Chapter 16

Morphology of Right Atrium Myocytes

Marina L. Bugrova

Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/intechopen.76311

Abstract

The chapter of this book is devoted to characteristics of the ultrastructure and function of secretory cardiac myocytes of the right atrium in norm and the experimental pathology in rats. The data were obtained at various models, such as clinical death, renovascular hypertension in rats, and in the isolated Langendorff-perfused rat hearts. We investigated the effect of the drug Mexidol on morphological and functional characteristics of endocrine cardiac myocytes in rats and in the isolated heart. Quantitative data on intracellular structures of atrial myocytes were presented. We estimated the accumulation of the atrial natriuretic peptide in granules and excretion into the sarcoplasm in secretory cardiac myocytes. The results were obtained using the methods of transmission electron microscopy and immunocytochemistry. The study makes an important contribution to histology and improves the understanding of the function of the heart as an endocrine organ. This chapter is intended for students of medical and biological universities and for experts.

Keywords: atrial secretory (or endocrine) cardiac myocytes, atrial natriuretic peptide (ANP)

1. Introduction

The concept of the heart's endocrine function was described by A.A. Galoyan with coauthors in 1967–1971 years [1]. Later, the ultrastructure of atrial secretory cardiac myocytes was described, and the atrial natriuretic peptide (ANP) contained in the granules of the myocytes was identified [2]. Currently, about 100 bioactive substances synthesized by atrial cells were determined [1].

Endocrine (or secretory) cardiac myocytes have not only well-developed contractile structures such as in ventricular myocytes but also electron-dense granules in the sarcoplasm.

Examining the heart morphology under the influence of various factors, the researchers focus on the myocardium left ventricle. It is widely accepted that the key role in the development of numerous cardiac pathologies is given to the ventricular contractile myocytes [3]. The role of atria is given less attention than ventricles, but their dilation determines the development of chronic heart failure and arrhythmia [4].

Studies of secretory atrial myocytes have scientific and practical importance, as these cells are the main source of production and storage of ANP [1, 2]. Peptide has a hypotensive effect due to the diuretic, natriuretic actions, and the suppression of the renin-angiotensin-aldosterone system [5, 6]. ANP is released from granules after tension of the heart wall [7], under the influence of hypoxia and neurohumoral factors [8]. The peptide inhibits the growth of smooth myocytes, endothelial cells, and the activity of fibroblasts [9]. The peptide is involved in the differentiation of cardiac myocytes [10], reduces hypertrophy [11], has anti-inflammatory effect, and so on [12]. The definition of ANP concentration in the blood has diagnostic and prognostic value [13]. Synthetic peptide is used in cardiology [14]; therefore, the study of interaction with drugs is an actual problem.

Despite more than 30 years of research, the question of "hormonal paradox" awaits a solution. It demonstrates the absence of a hypotensive effect of ANP in hypertension of different etiologies [6]. The role of ANP in the pathogenesis of cardiovascular diseases is ambiguous [8].The contradictory data of the study could partially be associated with the use of different methods for the determination of ANP. The few research works of atrial cardiac myocytes are devoted to their morphology only [15] or to the quantitative assessment of the hormone content without analyzing the ultrastructure of cells [16].

Morphometry of immunocytochemical-labeled granules in atrial myocytes with using the transmission electron microscopic analysis of the myocardium allows to investigate the localization of ANP along with changes in the ultrastructure of cells. It also evaluates the intensity of granulopoiesis in norm and in experimental pathology.

Special drugs for the correction of metabolic disorders caused by hypoxia are used in the intensive care unit. One such of drugs, Mexidol (ethylmethylhydroxypyridine succinate), is used in Russia. The neuro- and cardioprotective actions of the drug in the post-reperfusion period (PRP) were studied [17]. Mechanisms of the influence of the drug on the accumulation and excretion of ANP in secretory granules of myocytes have not been investigated.

Thus, the study does not give a complete picture of the morphological and functional features of secretory cardiac myocytes in different conditions. Therefore, we have applied histological techniques to study the right atrium and experimental models for the investigation of this type of cardiac myocytes in this work. This approach gives the possibility to quantitatively assess the dynamics of cardiomyocytes, granulopoiesis, and contents of ANP in norm, experimental pathology, and after the injection of Mexidol. The study makes a significant contribution to the discovery of the mechanism of the endocrine function of the heart.

2. Materials and methods

Experiments were carried out on white outbred Wistar male rats (n = 180) weighing 200–220 g. We used various models, such as clinical death [18], renovascular hypertension [19], and the Langendorff-perfused rat heart [17].

Clinical death (10 min) was induced using the method described by Korpachev [14]. Rats were anesthetized with Nembutal (25 mg/kg) and intubated; then, the cardiovascular fascicle was clamped with a special L-shaped hook without opening the chest. The heart completely stopped at 2–4 min after clamping. Before the start of resuscitation, 0.1% epinephrine solution (0.1 mL) was administered endotracheally. Resuscitation was performed by external cardiac massage and artificial respiration. We investigated experimental rats after 60 min and 60 days of post-reperfusion period. In experiments with Mexidol, the drug was administered after intraperitoneal resuscitation for the first hour, every 20 min [17].

Renovascular hypertension was induced using the method described by Kogan [19]. Rats were anesthetized, and then the artery of the left kidney was ligated. After 30 days of the procedure, hypertension was developed.

The model of a Langendorf isolated heart was used with the saline Krebs-Henseleit solution of the following composition (mmol/L): NaCl—130; KCl—4; $NaH_2PO_4 \cdot 2H_2O$—1.1; $NaHCO_3$—24; $MgCl_2$—1; $CaCl_2 \cdot 2H_2O$—1.8; glucose—5.6. The solution was saturated with Carbogen (95% O_2, 5% CO_2), with the pH of 7.3–7.4 at a temperature of 37°C. Two refrigerators were used to switch to perfusion with Mexidol: one with Krebs-Henseleit control solution and the other one with Mexidol in the dose of 25 mg/kg added [17].

