
Adaptability of Heart Tissue to Repeated Ischemia and the Efficiency of Recovering Processes

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The purpose of this experimental study was to determine quantitative changes of the connective tissue in all parts of the hearts, to point out a correlation between these changes and the morphological state of cardiomyocytes of repeated ischemia conditions. We have also investigated the influence of hyaluronidase on the process of the development and healing of myocardial infarction.

Experiments were carried out on 55 rabbits weighing 2.5–3.0 kg. The control group consisted of 5 rabbits and the study groups comprised 50 rabbits. Experiments in groups 1–6 and 10 were done inducing intravenous thiopental anesthesia (ITA). Rabbits in all groups were investigated functionally and morphologically. In groups 1, 6 and 10 measurements were taken of the ischemia basin and its adjacent areas, using an original electrode (5), and analysed according to ST interval dislocation in epicardiograms – Ae. Experimental data on myocardial function were compared with morphometric investigation results.

The investigation has shown that under conditions of chronic transitory short-duration occlusions of the coronary arteries, the cardiomyocytes apparently adapt to ischemic affects. On the grounds of such changes, a 20-minute occlusion of the coronary arteries results in less pronounced irreversible changes in the myocytes than those in the non-adapted myocardium.

Repeated myocardial infarction is a rather frequent condition in medical practice. Permanent occlusion of coronary arteries induces morphological changes in the tissue areas nourished by the vessel of the heart. Initially, there is a necrosis and some time later a scar is formed. Repeated myocardial infarction in a heart affected by the first infarction develops with difficulty, yielding much deeper pathological and morphofunctional changes in the cardiac tissues. The use of hyaluronidase in such cases helps to decrease the expressiveness of the pathological process. This is reflected in the quantitative changes of interstitial tissue and in cardiomyocyte nucleic acids, this condition of the normalization process is particularly evident after treatment with hyaluronidase.

Key words: experimental myocardial infarction, heart tissues, cardiomyocyte, repeated ischemia, hyaluronidase

INTRODUCTION

As a result of coronarogenic ischemia, the processes taking place in the heart wall deviate from the norm, depending on the duration and degree of the disturbance of coronary circulation (2, 3). The developing morphofunctional changes are of either reversible or irreversible nature. Such processes take place in the heart tissues not in isolation from other systems of

the body. Investigating experimental ischemic heart disease in order to explicate the questions of imminent danger and treatment, it is necessary to impartially evaluate the depth of pathological processes, their dynamics and development trend before resorting to therapeutic measures (1, 2, 6).

MATERIALS AND METHODS

Experiments were carried out on 55 rabbits weighing 2.5–3.0 kg. The control group consisted of 5

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Table 1. Structure of the experiment

Group No.	Type of occlusion (PO – permanent, OD – occlusion duration) and treatment, n = 55	Duration (days) of experiment
0	control group of healthy rabbits [5 rabbits]	
1	PO of anterior branch of left coronary artery [5 rabbits]	5
2	PO of anterior branch of left coronary artery [5 rabbits]	30
3	PO of right coronary artery [5 rabbits]	65
4	PO of right coronary artery and PO anterior branch of left coronary artery on the 30th experiment day [5 rabbits]	65
5	PO of right coronary artery followed by PO of anterior branch of left coronary artery and subcutaneously administered hyaluronidase 500v/kg every other day [5 rabbits]	65
6	OD 20 min [5 rabbits]	5
7	OD 5 min 2 times daily everyday [5 rabbits]	5
8	OD 5 min 2 times daily everyday and later OD 20 min [5 rabbits]	10
9	OD 5 min 2 times daily everyday, later OD 20 min and subcutaneously administered hyaluronidase 500 v/kg every other day [5 rabbits]	10
10	OD 20 min and treatment with hyaluronidase 500 v/kg [5 rabbits]	5

rabbits and the study groups of 50 rabbits. Experiments in groups 1–6 and 10 were done inducing intravenous thiopental anesthesia (ITA).

