I. Experimental problems

SUBMICROSCOPICAL DATA ABOUT THE DIFFERENTIATION OF ELASTIC FIBRES IN THE AORTIC WALL

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Key-words: aorta — elastic fibres — elastic lamellae — differentiation —electron microscopy

Aortic wall elasticity is determined by a fibro-elastic skeleton of specific structure. It undergoes age-dependent and pathological influences as well which induce structural and functional changes of elasic fibres (EF) forming it. The present work has the purpose to make an ultrastructural characteristics of EF during the main stages of their development with a view to the differentiation of definite elastic skeleton in the aortic wall. This will contribute to more complete understanding of age-dependent and pathological alterations which set in in the structure and function of elastic formations and reflect on aortic wall elasticity.

Material and methods

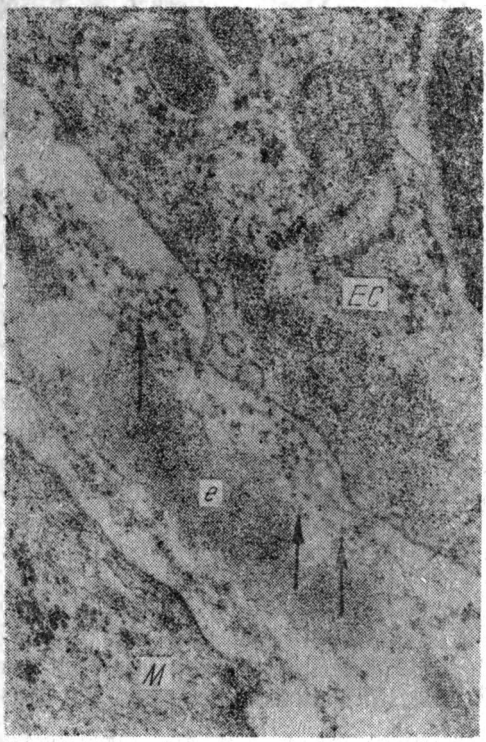
Thoracic aorta of 30 white rats (breed Wistar) of different pre- and postnatal age was electron microscopically investigated. Material was processed after a standard method for electron microscopy. An electron microscope JEM 7A was used in our study.

Results and discussion

EF are bicomponent systems under the electron microscope. They consist of a central amorphous part (protein elastin — 3,4) and peripherically located filaments formed by structural glycoproteins (2, 5, 7).

In 5-day old fetuses aortic wall presents a closed endothelial tube with 1—2 muscular layers. There is no structuralized fibrillary material in the extracellular space. However, endothelial and smooth muscle cells both are morphologically readily to synthesize and produce its precursors. In 15-days old fetuses developing elastic system of the aortic wall consists of 5—6 musculo-elastic units each of which possesses a smooth muscle cell layer of predominantly secretory type as well as a layer of elastic formations at different phases of maturity and structural organization. Subendothelial space is occupied by granulo-filamentous material with filaments dropped in. Some of them are on transverse section 8—13 nm in size and with tubular shape. Tubular wall is not continuous but consists of 3—5 subunits. In single cases filaments have only a central enlightenment and tubular wall subunits cannot be distinguished. According to the ultrastructure described, we can consider them filaments of elastic fibres (fig. 1 and 2). Aggregations of amorphous elastin can be observed in the subendothelium. Together with surrounding filaments it initiates the first bicomponent EF (fig. 1). In the media among smooth muscle cells there are large aggregations of elastic materi al

presenting incompletely fused EF aggregations forming incomplete elastic lamellae. Borderlines between single fibres within the aggregations are outlined