For electron microscopy analysis, samples were taken from the right atrium of intact and experimental animals. The heart tissue was fixed in 2.5% glutaraldehyde in phosphate buffer (pH 7.4), post-fixed in 1% osmic acid, dehydrated in ascending alcohols, and embedded in epon and araldite mixture according to the standard protocol. Cellular localizations of atrial natriuretic peptide was detected on ultrathin sections of the right atria using primary polyclonal anti-ANP (rabbit anti-atrial natriuretic factor (1–28) (rat), Peninsula Laboratories, LLC, Bachem) and secondary antibodies (Protein-A/Gold (15 nm), EM Grade, Electron Microscopy Sciences).

Ultrathin sections were analyzed under a Morgagni 268D (FEI) transmission electronic microscope. Morphometric analysis of the areas occupied by mitochondria, sarcoplasmic reticulum, myofibrils, and sarcoplasm of cardiomyocytes was performed using AnalySIS software.

In secretory myocytes of the right atrium, the number of immunodeficiency granules with ANP using the classification was evaluated [19]: counted granules of A-type ("reserving peptide") with a well-defined membrane and osmiophilic content, and B-type ("releasing peptide") without a membrane and with a less electron-dense content (**Figure 1**).

The results were evaluated using Mann–Whitney test and Spearman correlation coefficient. The differences were significant at $p \leq 0.05$. Data in tables are presented as mean (M) ± standard deviation (SD).

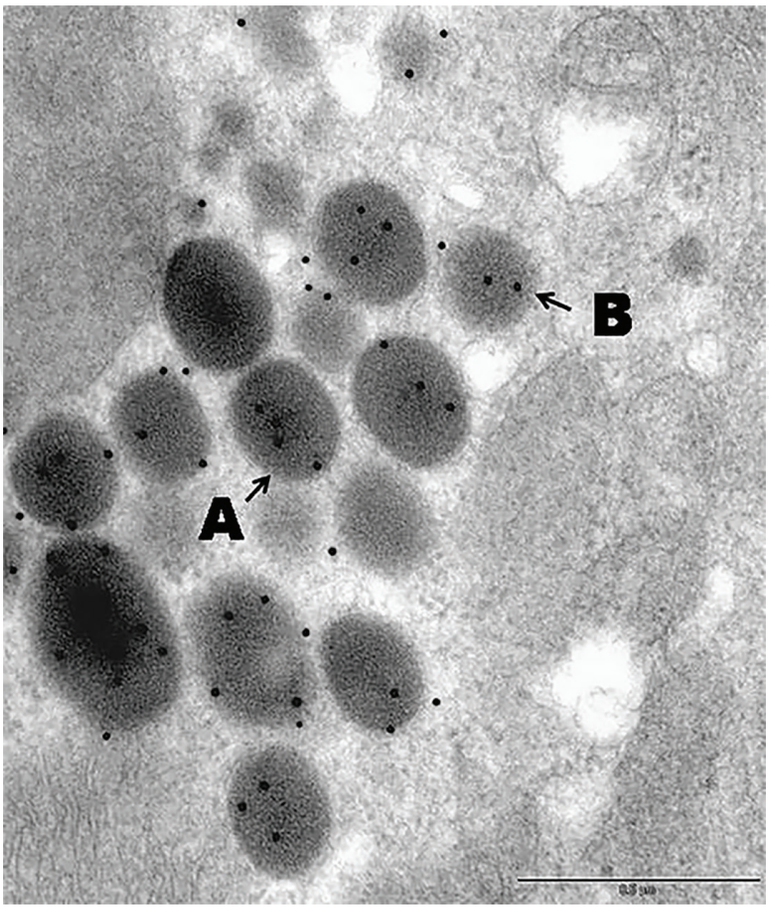

Figure 1. Immunocytochemical detection of ANP in the granules of right atrial cardiac muscle cells in rat. A and B, granules of A and B types, respectively. ×71,000.

3. Results

3.1. Secretory atrial myocytes in normal rats

Atrial secretory or endocrine cardiac myocytes differ from the contractile cardiac muscle cells by the presence of secretory granules in sarcoplasm (**Figure 2**).

The specific localization of this type of myocytes in the right atrium is not found. Cells with granules are mixed with the cells without those. Endocrine cardiomyocytes may differ from each other by the number of granules. We assume that all atrial myocytes have the potential ability for secretory function.

The most part of granules are localized in the perinuclear space near the Golgi complex and contain immunoreactive material of atrial natriuretic peptide (ANP). The quantitative distribution of A- and B-type granules with ANP was 63 and 37% in secretory cardiac myocytes (**Table 1**).

Quantitative values of areas occupied by myofibrils, mitochondria, sarcoplasmic reticulum, and sarcoplasm of atrial cardiac myocytes of intact animals are presented in **Table 2**.

