



HEART MICROVESSELS AND ITS MORPHOLOGICAL EVALUATION AT OPEN HEART SURGERY.

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ABSTRACT

Coronary microvascular dysfunction encompasses the range of structural and functional changes in the coronary microvasculature, presents as microvascular spasm and microvascular angina. This is not fully understood in physiopathological term and identification of them in daily clinical practice are still not well understood. During last decade the process of intussusceptive pathological growth of microvasculature bed (MCR) was shown. In 90th years of 20th century the images of MCR profiles of heart right atrii of patients with pulmonary artery mitral valve stenosis were obtained by our team for first time. These images indicated to a pathological growth of vessels by intussusception type. Our-days study show the possibility for semiquantitative evaluation of morphological status of MCR using routinely cutting biopsy material during cardisurgical procedure at heart open surgery. On semithin epoxy slices of biopsy material has been treated and fixed into epoxy resins by transmission electron microscopy method, stained by Azur II using our staining method (patent 2844A Republic of Armenia) and investigated under light optical microscopy. The vessels profiles could be obtained with their further evaluation in points. Based on 10 evaluation criteria, include status of vessel wall, vessel formation was developed the scale of pathological growth by intussusceptive type.

KEYWORDS: Coronary microvasculature, intussusceptive grows, scale, light optical microscopy.

INTRODUCTION

Coronary microvascular dysfunction (CMD) encompasses the range of structural and functional changes in the coronary microvasculature that cause diminished coronary blood flow and consequently myocardial ischemia.^[4] Coronary microvascular disease plays a major role in determine myocardial ischemia in many cardiovascular conditions.^[12] Structural impairments accompanying CMD are predominantly characterized by intramural arteriole and capillary luminal narrowing, perivascular fibrosis, and microvascular refraction often associated with left ventricular muscle hypertrophy.^[4,22] Various methods have been proposed for the analysis of the functional state of coronary microcirculation.^[21] CMD can only identified indirectly through measurements of changes in coronary blood flow and coronary vascular resistance in response to particular triggers. In recent decades, new nosological entities have emerged such as coronary vasospasm, microvascular spasm and microvascular angina. The correct definition of these disorders is not yet fully understood in physiopathological term, and the knowledge and identification of them in daily clinical practice are still not well understood.^[21]

It's quite important to reveal structural changes of vasa vasorum treated the wall of large vessels.

The importance of the vasa vasorum in the performance of human saphenous vein (hsv) used for coronary artery bypass grafting (CABG) has been highlighted in various publication over the last 15 years.^[1] The role of the vasa vasorum and blood supply to the wall of HSV used for (CABG) is briefly discussed.^[1] The question whether the primary increase of vasa vasorum of venous wall plays an initial role in varicogenesis.^[14] During last decade have been shown that pathological vascularization is crucial for progression and morbidity of serious diseases such as cancer, diabetic retinopathy and age-related macular generation.^[23] The group of Burri, including Djonov and Patan, initiated and developed the vessel intussusception concept in blood vessels.^[16] Intussusception is a form of vascular remodeling and growth involving the splitting of vessels by extending cellular processes generated by endothelial and vascular mural cells into vascular lumen.^[2; 8] Intussusception may, however, be induced by VEGF-signaling implying that the role of VEGF in regulating vessels sprouting versus intussusception is still incompletely understood and may be content dependent.^[2,9] Vascular intussusception entails

the internal division, splitting of vessels into two daughter vessels.^[5]

However a role for intussusceptive angiogenesis in rebuilding a vessels network after ischemic muscle injury is unknown.^[6, 24]

Our previous investigation of human biopsy material at pathology revealed structural alteration of microcirculatory bed (MCR) by the type of intussusception, as well as MCR de novo vessels formation.^[10, 15, 18, 19], which indicated that these effects can be manifested as the structural abnormality of the coronary microvasculature, that can lead to CMD.^[22] This in its turn gave opportunity to come closer to the question of semi quantity gradation of the direction of pathological intussusception process.

MATERIAL AND METHODS

In current study small peaces of auricular right atria were taken a routine cardiovascular procedures.