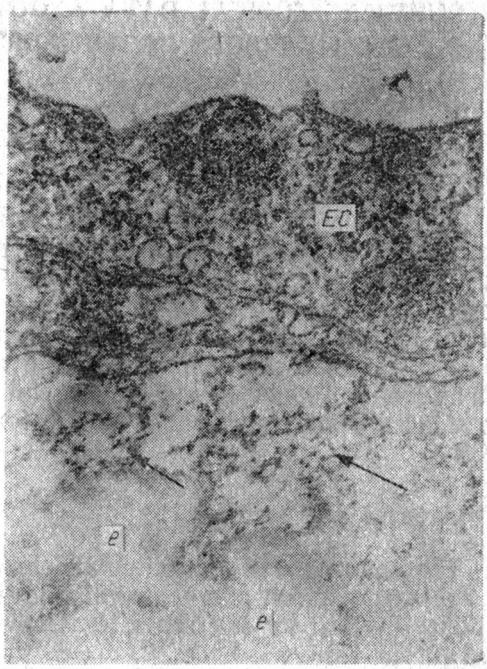


Fig. 1. Thoracic aorta — 15-day old fetus.
Subendothelial space. Filaments of elastic fibres (arrows). Elastin (e). Endothelial cell (EC). Myocyte (M).

Fig. 2. Thoracic aorta — one-day old rat.
Subendothelial space. Elastin (e). Filaments of elastic fibres (arrows). Endothelial cell (EC)

by a stronglier contrasted regions with granulous shape. During EF maturation these regions gradually disappear and elastin remains regularly weakly contrasted.

In newborn animals subendothelial space continues to be occupied by filamentous material and single EF. The amount of amorphous elastin is strongly increased (fig. 2). We establish a dense contact between the amorphous elastin and the basal part of an endothelial cell first in this age group. Such cell-elastin contracts occur only in the limits of the media in prenatal age, too. In the media elastin lamellae can be seen which occupy an even greater part of the space amids smooth muscle cells and of the vascular circumference as well.

In 18-day old animals it is to be noted that there is a strongly expressed reduction of subendothelial filaments on the account of amorphous elastin augmentation. Single EF occur more rarely, commonly in proximity of the first elastic lamella which is already almost completely formed. In the interior of the lamellae elastin is weakly contrasted as a contrary of its peripherical parts where

single EF undergo fusion with it. Elastic lamellae are completely formed in the media. They are thick, with regularly weakly contrasted elastin and scanty filament number. Amids them single or organized in bundles collagen fibres, 25—40 nm in size, and with manifested periodicity begin to appear.

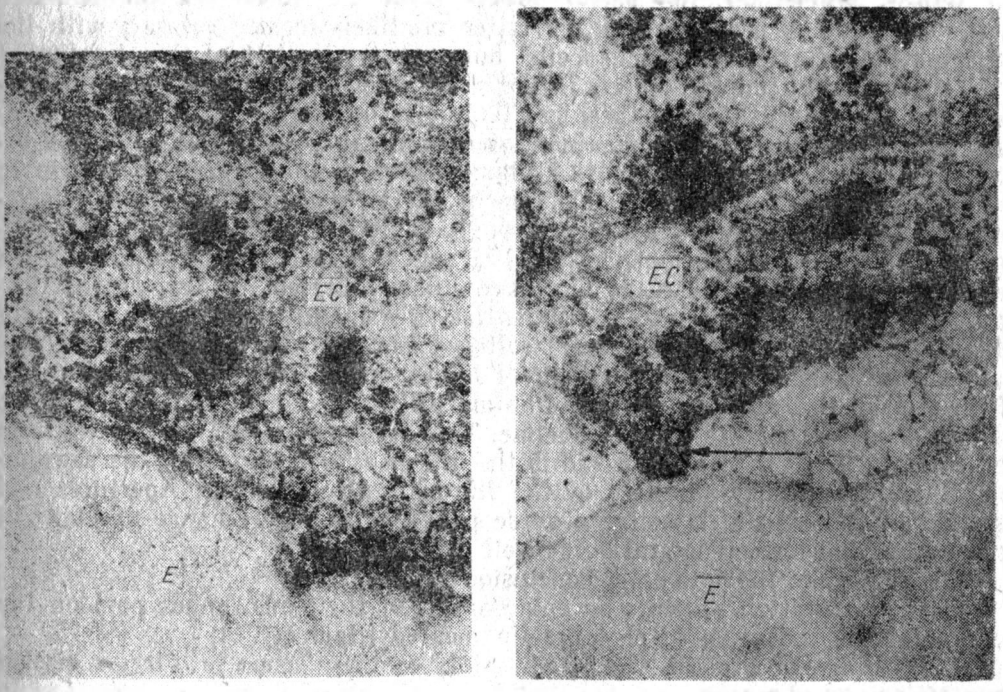


Fig. 3. Subendothelium of thoracic aorta — 45 day old rat.
a. First elastic lamella (E) is located closely to the basal membrane of endothelial cells (EC).
b. Larger free spaces remain only among basal processes (arrow) of endothelial cells (EC) attaching to the elastic lamella (E)