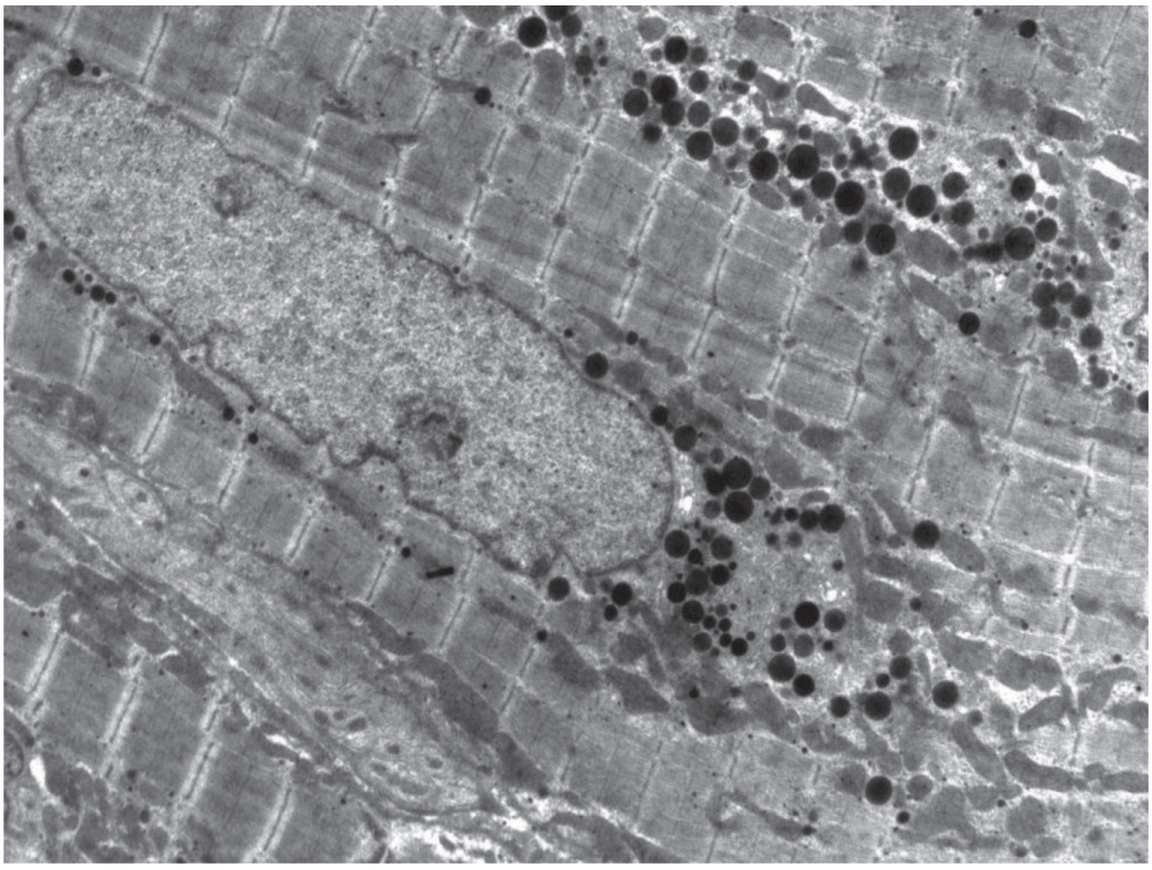

Figure 2. Cardiomyocytes of the right atrium of the intact rat. ×4400.

In the previous study, we revealed the individual peculiarities in the right atrium and left ventricle of intact animals [20]. The areas occupied by various organelles in atrial secretory cardiomyocytes include mitochondria (23%), myofibrils (46%), sarcoplasmic reticulum (1%), and sarcoplasm (30%). Similarly, in ventricular cardiomyocytes, the area was distributed as follows: mitochondria (33%), myofibrils (56%), sarcoplasmic reticulum (0.5%), and the sarcoplasm (10.5%).

Experimental conditions	A-granules	B-granules	Total number of granules
Intact rats	65.75±19.49	38.90±19.63	104.65±33.41
60-min PRP	85.64±20.78*	56.48±17.00*	142.12±36.53*
Langendorff-perfused rat heart	88.54±19.22*	42.17±14.53	130.71±29.79*
Langendorff-perfused rat heart after 10-min ischemia	99.97±33.40*	65.93±23.36*	165.90±55.08*
60 days of PRP	105.17±28.27*	54.71±19.66*	159.88±44.55*
Renovascular hypertension	71.45±24.84	23.75±10.58*	95.20±32.82

Note: $p < 0.05$ in comparison with *the intact animals (Mann-Whitney test).

Table 1. Content of A and B granules containing ANP—granules in atrial cardiac myocytes in experiment (number of granules in visual field, M ± SD).

Ultrastructural element	Intact rats	60-min PRP	60 days of PRP	Renovascular hypertension
Mitochondria	6.82±2.14	7.58±2.13	7.34±1.69	7.,51±1.62
Myofibrils	15.50±2.79	15.70±4.51	17.65±2.41*	14.86±2.55
Sarcoplasmic reticulum	0.31±0.22	0.50±0.29*	0.55±0.36*	0.27±0.30
Sarcoplasm	9.77±2.90	8.91±3.38	6.87±1.56*	9.13±2.61

Note: $p < 0.5$ in comparison with *the intact animals (Mann–Whitney test).

Table 2. Areas occupied by ultrastructural elements of cardiac myocytes in the right atrium in experiment (μm^2, M ± SD).

These data attest to individual functional differentiation of the myocytes: in comparison with ventricular cardiomyocytes, the atrial endocrine ones were characterized by less developed contractile and the energy-providing apparatuses (according to the area occupied by myofibrils and mitochondria) as well as an increased content of the synthetic apparatus (reflected by sarcoplasmic reticulum) and greater sarcoplasm areas [20].

3.2. Secretory atrial myocytes after 60-min post-reperfusion period

After 60 min of post-reperfusion period, when blood stream had been restored, we revealed the heterogeneity of myocytes. Most of the cells were without any changes, some were with degenerative disorders. The areas occupied by mitochondria, myofibrils, and myofibril-free sarcoplasm in atrial secretory cardiomyocytes did not differ significantly from the corresponding values of intact rats. By contrast, sarcoplasmic reticulum area increased by 61% (**Table 2**). Some areas of myocardium with interstitial edema were identified (**Figure 3**). The evident damages were not observed in most of the cells. It should be noted from the previous study that the ventricular cardiomyocytes changed more severely than the atrial ones [20].

After 60 min of post-reperfusion period, submicroscopic examination of the myocytes of the right atrium revealed a pronounced increase in the content of ANP containing A- and B-granules by 30 and 45%, respectively, whereas the total content of secretory granules increased by 36%. These data attest to intensive accumulation and secretion of ANP (**Table 1**). There was a loose positive correlation between the total number of ANP-storing granules and SR area ($r = 0.37$). The study reports upregulation of granule formation in atrial cardiomyocytes via receptors associated with G proteins (Go and Gq), which trigger $Ca2^+$-activated (SK4) potassium channels residing in the sarcoplasmic reticulum [21]. The calcium ions activate protease corin, which converts ANP precursor (pro-ANP) to mature and active ANP [22]. However, no correlation between the total number of granules and the areas occupied by mitochondria or myofibrils was revealed.