1. Patients with pulmonary artery valve stenosis (PAVS). The archived pic has got at 90th years of 20th century in Institute of Michaelyan had not been published earlier;
2. Patients with coronary artery disease (CAD) - 4 patients.
3. Patients with rheumatic heart disease (mitral valve replacement) - 2 patients.
4. HSV, used at surgery (CABG) -2 patients.



Fig. 1: (archive) x 400.

The constriction in vessels of MCR is formed. The vessels wall is slightly thickened.

At patients with CAD the degree of MCR vessels involving in pathological process was expressed in majority vessels and showed up in vessels wall thickness alteration, as well as its twisty.

Typical for this disease was formation of De novo vessels. It must be noted that the thickness of formatted vessels are different.

Small pieces of investigated tissue have immediately put in a cold mix of paraformaldehyde and glutaraldehyde, with following post fixation in %OsO₄, then dehydration in ascending series of spirits, acetone, and embedded into epoxy resins.

Observation under light microscope; semithin epoxy slices, with up to 1 mkm thickness were made using ultra cut Reichert (Austria), stained with Azur II and studied under light microscope supplied 40x10 ocular lens.

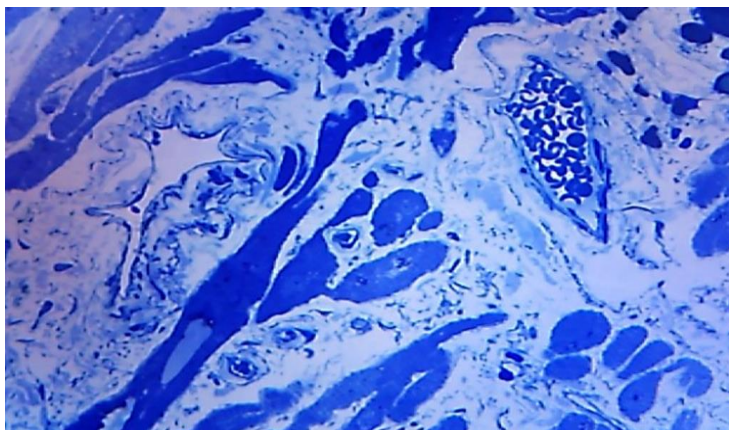
All procedures involved human subject were approved by institutional revent board bioethical committee (Yerevan State Medical University RA) conformed to the Legal Aspects of Research Ethics and Science in European Community directive (2001/20/EC).

GOALS OF STUDY

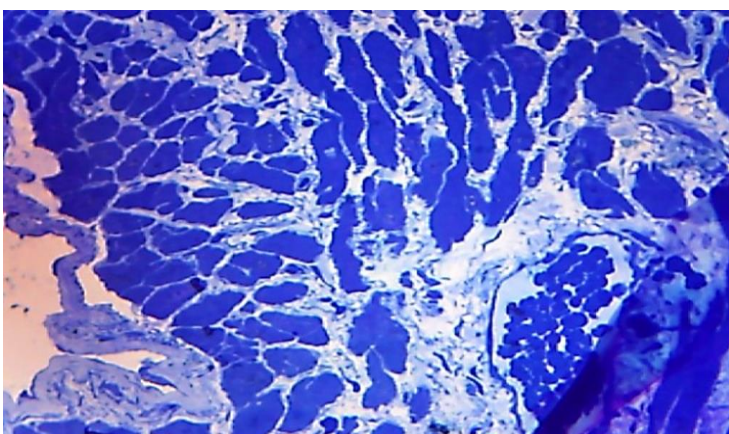
Creation of 5th scare scale of pathological growth of the microcirculatory bed vessels on the intussusception type based on the previously obtained dates as well as a new one.

RESULTS

As have shown the results of our study, single patients with PAVS at expressed pathological processes revealed the pathological growth of MCR by the type of intussusception;