Rabbits in all groups were investigated functionally and morphologically. In groups 1, 6 and 10 measurements were taken of the ischemia basin and its adjacent areas, using an original electrode (5), and analysed according to ST interval dislocation in epicardiograms – Ae. Experimental data on the myocardial function were compared with morphometric investigation results. Stereologic-morphometric myocardial studies were carried out on the fifth day of the experiment. Serial total histological sections performed by Masson dye method were used for the study. Total heart sections and their necrosis zone were determined with the help of photo enlarger and depicted on paper. Stereologic-morphometric measurements were performed with an “infarct ruler” of our own construction. Nucleic acid concentration was investigated with a luminescence microscope (histological sections being stained with acridine orange) and the volume of the interstitium in the myocardium was assessed by calculating the ratio of the area taken by interstitium and the investigated area of the myocardium. Macro devices fixed to the Opton microvideomat were used for investigation of microphotographs made of histological micropreparations stained by original technique (orange used for cardiomyocytes and dark blue for the interstitium).

RESULTS AND DISCUSSION

The results of our study demonstrated that following 20 min of coronary occlusion, transmural my-

ocardial necrosis on the fifth day developed in the anterior cardiac wall and the interventricular septum in 80 per cent of cases, and small focused necrosis developed in the rest of cases. The necrosis comprised 9 to 10 per cent of the total cardiac muscle volume.

During occlusion a single medicamental injection reduced the incidence of transmural myocardial necrosis to 50 per cent. The volume of necrosis on the fifth day was reduced to 5 per cent. These positive changes or morphological parameters of myocardial necrosis corresponded to positive ECG dynamics. A comparative analysis of the results of morphofunctional investigations (Figs. 1–3) has revealed the following facts: the recorded epicardiograms show that deviation of ST segment is an informative index, and the analysis of these indices makes allows assessing changes of coronary blood circulation in the ischemia basin of the occluded blood vessels as well as in the adjacent areas; during occlusion, maximal changes in the heart wall tissues develop in the central part of ischemia basin, and they are much less in the outlying area; at the beginning of occlusion, due to inner reserves and the remains of blood circulation as well as involvement of colateral circulation, there are attempts to compensate for the developing morphofunctional disturbances in cardiomyocytes and their adjacent tissues; in the course of occlusion of coronary blood vessels, pharmacological preparations were used, helping to limit the depth and scope of the pathological processes; searching for some new therapy methods of ischemic heart disease, using analysis of epicardiograms, it is easier to select the proper time, dose, and structural complex of medicaments and to assess the

effectiveness of experimental treatment at an early stage of acute ischemia; the use of original methods of functional investigation in the experiment allowed us to prognosticate the scope of the possible necrosis basin in later experiments, using only the data of functional investigations obtained during the first hours, thus making it possible to compare them with the morphometric data of analysis of a stereological picture of heart wall necrosis in an analogous ischemia basin evaluating the areas of maximal, average and minimal level of damage of cardiomyocytes (Figs. 4, 5).

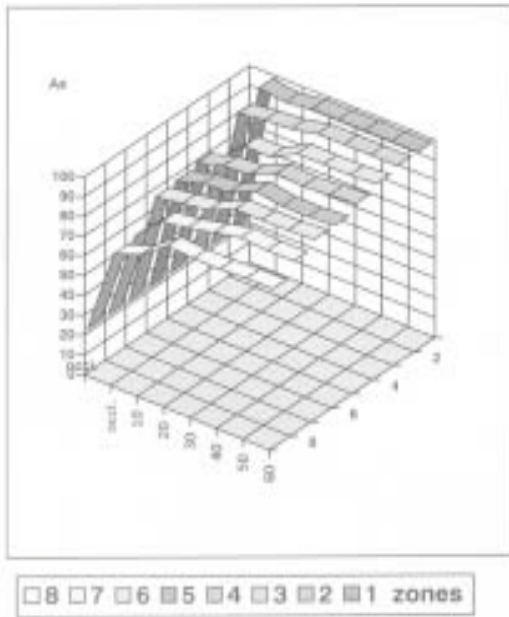


Fig. 1. Dynamics of changes of the index of ST interval dislocation (Ae) in epicardiograms of the ischemia basin during and after occlusion (group 1)