According to our previous study [23], the applied heart rate variability (HRV) analysis and arterial pressure (AP) measurement enabled to conclude the following: within the first minutes of post-reperfusion period, a short-term AP increase and the activation of

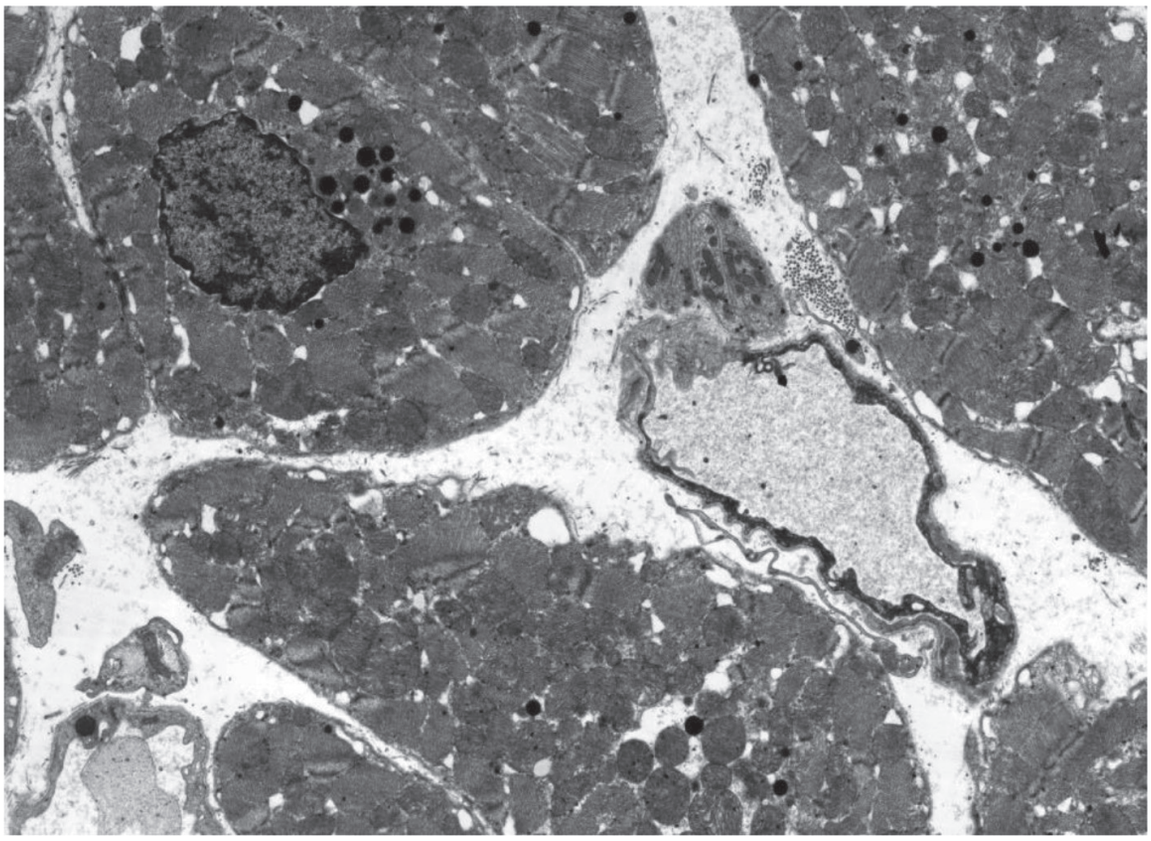

Figure 3. Cardiac myocytes of the right atrium after 60 min of post-reperfusion period: The area of myocardium with interstitial edema, ×8900.

sympathoadrenal, pituitary-adrenal, and rennin-angiotensive systems had no effect on ANP synthesis and secretion in the right atrial myocytes. On the 60th min of post-reperfusion period, a high intensity of ANP synthesis, and accumulation and secretion in atrial myocytes were associated with a stimulating effect of hypoxic and ischemic factors during this period [24].

3.3. Secretory atrial myocytes in Langendorff-perfused rat heart

According to the study and our own research, the heart starts the autonomous functioning after 60 min of post-reperfusion period [24, 25]. The influence of external neurohumoral factors on morpho-functional characteristics of secretory cardiomyocytes was studied in Langendorff-perfused rat heart.

According to Arjamaa and Nikinmaa [26], the myocardium of isolated perfused heart is experiencing a small hypoxia due to the lower oxygen content in the solution compared to the blood. The cardiac myocytes of the isolated heart mainly retain their structure and have adaptive changes under the influence of hypoxia: expanded sarcoplasmic reticulum. We found the small intercellular edema (**Figure 4**).

