arch. Histol. Jpn.* 32:445-54, 1971.
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This paper describes some advantages of vascular casting and scanning electron microscopy in the study of microcirculatory systems and introduces a methacrylate method that allows a thorough reproduction of the whole blood vascular beds (arteries, capillaries, and veins). [The *SCI*® indicates that this paper has been cited in over 210 publications, making it the most-cited paper from this journal.]

Vascular Casting and Scanning Electron Microscopy

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When I was a postgraduate student (1965-1969) of our department, Professor Hiromu Outi advised me to study the microvascular systems under the guidance of Professor Tsuneo Fujita, who introduced in 1965 a scanning electron microscope (SEM) in our medical school. I began in 1967 to study these systems and developed in 1968 a methyl methacrylate method useful for thorough casting of the whole blood vascular beds (arteries, capillaries, and veins) and also useful for detailed analysis of these beds with the SEM. Because of my poor English, it took about two years to publish this work in my favorite journal, *Archivum Histologicum Japonicum* (Archives of Histology and Cytology), which was founded in 1950 by Professor Masaji Seki in our department.

I had to use a large amount of methacrylate (40 liters or more) and also had to sacrifice a large number of animals, especially rats (100 or more), to prepare the satisfactory casting media. During the preparation, I accidentally caused a fire. Professor Outi kept this fire secret and allowed me to continue my work. Furthermore, I accidentally set a monkey free and had to ask the police to catch this monkey. Although this accident was written up in newspapers and regarded as a criminal act, Professor Fujita went to the police and saved me from punishment.

Our methacrylate casting media, as well as diluted and refined media,[1-6] are widely available for replication of the blood vascular beds, lymphatic vessels, lung airways, biliary tracts, and other tubular systems of various animals, including human autopsy samples. The use of such media permit fine SEM studies of basic architecture, developing patterns, age-related vicissitudes, species variations, anomalies, physiological changes, and pathological changes.[2-6]

The most valuable and reliable findings obtained by vascular casting and SEM are the details of vascular connections, arrangements, and distributions.[2-6] Computer-aided scanning microscopy of the casts allows measurement of vascular densities. Nuclear protrusions, cell boundaries, cushions, valves, sphincters, and other mural and endothelial elements are visible on the casts.[3] Vascular contractions caused by resin injection are also replicated.[3] Estimations of specific gravity and weight allow calculation of the volume of the casts or injected vascular beds.[3] Measurements allow calculation of length and diameter of vessels of interest. Incomplete injections through the arteries of casting media as viscous as blood are useful to study blood flow.

Positional relations of each vessel to the tissue elements cannot be examined in the cast samples. Despite this problem, our technique of displaying microvascular patterns as independent structures has been valuable in biomedical research.[1-6] To my knowledge, 1,000 or more biomedical papers have used the technique since 1971.

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2. **Murakami T.** Methyl methacrylate injection replica method. (Hayat H A, ed.) *Principles and techniques of scanning electron microscopy*. New York: Van Nostrand Reinhold, 1977. Vol. 6. p. 159-69.
3. **Murakami T, Ohtani O, Ohtsuka A & Kikuta A.** Injection replication and scanning electron microscopy of blood vessels. (Hodges G M & Carr K E, eds.) *Biomedical research applications of scanning electron microscopy*. London: Academic Press, 1983. Vol. 3. p. 1-30.
4. **Murakami T, Kikuta A, Kaneshige T & Naito I.** Minute structure of human kidney glomerulus, its embryonic development and age-related changes: a scanning electron microscope study of vascular casts. (Seno S, Copley A L, Venkatachalam M A, Hamashima Y & Tsujii T, eds.) *Glomerular dysfunction and biopathology of vascular walls*. Tokyo, Japan: Academic Press, 1985. p. 103-17.
5. **Murakami T, Ikebuchi Y, Ohtsuka A, Kikuta A, Taguchi T & Ohtani O.** The blood vascular wreath of rat ovarian follicle, with special reference to its changes in ovulation and luteinization: a scanning electron microscopic study of corrosion casts. *Arch. Histol. Cytol.* 51:299-313, 1988.
6. **Yamamoto K, Itoshima T, Tsuji T & Murakami T.** Three-dimensional fine structure of the biliary tract: scanning electron microscopy of biliary casts. *J. Electron Microsc. Tech.* 14:208-17, 1990.

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