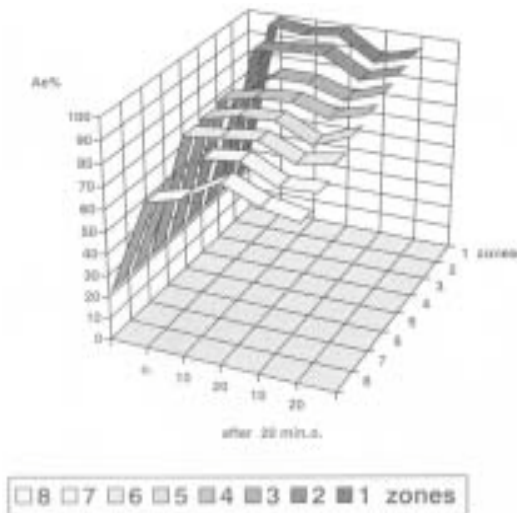


Fig. 2. Dynamics of changes of the index of ST interval dislocation (Ae) in epicardiograms of the ischemia basin during and after occlusion (group 6)

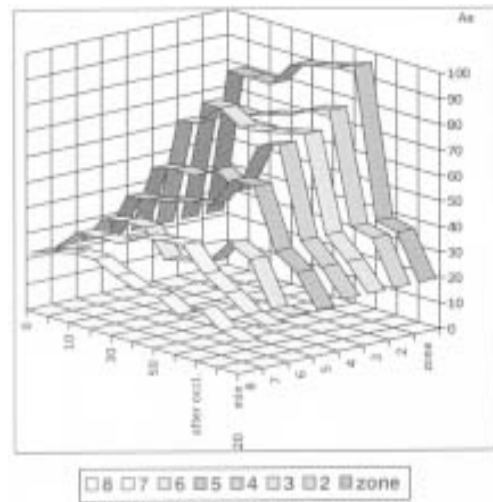


Fig. 3. Dynamics of changes of the index of ST interval dislocation (Ae) in epicardiograms of the ischemia basin during and after occlusion (group 10)

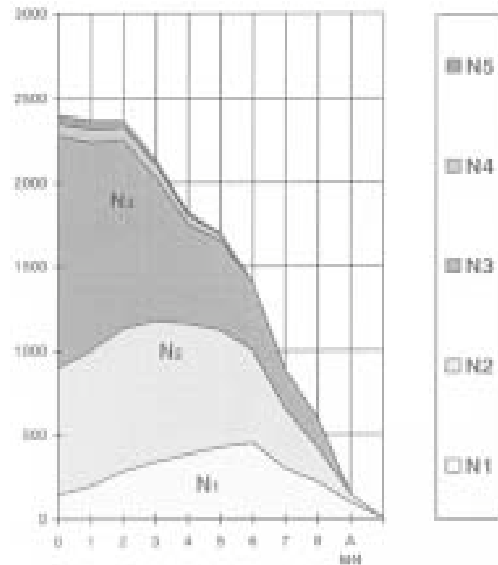


Fig. 4. Results of morphometric investigation of the area of myocardial necrosis (occlusion 20 min) on the 5th day of experiment. AMN – area of myocardial necrosis, N1 < 10%, 30% > N2 > 10%, 60% > N3 > 30%, 75% > N4 > 60%, N5 > 75%

Our results have shown that permanent occlusion of coronary vessels induces morphological changes in the heart tissues in the area of the nourishing vessel, with necrosis developing first, followed by cicatrization of the area. Quantitative changes also develop in the interstitium of the cardiac wall. These changes manifest themselves differently in the periinfarct area and in the intact left or right ventricular areas (Fig. 6). Normalization of the amount of interstitium in the cardiac wall can be markedly affected by hyaluronidase treatment. Repeated myocardial infarction in the heart affected by the first infarction develops in aggravated conditions, and the

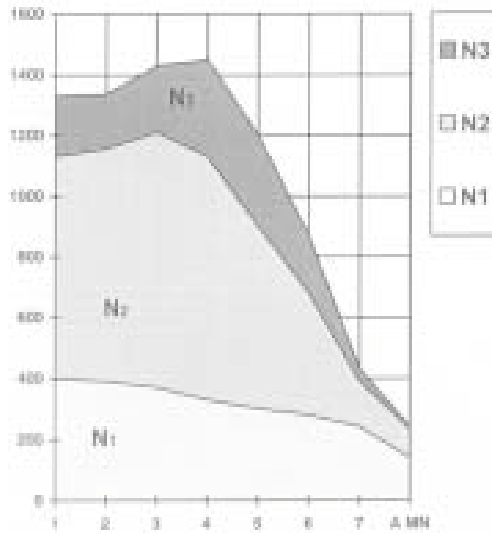


Fig. 5. Results of morphometric investigation of the area of myocardial necrosis (occlusion 20 min and treatment) on the 5th day of experiment. N1 < 9%, 30% > N2 > 10%, 60% > N3 > 30%

