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AN Gansburgsky^{1*} and AV Yaltsev²

¹Department of Histology, Cytology and Embryology, Yaroslavl State Medical University, Russia

²Department of Pathological Anatomy with the Course of Clinical Pathology, Yaroslavl State Medical University, Russia

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***Corresponding author:** AN Gansburgsky, Department of Histology, Cytology and Embryology, Yaroslavl State Medical University, Russia, E-mail: profang@mail.ru

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Research Article

Vascular Smooth Muscle Cells in the Branching of Renal Arteries

Annotation

Histological, morphometric, cytophotometry and statistical methods studied isolated smooth muscle cells at sites of the renal arteries with different hemodynamic conditions in newborns. It is shown that the population of smooth muscle cells on the straight sections of the renal arteries represented subpopulations of small, medium and large mononuclear cells. In the area of the arteries blood flow dividers, characterized by turbulence, twists, recycling and complex distribution of shear stress on the vascular wall, there is a pronounced polymorphism in the structure of the arteries of the population. Smooth muscle cells in these hemodynamic conditions, undergo hypertrophy and hyperplasia polyploidization. Phenotypic modulation myocytes occurring in arteries with high branching, anastomosing, many turns may initiate the development of hypertension, heart failure, ischemia, diabetes and obesity.

Abbreviations

SM: Smooth Muscle Cells; DNA: Deoxyribo Nucleic Acid; MCM: Micrometer; NM: Nanometer

Introduction

Areas of vascular branching attracted the attention of researchers since the mid 50-ies of the last century [1, 2]. Dividers blood flow correspond to areas of increased permeability of the endothelial lining and are prone to the formation of atheroma [3, 4]. As the analysis of the literature [5], the least studied structural component of the vascular wall in hemodynamic stress zone are smooth myocytes (SM). It is known that SM can acquire branched form, and their contact with the basement membrane of endothelial cells alone [6]. Hemodynamic conditions may be a factor influencing the properties of the SM of the vascular wall [7].

The purpose of work

Morphometric and cytophotometry analysis isolated smooth muscle cells in arteries areas with different hemodynamic conditions.

Material and Methods

We studied the right and left renal arteries in the branch at the gates of the body 26 newborn infants who died of cerebrovascular accidents in case of damage of the skull bones. For comparison, the study area is the same vascular blood flow dividers. Specimens were fixed in 10% neutral formalin, 0.1

M phosphate buffer at pH 7.0. Isolated smooth muscle cells (SM) was prepared by alkaline cell dissociation and stained with hematoxylin and eosin Feulgen (hydrolysis of a 5 N HCl at 37°C 12 minutes). We counted the relative abundance of two-nuclear elements, screw ocular micrometer measured linear parameters of the nucleus and cytoplasm of single-core and dual-core SM, as well as calculate their area and volume. The level of DNA in the nuclei of single- and dual-core assayed for SM cytophotometry "MIF-K" (Moscow State University) at a wavelength of 580 nm. Quantitative data were processed by the method of variation statistics.

Results of the Study

Population structure of smooth muscle cells of the renal arteries of newborn children on the straight sections of the vessel and presented heterogeneous, mostly single-core elements (Figure 1). The morphometric analysis helps identify a subpopulation of small (up to 1000 mcm³ to 24.8% - (Figure 2)), medium (1000 - 3000 mcm³ to 53.4% - (Figure 3)) and larger GM (more than 3000 mcm³ 21.8 % - (Figure 4)). It was also found a small amount (0.2%) dual-core forms of SM (Figure 5). Cytophotometry analysis shows that SM population comprises mononuclear preferably diploid elements (Figure 6).

The population of the area of vascular myocytes branching arteries expressed different heteromorphic. Often there are different forms branched SM (Figure 7), with Um tortuous (twisted) nuclei (Figure 8), the peripheral position of the nucleus (Figure 9). In addition, the share of dual-core four times (to 0.8%) SM. There are muscle cells with micronuclei



Figure 1: Isolated mononuclear smooth myocytes newborn renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 600.

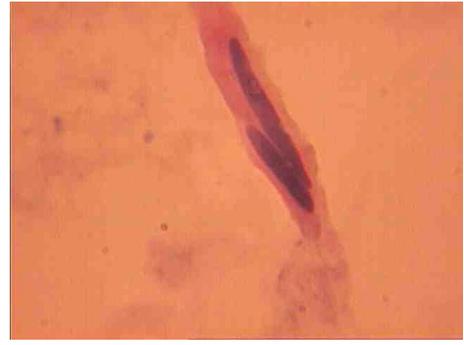


Figure 5: Dual-smooth myocyte newborn renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.



Figure 2: Small single-core smooth myocyte newborn renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.