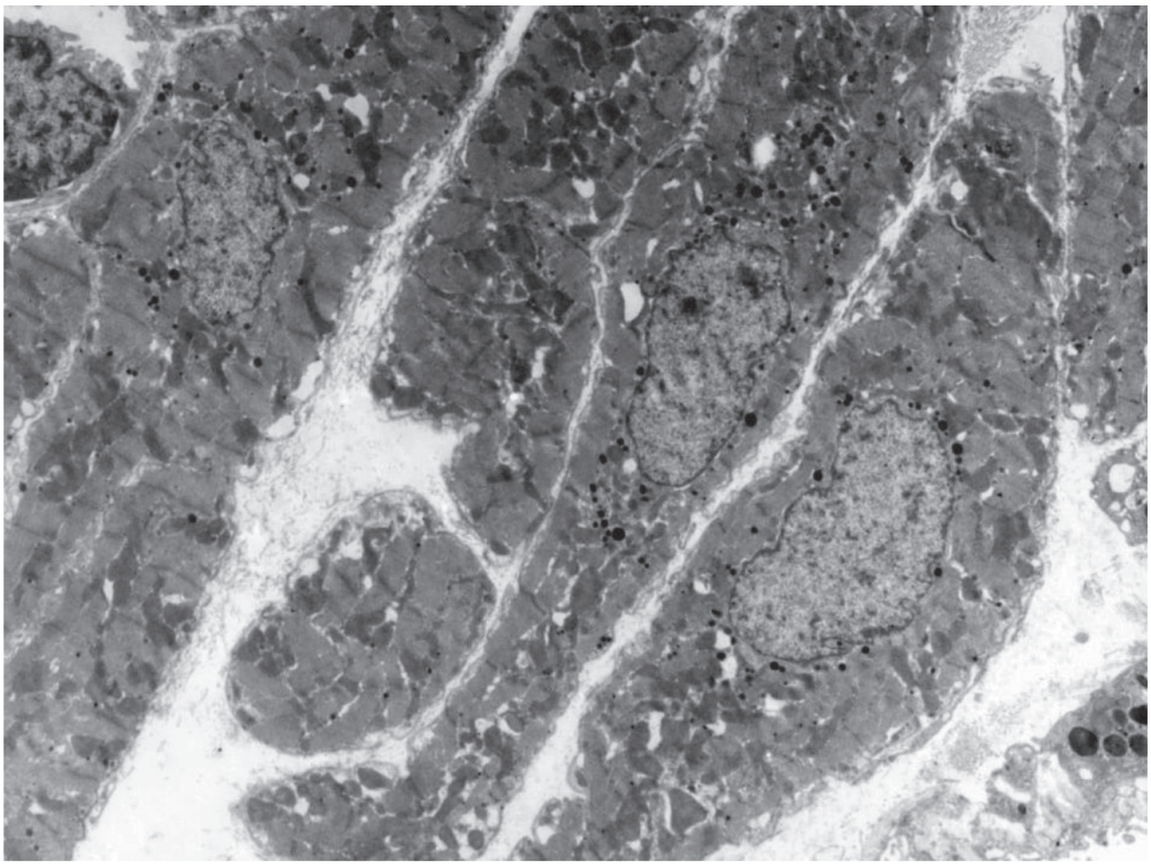

Figure 4. The atrium of isolated rat heart with the intercellular edema: Cardiac myocytes with expanded sarcoplasmic reticulum. ×2800.

The accumulation of ANP in atrial myocytes of isolated heart enhanced: number of A-type of granules increases at 35% and the total number of granules on 25% in comparison with indicators of intact rats (**Figure 5**). According to the authors [26], hypoxia provokes an increase in the transcription of the peptide due to the activation of HIF—"hypoxia inducible factor."

We investigated the contribution of ischemia and reperfusion in the change of ultrastructure and granulopoiesis in atrial myocytes using an experiment with simulations of 10-min ischemia and reperfusion in an isolated heart. There are intracellular destructive changes in comparison with the control group of isolated hearts. Almost all myocytes have condensed or vacuolated form of mitochondria: the dilatation of the sarcoplasmic reticulum. Lysis of myofibrils of cardiomyocytes was observed in some cells (**Figure 6**).

A 10-min period of ischemia and reperfusion stimulates the accumulation and excretion of ANP in endocrine myocytes isolated heart. An increase in the granules A-type by 13%, B-type by 56%, and the total number of granules by 27% compared with the control group of isolated heart is shown (**Table 1**, **Figure 5**). It should be noted that quantitative data and changes in the ultrastructure of cardiac myocytes are similar to the characteristics of rats after 60 min of post-reperfusion period.

http://dx.doi.org/10.5772/intechopen.76311

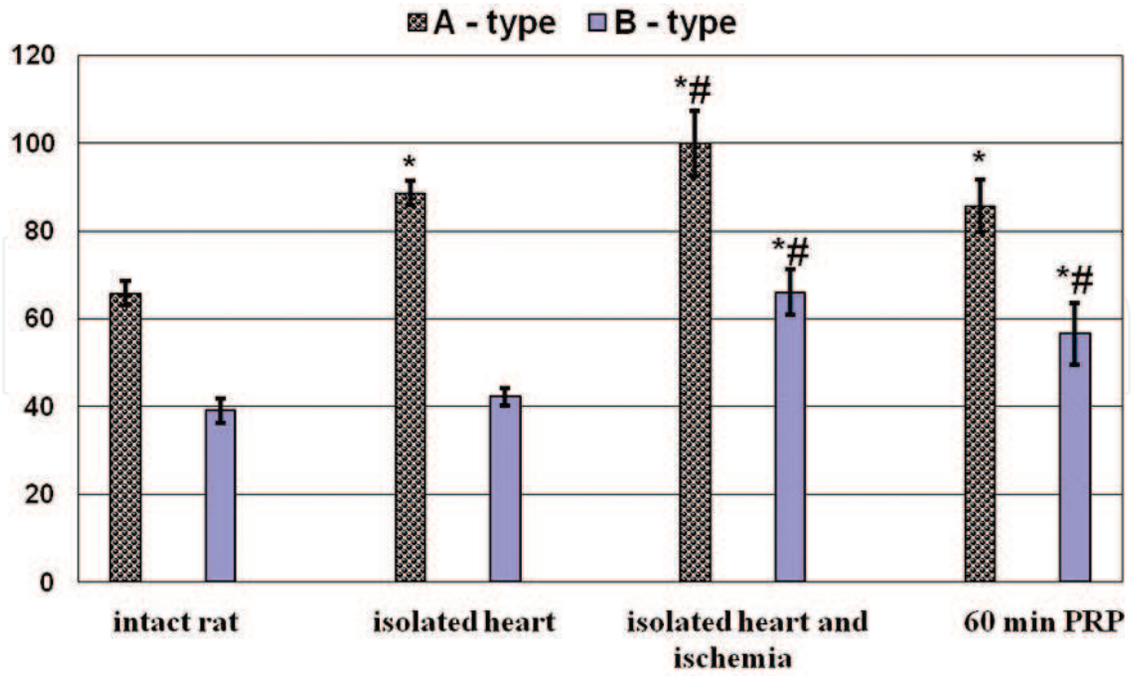

Figure 5. Quantitative distribution of the granules with ANP in the intact rat hearts, isolated perfused hearts, isolated perfused hearts after 10-min ischemia, and rat after 60-min PRP (post-reperfusion period). Asterisk indicates significant differences from intact animals; hash indicates significant differences from isolated heart; $p < 0.05$ (according to Mann-Whitney test).

