ROLE OF THE INTERNAL ELASTIC MEMBRANE IN THE FORMATION OF THE SURFACE MICRORELIEF OF BLOOD VESSELS

L. D. Krymskii, G. B. Nestaiko, and A. B. Shekhter

UDC 611.132-018-086.3

A scanning electron-microscopic study of areas of the aorta with intact endothelium and after removal of the surface layer of the intima showed that the large folds of the microrelief of the blood vessels are formed by the internal elastic membrane.

KEY WORDS: microrelief of inner surface of blood vessels; internal elastic membrane; endothelial cells.

Studies with the scanning electron microscope have shown that the inner surface of blood vessels is not a smooth-walled cylinder but it has a complex relief. In particular, the microrelief of arteries of elastic type has been shown to consist of large folds (of the first order), arranged parallel to one another [2, 3, 5, 6]. The structures responsible for the formation of these folds have not yet been identified. It has been suggested that they are formed by endothelial cells arranged in a row, one after the other [6, 8]. According to other workers, the first order folds are formed by the internal elastic membrane [2-5].

The object of this investigation was to continue the study of the role of various components of the intima in the formation of the microrelief of the inner surface of blood vessels.

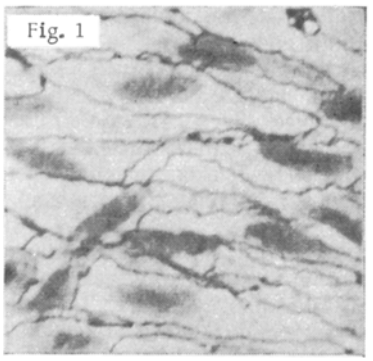
EXPERIMENTAL METHOD

The abdominal aorta of five mongrel dogs and five chinchilla rabbits was studied. Immediately after autopsy pieces of the blood vessels measuring from 1 to 1.5 cm^2 in area were excised, washed with physiological saline for 5-10 min, and impregnated with silver nitrate by Sinapius' method [7] to reveal the boundaries of the endothelial cells (the "cement lines"). In some preparations the endothelial layer was removed mechanically. All preparations were fixed in 1.5% glutaraldehyde solution in phosphate buffer, pH 7.4, and dehydrated in alcohols of increasing concentration. The endothelial layer or the whole of the intima down to the internal elastic membrane was removed from some of the dehydrated preparations with collodion film by Kochetov's method [1]. The integrity of the endothelial layer was judged from observations on the vessel wall with the optical microscope in reflected light and in film preparations in transmitted light. All the preparations were sprayed with a thin layer of carbon and silver and examined in the SSM-S1 and Stereoscan scanning electron microscopes with an accelerating voltage of 10 and 20 kV and magnification of between 100 and 10,000 × .

EXPERIMENTAL RESULTS

Examination of the inner surface of the aorta in reflected light and in film preparations of the detached endothelial layer clearly revealed the boundaries of the endothelial cells (Fig. 1). The "cement lines" were absent in de-endothelized areas.

A. N. Bakulev Institute of Cardiovascular Surgery, Academy of Medical Sciences of the USSR. Central Research Laboratory, I. M. Sechenov First Moscow Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR Yu. M. Lopukhin.)
Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 80, No. 12, pp. 94-96, December, 1975. Original article submitted April 28, 1975.



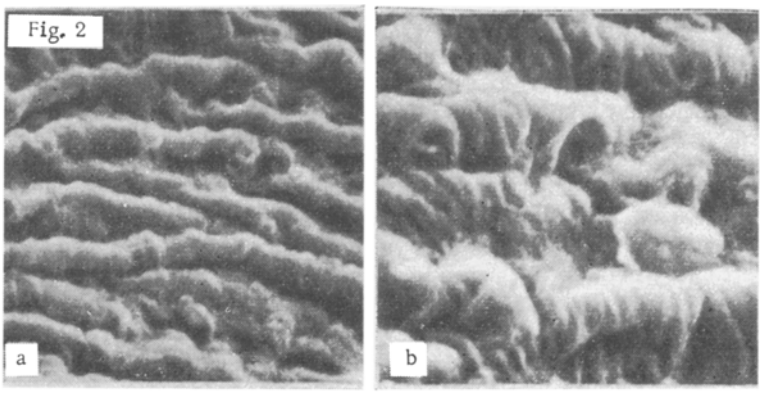


Fig. 1 Fig. 2

Fig. 1. Film preparation of detached endothelium. Boundaries between endothelial cells are clearly visible (2000×).