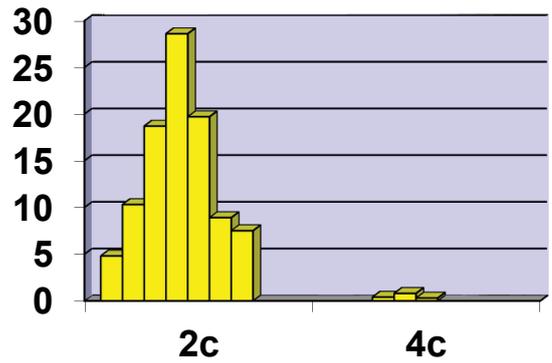


Figure 6: Histograms of DNA content in the nuclei of smooth muscle cells isolated straight sections neonatal renal artery. The x-axis: amount of DNA in SM; ordinate: relative content mononuclear smooth myocytes%.



Figure 3: The average single-core smooth myocyte newborn renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.



Figure 7: Smooth myocyte with branched end in the renal artery of the newborn. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.



Figure 4: Large single-core smooth myocyte newborn renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.



Figure 8: Smooth myocyte crimped with the core in the renal artery of the newborn. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.

(Figure 10), dividing by mitosis (Figure 11) and signs of death (Figure 12). In a population of mononuclear myocytes varies the ratio of small (31.4%), medium (40.5%) and high (28.1%) SM. Volumetric detailed pie charts (Figures 13,14) to evaluate the size of SM modulation classes in straight sections and areas of branching renal arteries newborns.

SM dual-characterized, compared to single-core, large size (Figure 15). Thus, their average length is increased to 1.8 ($P < 0.01$), and the cross section - in a 1, 7 ($P < 0,01$) times. Area and volume dual-core cell above, respectively, 4.5 ($P < 0.01$) and 10,4 ($P < 0,01$) fold. This same pattern was observed in the measurement of myocyte nuclei: their length exceeds a



Figure 12: Necrosis smooth muscle cells in the renal artery of the newborn. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.



Figure 9: Smooth myocyte with a nucleus at the periphery of the cells in the renal artery of the newborn. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.

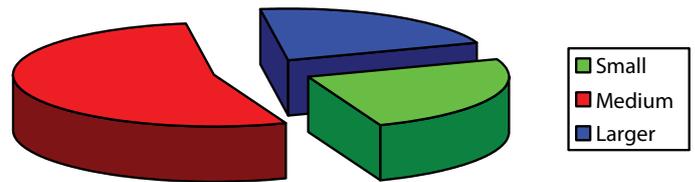


Figure 13: The structure of the population of mononuclear smooth muscle cells in the straight sections of arteries (laminar flow of blood).

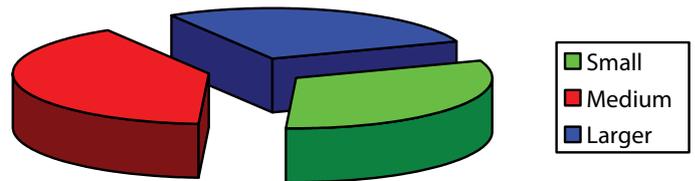


Figure 14: The structure of the population of mononuclear smooth muscle cells in the area of the arteries with blood flow dividers (turbulent blood flow).



Figure 10: Dual-smooth myocyte microkernel in the renal artery of the newborn. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.



Figure 15: The dual-core and single-core newborn myocytes renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.

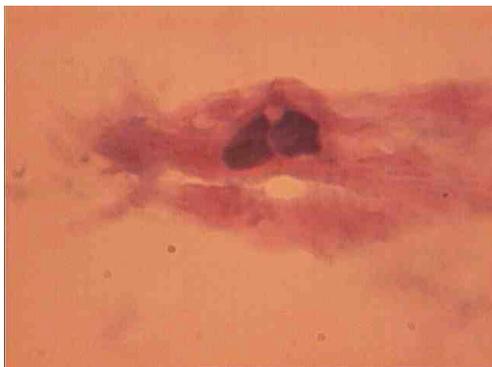


Figure 11: Telophase of mitosis smooth muscle cells in the renal artery of the newborn. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.

mononuclear 1,3 ($P < 0,01$), a cross-section - in 1,4 ($P < 0,01$), and the area and volume of a 2.8 ($P < 0,01$) and 2,7 ($P < 0,01$) fold.

Quantitative analysis of dimensional parameters of dual-core SM shows that in their composition, there are three subpopulations: small (up to 5000 mcm^3 9.4% - (Figure 16)), medium (5 000 - 10 000 mcm^3 65.3% - (Figure 17)) and large (more than 10,000 mcm^3 25.3% - (Figure 18)). The ratio

between the bulks represented in subpopulations expanded pie chart (Figure 19).

Dual core myocytes also differ in their nuclei topography. There SM with nuclei lying one behind the other (Figure 10) or in parallel (Figure 17).

Cytophotometry redistribution study shows ploidy classes in the population of smooth muscle cells in the zone of the branching renal arteries (Figure 20), Table 1).