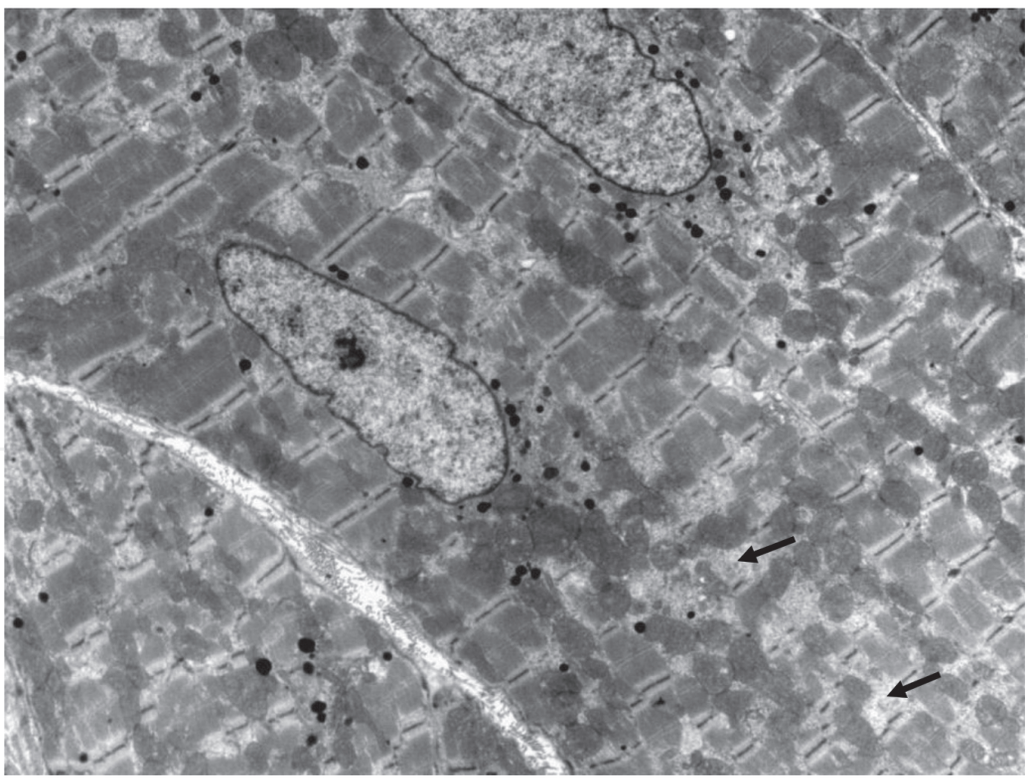

Figure 6. The atrium of isolated rat heart after 10-min ischemia: Cardiac myocytes with lysis of myofibrils (arrows). ×3500.

Thus, granulopoiesis and changing the accumulation and excretion of ANP in secretory cardiac myocytes occur regardless of external neurohumoral factors after 60 min of post-reperfusion period. The processes are influenced by ischemia and reperfusion. This study confirms the existence of a functional isolation of the heart after 60 min of post-reperfusion period.

3.4. Effects of Mexidol on secretory atrial myocytes

The effect of Mexidol used in the correction of ischemic myocardial damage on the secretory myocytes was investigated in rats after 60 min of post-reperfusion period and in Langendorff-perfused hearts (control group and after 10 min of ischemia period and reperfusion).

The nuclei of atrial cardiomyocytes contain euchromatin and nucleoli, the expanded Golgi complex, and an increased number of granules of glycogen in the sarcoplasm in the group after 60 min of post-reperfusion period with the injection of Mexidol (**Figure 7**).

The area of the sarcoplasmic reticulum does not differ from the values of intact animals. The increase of the mitochondrial area of cardiac myocytes by 16% is believed to indicate the increase of functional activity (**Table 2**). Researchers have observed this state of mitochondria in suspension during aeration, in the addition of adenosine triphosphate [27], and in aerobic respiration in the cells [28].

The positive effect of Mexidol on the synthetic activity of secretory cardiac myocytes is manifested by the increase of the content of ANP in comparison with the control group: granules