**Fig. 2: x400**

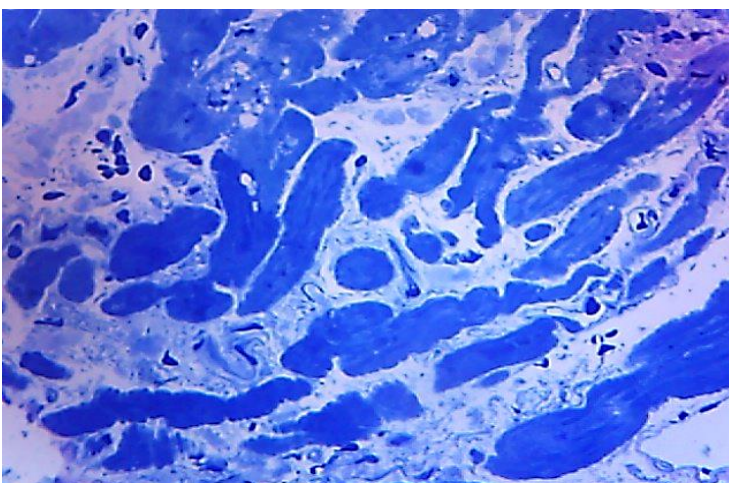
Alteration of arteriole walls, the presence of its twisty and thickness. Formation of De novo vessels. 3 such vessels are observed.

**Fig. 3: x 400**

Merging the opposite walls of arterioles and formation of a new lumen.

Patients with rheumatic heart disease the level of heart vessels wall's alteration expression is not significant

compare to patient with CAD. At the same time the process of De Novo vessels formation present.

**Fig. 4: X400**

Vessels wall is not significantly thickened. The separated De Novo vessels are presented.

Vasa vasorum HSV also involved in the process of pathological growth by intussusception type.

Along the vessels way they have irregular lumen from very wide in some parts to very narrowed in others, which could be a problem during Venus using at cardiosurgical corrections at aorta coronary bypass.

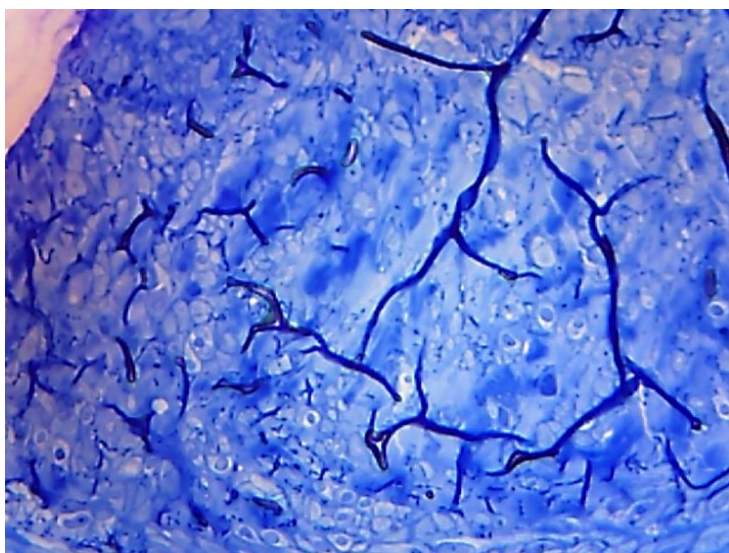


Fig. 5: x400

The process of formation and increasing of MCR Venus arterioles by intussusception type is expressed.

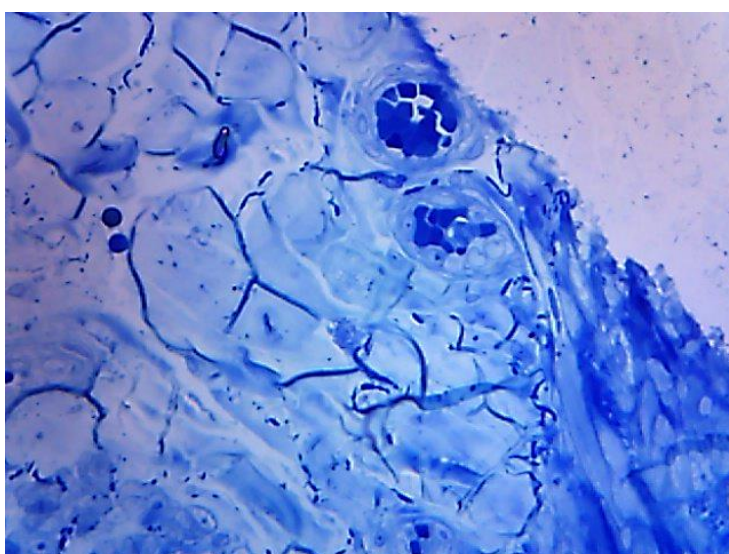


Fig. 6: x400

Formation of MCR vessels de Novo. Close location of 2 not completely separated profiles. Increasing of MCR capillaries by intussusception type.