In 45-day old animals the elastic skeleton of the aortic wall is formed by mature elastic lamellae. The first one is located in the close proximity of the basal membrane of the endothelial cells, rather densely in some places (fig. 3-a). Only amids basal processes of endothelial cells attaching to the lamella one can see larger free spaces (fig. 3-b). In the media elastic lamellae are mature with characteristic weak contrast of the elasin and a small amount of peripheral filaments. The lamellae formed do not present a continuous barrier between the cells. They possess fissurae and apertures filled with collagen fibres, filaments, and even cell evaginations (8).

EF differentiation in the aortic wall includes their origination by a definite way, their growth by elastin sedimentation on a network of filaments to a definite critical size followed by fusion and enlargement of elastin aggregations resulting in elastic lamella formation. It is known that first EF filaments appear in the course of elastogenesis (2, 6). Then an elastin sedimentation on them sets in (5). By this way the first bicomponent EF are formed. We establish that their growth is realized, by the one hand, by means of an additional amorphous elastin sedimentation, and by fusion of single EF and their enlargement into complex

aggregations, on the other hand. Newly-formed EF are rich in filaments and their elastin is irregularly contrasted. With age peripheral filaments reduce strongly in number but elastin remains regularly weakly contrasted. Therefore, the wealth of filamentous material and the single subendothelial EF described in 15-day old fetuses argues for the active processes of EF formation taking place in the intima. The precursors of the latter are likely formed not only with the participation of smooth muscle cells but also with that of the endothelial ones. At the same time the process in the media is in a considerably more advanced phase — among the cells there are large elastin aggregations formed by fusing EF. This process continues in newborn animals, too. There are quantitative differences, namely an essential augmentation of the amorphous elastin in comparison with the fetuses. The enlargement of elastin aggregations leads to elastic lamella formation in the media. A strongly expressed reduction of the filamentous material and of single subendothelial EF sets in with 18-day old animals when the first elastic lamella is already almost completely formed. This argues that formation of new EF decreases but differentiation is presented mainly by fusion of EF into lamellae. The appearance of collagen fibres in the media of the aortic wall can be considered an expression of higher-degree morphological maturity of the wall in this age. In 45-day old animals fibro-elastic skeleton of the aortic wall is formed by mature elastic lamellae. The presence of cell-elastic contacts and collagen fibres in the media and in the intima as well argues for the morphological and functional maturity of the fibrous aortic skeleton. Apertures and fissurae in the elastic lamellae provide possibilities for exchange of material information and for cell migration as well (9).

We can make the following conclusions:

1. EF maturation presents an enlargement of their amorphous part on the account of the reduction of peripherally located filaments.

2. With age EF growth is realized mainly by their fusion into larger elastin aggregations or plaques.

3. The differentiation of the elastic system in the aortic wall is of longer duration in more active zones in respect to growth and adaptation — intima and borderline media regions.

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СУБМИКРОСКОПИЧЕСКИЕ ДАННЫЕ О ДИФФЕРЕНЦИАЦИИ ЭЛАСТИЧЕСКИХ ВОЛОКОН АОРТНОЙ СТЕНКИ

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РЕЗЮМЕ

Методом электронной микроскопии исследована грудная аорта белых крыс линии Вистар в различном пре- и постнатальном возрасте. Сделана ультраструктурная характеристика эластических образований (волокон, пластинок) во время их развития — с момента их первоначального появления до образования окончательного эластического скелета аортной стенки. Дифференциация эластической системы аорты протекает с различной продолжительностью в различных зонах стенки. В зонах с более интенсивным ростом и более активной адаптацией дифференциация длится больше. Такими зонами являются интима и граничные участки медии.