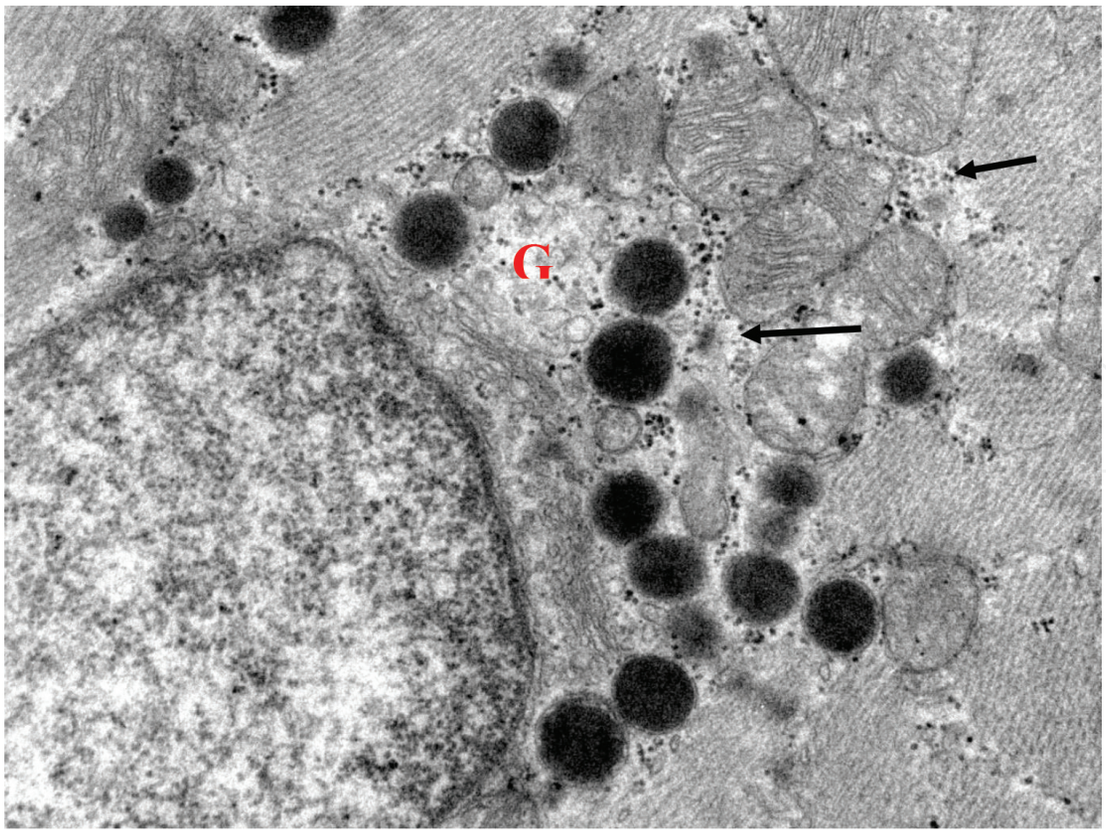

Figure 7. Cardiac myocytes of the right atrium after 60 min of post-reperfusion period with the injection of Mexidol: the expanded Golgi complex (G) and an increased number of granules of glycogen (arrows). ×18,000.

A-type was increased by 38%, B-type by 42%, and the total number of granules by 37% (**Table 1**). A direct correlation between the total number of granules and the area of mitochondria (r = 0.44) allows us to consider the increased synthesis of adenosine triphosphate to promote granulopoiesis in myocytes.

Previously, it was shown that the heart after 60 min of post-reperfusion period is in a state of functional isolation. The effect of Mexidol on the endocrine cardiac myocytes in the conditions of complete isolation of the heart and under the influence of factors of ischemia/reperfusion was investigated in isolated hearts and after 10-min period ischemia and subsequent reperfusion.

The morphological picture was similar to the group after 60 min of post-reperfusion period with the injection of Mexidol. The number of granules containing the ANP was more than in the control myocardium of isolated hearts: granules of A-type by 33%, B-type 53%, and the total number by 39%. A dramatic increase in the A- and B-type granules indicated a beneficial effect of Mexidol on ANP formation and release in the isolated rat heart. Apparently, it was related to the cytoprotective effect of the drug, which manifested itself on the myocardium ultrastructure as a high content of glycogen cytogranules in the sarcoplasm and sarcoplasmic reticulum without dilatation cisterns. The identified increase in the average value of the mitochondria area with the preservation of membrane structures and matrix indicated the energized state of the organelles that arise, according to the authors [27, 29], at media aeration, with oxidation substrates or ATP added. The membrane-protecting effect, improvement, and preservation of high-energy compounds synthesis with Mexidol administration had a positive impact on energy input processes of ANP formation and release (**Figure 8**).

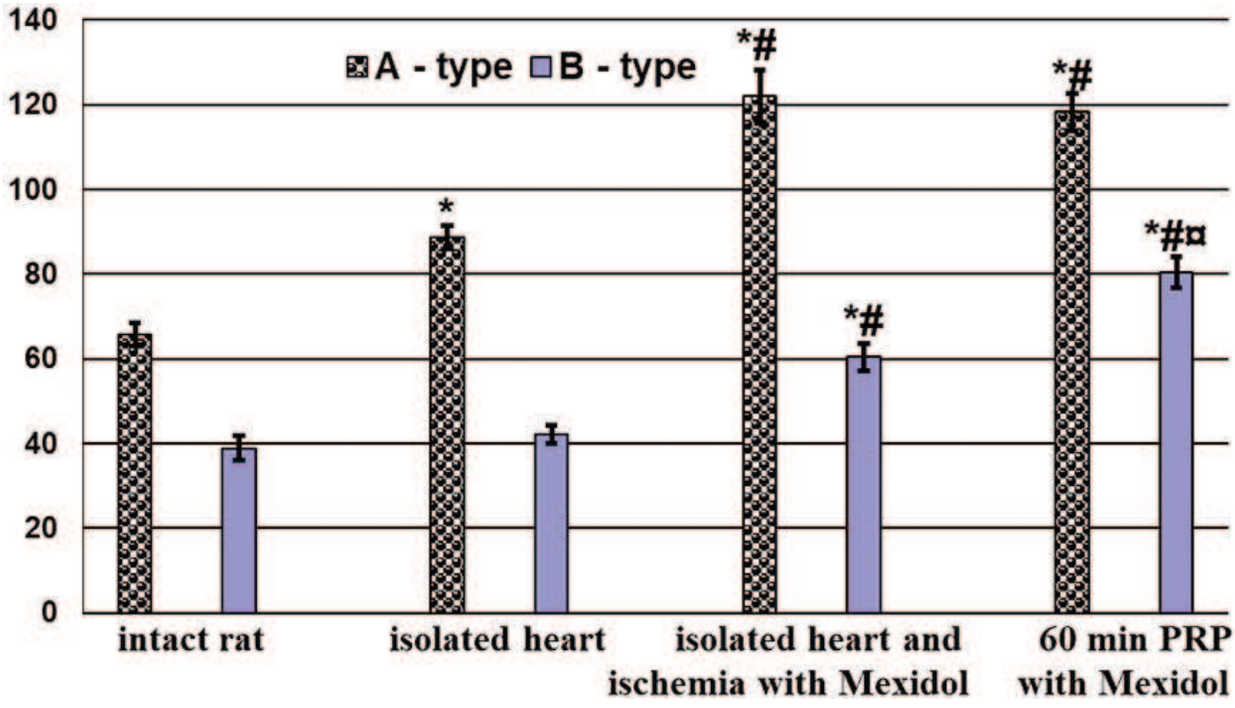

Figure 8. Quantitative distribution of the granules with ANP in the intact rat hearts, isolated perfused hearts, isolated perfused hearts after10-min ischemia with Mexidol administration, and rat after 60-min PRP with Mexidol administration (post-reperfusion period). Asterisk indicates significant differences from intact animals; hash indicates significant differences from isolated heart; $p < 0.05$ (according to Mann-Whitney test).