In current study is comparison of different pathological material the process of pathological growth of capillaries MCR and as a result formation of de NOVO vessels was occurred.

This as well as previous studies (.....) gave us opportunity to move from visualisation of characteristic of MCR structure to semiquantify score. Based on 10 grade criteria, taken into consideration of the vessel profiles lumen, process of de Novo Vessel formation the 5 degree morphological scale of pathological growth of MCR vessels was developed.

The scale of pathological growth of microcirculatory bed vessels by intussusception type.

DISCUSSION

MCR, its morphology increase a quiet big interest as in a science as well as in clinical medicine. The process of pathological growth MCR by intussusception type become more actual in our days.

Vascular intussusception entails the internal division or splitting of a vessel into daughter vessels. Vessel splitting begins with the formation of peculiar endothelial based projection that cross from one side of the lumen to the other, a structure referred as pillar.^[5;6;17;24] Pillar formation has been mostly studied in the lung and extra embryonic circulation but has been observed in muscle following forced expression of VEGF.^[8;9]

New dates indicate that intussusceptive growth in skeletal muscle is a slowly flowing process.^[11] Study of skeletal muscle mainly affect capillary remodeling but do not take into consideration alterations take place in arterioles. Also there are few morphological studies of MCR on byoptates of human skeletal muscle.^[15,19]

Different methods are used for MCR study. One of the method is MCR investigation on semithin epoxy slices with thickness up to 1 mkm with its staining by Azur II.^[20]

The material samples were treated by standard method used in traditional transmission electron microscopy with following investigation of obtained semithin slices for MCR study, which will let discover mechanisms, take place in MCR structure of heart muscle at pathology as well as vasa vasorum HSV for coronary artery bypass grafing. (Fig 2,3,4,5).

It must be mentioned that in 90th years of 20th century, during investigation of hypertrophy of right parts of heart changes of cardiomyocytes of patients with PAVS using transmission electron microscopy were revealed structural which let us think about alterations of MCR.

Also was conducted light optical study on semithin epoxy slices stained by Azur II and for first time phenomenon of pathological growth of MCR by the type of intussusception in the heart muscle was revealed (Fig 1).

Our new studies give opportunity for easy visualisation and score of MCR structural status. This make closer to solving the semiquantify score of MCR in scale by 5 degree scale and give opportunities of drug therapy.^[4, 22, 23]

The scale of pathological growth of microvessels by intussusceptive type.

Group	Investigation signs	Evaluation of vessels growth	Mark
I	The growth of vessels lumen. A bit expanding of vessels wall, with rarely presense of invagination into some vessels walls, and presence of thin wall in other vessels.	Insignificant changes	1
II	The lumen of vessels are different, often present expended one with involving of some vessels. Takes place decreasing process of uneven expanding, with often presence of invagination into wall, with connection of neighbour area. In such vessels wall forming of 5 new lumen. In other vessel observed thin, sometimes scrolled wall, as well as convergence of opposite sides of first vessels.	Slightly viewable changes	2
III	Extra growth of vessels profile in intestical space between tissue cells. The lumen of vessel is thin - expanded. The wall of many vessels is uneven expended, scroled, invaginated to the wall direction, forming new lumen in the vessel wall and thin scroled wall presence in others. Viseable enclosing of oposite walls of vessels. Often obtained formation of new bridges in the vessel lumen. Often observed presence of vessels with not completely divided wall close to each other.	Temperately viewable changes	3
IV	Extragrowth of vessels profile in intestical space between tissue cells with termination of lasts. The lumen of vessels is thin-small. The vessel wall is quite uneven expanded and scrolled up to one profiles and thin, extremely scrolled up to others. The opposite walls of many of vessels reached close to each other creating bridges in vessels lumen. At the same time observed vessels profiles located close to each other, which have the one wall, and are not separated totally — multiple.. De novo different size vessels creation - singular.	Viewable changes	4
V	Formation of de novo profiles from small to very small: multiple	Brightly viewable changes	5

CONCLUSION

Based on 10 criteria, the 5 degree scale for evaluation of pathological growth of microcirculatory bed vessels by the type of intussusception was made. This give opportunity to evaluate the changes and give prognosis for its further state.

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