Discussion of the Data

Unlike straight segments laminar character movement, and

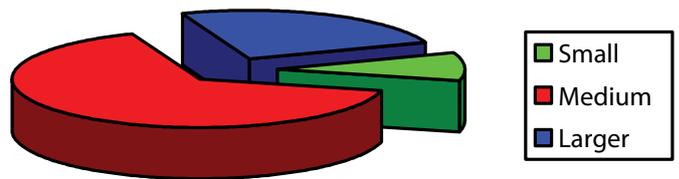


Figure 19: Population structure of dual-core smooth muscle cells in the area of the arteries with blood flow dividers (turbulent blood flow).

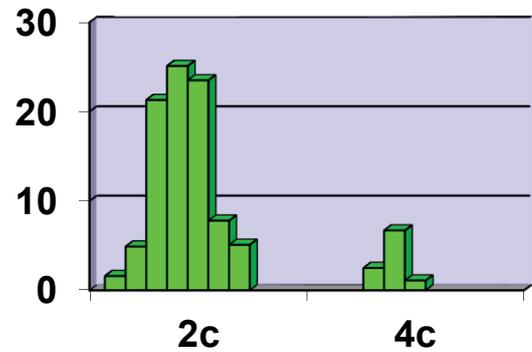


Figure 20: Histograms of DNA content in the nuclei of smooth muscle cells in isolated sections of the branching of renal arteries newborns. The x-axis: amount of DNA in SM; ordinate: relative content mononuclear smooth myocytes%.

Table: Ploidy classes of smooth muscle cells of various departments of the renal arteries of newborn.

The studied group	The percentage of each class of cells			
	2 c	2c x 2	4 c	4c x 2
The straight sections of arteries	98,5	0,2	1,3	-
The blood flow dividers zone	89,3	0,8	9,6	0,3



Figure 16: Small dual-smooth myocyte newborn renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.



Figure 17: Average dual core smooth myocyte newborn renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.



Figure 18: Patient dual-smooth myocyte newborn renal artery. Alkaline dissociation. Stained with hematoxylin-eosin. x 1000.

field bends flow dividers are special sections with turbulence, swirling, recirculation, and complex shear stress distribution on the nature of the arterial wall [5,8]. It is important to bear in mind that the pressure (stress / strain) and flow (shear) can regulate the function of blood [9], differentiation, proliferation and migration of vascular myocytes [9,10].

The morphological laboratories Yaroslavl Medical University found that areas dividers fetal blood and adult arterial flow distinguished by the presence of additional regulatory smooth complexes in the inner membrane of vessels [11,12]. SM artery walls respond to hemodynamic stress cytoskeleton reorganization and migration [13]. Development in the tunica intima of the SM and the musculo-elastic sphincter is a consequence of the penetration of SM medium choroid into the interior through the Fenestra in the elastic membrane [14]. The role of migration in SM endothelin and platelet-derived growth factor [14,15].

It was established that in the arterial wall area of blood flow dividers marked a pronounced polymorphism of smooth muscle cells, their hypertrophy and polyploidy. In the analysis of mechanisms polyploidy smooth muscle tissue was shown that growth of tangential stress and increased pressure in the vessels associated with activation of DNA synthesis in the nucleus of muscle cells [12, 16]. In this work in areas of

branching renal arteries newborns revealed mitotic division of smooth muscle cells, and increasing the number of dual-core form. It is known that various damaging effects lead to an increase in the amount of cell population's binucleate forms vascular wall [17], it is considered as a variant of polyploidy [18].

In our laboratory we show that the regeneration conditions morphogenesis rat aortic population becomes more reactive SM tunica: labeled nuclei index compared to control them in 53-fold increases in [19]. Thus, both types of SM polyploid with increasing load receptacle (turbulent motion twists and blood recirculation) are formed by polyploidy mitosis. Last according V.Ya. Brodsky & I.V. Uryvaeva [18] is a variant of the conventional mitosis stopped at one stage or another. About the origin of the mitotic dual-core elements is also evidenced by its own data, showing that the relationship between the DNA content in the nuclei of the same dual-core myocyte close to 1.0. DNA replication provides the appearance of new cells or existing polyploidy, ie strengthening of intracellular regeneration processes [20].

In the course of the study shows that the population of the SM renal arteries expressed different heteromorphic. This is especially demonstrated by the presence of small, medium and large single and dual-core vascular myocytes. It is known that SM internal organs (visceral SM) have similar sized subpopulation, where it is attributed to a different functional purpose [21]. The authors believe that a group of small SM is young, actively synthesizing protein cells having proliferative activity; SM's average form the basis of population and retain proliferative potential; SM is a large terminal elements lose their ability to proliferate.

Thus, these data confirm the information [10,21] about the heterogeneity of the SM populations. Isolated phenotype small phase (fast) SM, capable of passing through hypertrophic growth and tonic (slow) vascular SM undergoing hyperplasia [9]. Load changes can cause phenotypic modulation of myocyte proliferation and SM [9,10]. Thus, hemodynamic conditions can be a factor affecting the properties SM vascular wall. Phenotypic modulation myocytes, which occur in the arteries that are highly branched, anastomosing, many turns, are discussed to initiate the development of hypertension, heart failure, ischemia, obesity and diabetes [22].

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