pathological morphofunctional changes in the myocardial tissues become more pronounced. Investigation of quantitative changes in the cardiomyocyte nucleic (Fig. 7) acids has contributed to a better understanding of the structural reorganization of the affected cardiac muscle. This is due to the fact that adaptation mechanisms of this organ and the intracellular regeneration processes are closely linked with nucleic acids. It has been found that quantitative normalization processes of cardiomyocyte nucleic acids are positively affected by hyaluronidase. Nucleic acids of cardiomyocytes vigorously react to the developing changes in the myocardium due to coronarogenic ischemia. This reaction takes place in all cardiac areas, but its intensity differs.

Recurrent ischemia of the myocardium presents a relevant problem, as understanding the interrelation between the duration of the disturbed coronary circulation and the dynamics of structural and functional changes developing in response to ischemia, which lead to destruction of the cardiac tissues, especially in the early stages of the disorder due to repeated coronary spasms, extends the possibilities of prevention and restriction of the pathological changes. The investigation has shown that under conditions of chronic transitory short-duration occlusions of the coronary arteries, the cardiomyocytes apparently adapt to ischemic effects. On the ground of such changes, a 20-minute occlusion of the coronary arteries results in less pronounced irreversible changes in the myocytes than those in the non-adapted myocardium. During the process of the development and healing of myocardial infarction a connective

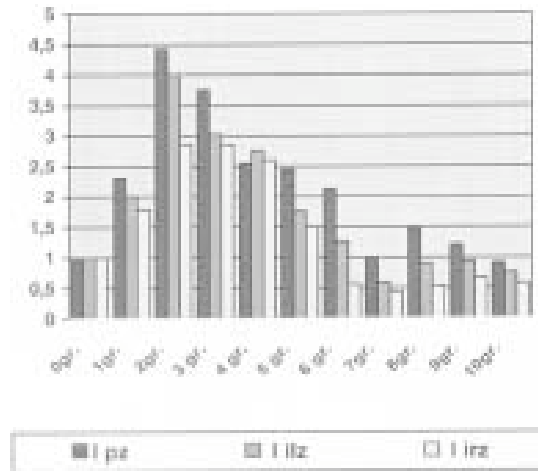


Fig. 6. Quantitative changes of interstitial tissue during MI. I – quantitative ratio of interstitial tissue in experimental group as compared to that of healthy myocardium: pz – periinfarction zone, ilz – infarction zone of the left ventricle, irz – intaction zone of the right ventricle

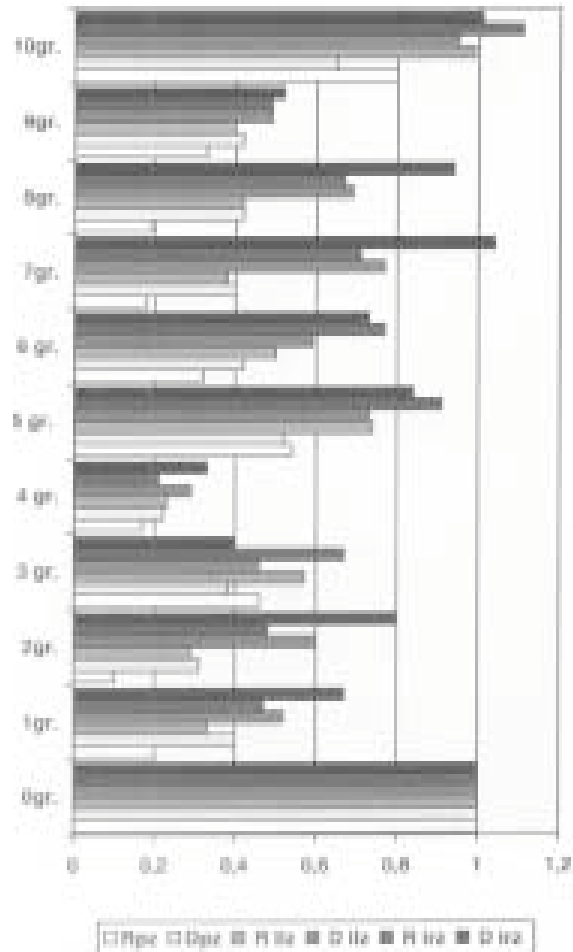


Fig. 7. NA concentration in the cardiomyocytes in the zones of MI: pz – periinfarction zone, ilz – intaction zone of the left ventricle, irz – intaction zone of the right ventricle; R – RNA/RNAo, D – DNA/DNAo, Nao – nucleic acid of the myocardium of healthy experimental animals, NA – nucleic acid of the myocardium of experimental animals