Fig. 3. Inner surface of aorta with intact endothelium (scanning electron microscope): a) first-order folds (1000 ×); b) endothelial cells whose central part projects into lumen of vessel (5000 ×).

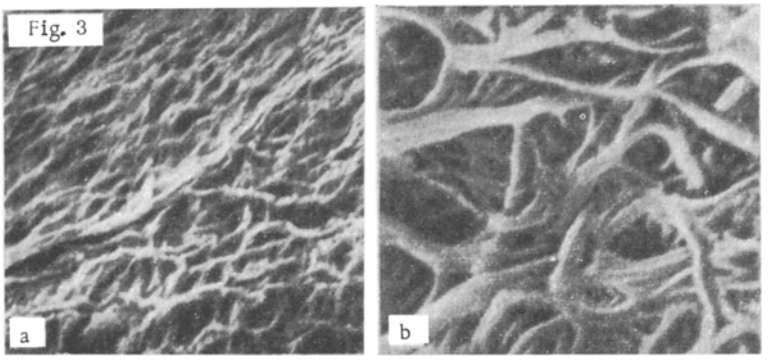


Fig. 3. De-endothelized area of inner surface of aorta (scanning electron microscope): a) first-order folds intact (500×); b) reticular, fenestrated structure formed by fibers of internal elastic membrane (5000×).

In the scanning electron microscope the inner surface of the aorta with its endothelium intact consists of large folds arranged along the long axis of the vessel, slightly skew, and with a helical twist (Fig. 2a). The distribution of the folds on the inner surface of the vessel is quite uniform, and together with their parallel arrangement this explains the regular microrelief. At the apex of the folds and in the gaps between them there are projections of different shapes measuring 5-10 μ, separated from each other by small hollows (Fig. 2b). The distances between the central parts of these projections are approximately equal to the distances between the nuclei of neighboring endothelial cells in film preparations.

In the de-endothelized areas of the surface of the aorta no projections measuring 5-10 μ are present. Meanwhile the first-order folds are intact (Fig. 3a). Under low power they are hardly distinguishable from the corresponding folds on the inner surface of the aorta with the endothelial cells intact. However, under magnifications of 1,000-3000 × it can be seen that the first-order folds consist of fibrous structures, arranged in bundles but joined together by fibers of smaller diameter (Fig. 3b). The connective-tissue fibers as a whole create a reticular, fenestrated structure characteristic of the internal elastic membrane.

The structure of the inner surface of the aorta with the endothelial layer intact as described above is identical with the observations made by the present writers and other

workers previously [2-6, 8]. There is some evidence that the projections into the lumen of the vessel, measuring 5-10 μ, are formed by the nuclei of endothelial cells covered by a thin layer of cytoplasm [3, 5, 6]. The absence of such projections found in the present investigation in the de-endothelized areas of the aorta confirms this hypothesis. Further support for the identity of the projections observed in the scanning electron microscope with the endothelial cells discovered in film preparations is given by their corresponding size.

First-order folds in areas of the aorta from which the endothelium is removed mechanically still persist. This indicates that the large folds are formed, not by endothelial cells, as some workers [6, 8] consider, but by the subendothelial connective-tissue layer of the intima and, in particular, by the internal elastic membrane. A similar view has been expressed in the past [2, 4, 5], but now for the first time, as a result of the use of special techniques (removal of the endothelial layer and intima to different depths under control of the optical microscope, followed by the study of these preparations in the scanning electron microscope), convincing proof in support of this hypothesis has been obtained. By means of these techniques, the three-dimensional structure of the internal elastic membrane could also be studied. It was shown to consist of separate fibers, interweaving in longitudinal and transverse directions, which together form a reticular, fenestrated structure.

LITERATURE CITED

1. N. A. Kochetov, Trudy S-Pb. Obshch. Estestvoisp., 39, No. 1, 134 (1908).
2. L. D. Krymskii and M. B. Il'ina, Arkh. Pat., No. 12, 45 (1973).
3. L. D. Krymskii, G. B. Nestaiko, and Yu. A. Stanishevskii, Éksp. Khir., No. 3, 84 (1974).
4. G. M. Chisolm, J. L. Gainer, and G. E. Stoner, Angiologica, 10, 10 (1973).
5. B. Christensen and C. Garbarsch, Angiologica, 9, 15 (1972).
6. T. Shimamoto, T. Yamashita, and T. Sunaga, Proc. Jap. Acad., 45, 507 507 (1969).
7. D. Sinapius, Z. Zellforsch., 44, 27 (1956).
8. G. Weber, P. Tosi, and C. Callesi, Arch. De Vecchi Anat. Pat., 44, 27 (1970).