tissue scar is formed in the zone of circulation disturbance. Morphological changes are taking place in the whole heart. The purpose of this study was to determine quantitative changes of the connective tissue in all parts of the heart, to point out a correlation between these changes and the morphological state of cardiomyocytes. We have also investigated the influence of hyaluronidase on the process of the development and healing of myocardial infarction.

CONCLUSIONS

1. The recorded epicardiograms show that deviation of ST segment is an informative index, and the analysis of these indices makes it possible to assess the changes of coronary blood circulation in the ischemia basin of the occluded blood vessels as well as in the adjacent areas.

2. The use of original methods of functional investigation in the experiment allowed us to prognosticate the scope of the possible necrosis basin in later experiments, using only the data of functional investigations obtained during the first hours, thus making it possible to compare them with the morphometric data of analysis of a stereological picture of heart wall necrosis in an analogous ischemia basin, evaluating the areas of maximal, average, and minimal level of damage of cardiomyocytes.

3. Due to permanent occlusion of coronary arteries myocardial infarction develops in the cardiac wall, embracing about 10% of the entire volume of the cardiac muscle on the 5th day of experiment, and on the 30th day a scar is found in the place of infarction.

4. Neither the amount of nucleic acids of cardiomyocytes nor the volume of interstitium can restore itself in the cardiac wall on the 30th and 65th day of experiment, the normalization process taking place due to the effect of hyaluronidase.

5. In the presence of permanent occlusion of the left coronary artery or of the right one, the quantitative changes of nucleic acid in the interstitium of the heart wall are different, this being associated with the peculiarities of blood circulation in the heart of rabbits.

6. Restorative processes of heart tissues affected morphologically and functionally by primarily sustained myocardial infarction are found to be aggravated following a repeated infarction, and this is shown by changes of NA in the cardiomyocytes and in the cardiac interstitium.

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ŠIRDIES AUDINIŲ ADAPTACIJA PASIKARTOJANČIAI IŠEMIJAI IR ATSISTATYMO PROCESŲ EFEKTYVUMAS

S a n t r a u k a

Šis darbas yra eksperimentinis. Jame panaudoti 55 triušiai, kurie sudarė 10 grupių. Modeliuota laikino ir permanentinio pobūdžio širdies audinių ischemija, kad būtų galima ištirti pasikartojančios širdies audinių ischemijos poveikį širdies audiniams. Mūsų tyrimo rezultatai parodė, kad širdies audiniuose dėl ischemijos vykstantys pakitimai priklauso nuo jos trukmės. Ischemijos laikas, po kurio įvyksta negrįžtami pokyčiai kardiomiocituose, yra dvidešimt minučių. Dvidešimt minučių trukmės bei permanentinė vainikinių širdies kraujagyslių okliuzija kraujagyslės maitinamame balseine sukelia eksperimentinio gyvuliuko širdyje morfologinius širdies audinių pokyčius. Iš pradžių vystosi nekrozė, vėliau čia formuojasi randas. Trumpalaikė bei laikino pobūdžio ischemija „treniruoja“ miokardą deguonies stokai, širdies audiniuose vyksta adaptacinio pobūdžio persitvarkymai. Pakartotinė miokardo ischemija jau pakenktoje po pirmojo miokardo infarkto širdyje vystosi sunkesnėmis sąlygomis, patologiniai morfofunkciniai širdies audinių poslinkiai yra gilesni. Mūsų eksperimente hialuronidazės panaudojimas padėjo sumažinti patologinio proceso išreikštumą. Tai atsispindi tiek širdies intersticijumo, tiek kardiomiocitų nukleino rūgščių kiekybiniuose poslinkiuose, jų kitimų normalizacijos linkme taikant gydymą.

Raktažodžiai: eksperimentinis miokardo infarktas, širdies audiniai, kardiomiocitas, pakartotinė ischemija, hialuronidazė

Received 2 April 2001

Accepted 1 June 2001