According to the research data [24, 25], ANP introduced into the ANP perfusion solution has a cardioprotective effect on the cardiomyocytes of an isolated perfused heart. The ANP effect on the electrophysiological heart function is also known [30]. It is put into effect in two ways in the isolated heart: (1) directly, through the autonomic nervous system (according to the authors, ANP depresses the sympathetic and activates parasympathetic component of the autonomic nervous system; (2) through calcium canals: ANP weakens the calcium flow into the cell, inhibiting ICaL canals. Herewith, the cyclic guanosine monophosphate (cGMP) activated by the peptide facilitates the performance of calcium ATPases which carries intracellular calcium into the sarcoplasmic reticulum and reduces the risk of calcium overload. Besides, ANP is shown [31] to prevent the so-called electrical remodeling leading to atrial fibrillation.

Thus, we found a positive effect of Mexidol on the ultrastructure of secretory cardiac myocytes, granulopoiesis, and the secretion of ANP after exposure to ischemia and reperfusion in body rats and in the isolated hearts. The revealed effect of Mexidol perhaps discovers another mechanism of its cardioprotective action and can be used in pharmacology and medicine.

3.5. Secretory atrial myocytes after 60 days of post-reperfusion period

After 60 days of post-reperfusion period, the morphological diversity of the myocytes of the atrium was revealed (**Figure 9**). We found the cardiac myocytes without visible changes and cells with some degenerative changes in the nuclei or appeared apoptotic bodies (**Figure 9**).

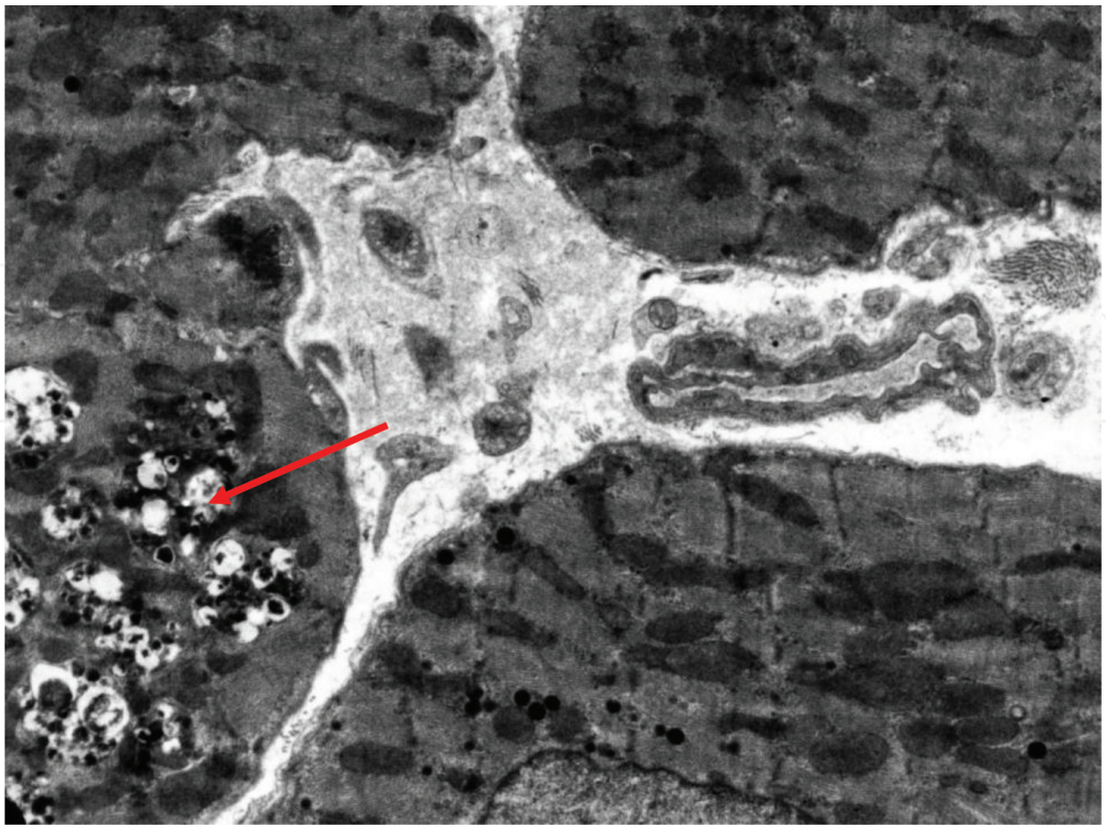

Figure 9. The right atrium after 60 days of post-reperfusion period: apoptotic bodies in the myocyte (arrow). ×5600.

We identified condensed forms of mitochondria or organelles with the enlightenment of the matrix and disorientation of cristae (**Figure 10**).

The areas of myofibrils and the sarcoplasmic reticulum increased in right atrial cardiomyocytes by 14 and 77%, respectively, while the area occupied by mitochondria did not differ from the intact value. In parallel, the area of myofibril-free sarcoplasm decreased by 30% in comparison with the initial value (**Table 2**). There were a large number of granules with immunoreactive label to ANP. At this, the content of A- and B-granules increased by 60 and 41%, respectively, whereas the total content of granules increased by 53% in comparison with the intact values (**Table 1**). The A:B granule content ratio was 66:34%. A moderate positive correlation was revealed between the total content of secretory granules with ANP and the sarcoplasmic reticulum area ($r = 0.36$). By contrast, there was no correlation between the total content of granules and the area occupied by mitochondria or myofibrils. An enhanced endocrine activity of the secretory cardiomyocytes was observed against the background rise of arterial pressure by 23% [23], which in the view of some researches upregulates not only secretion but also the synthesis of ANP [15, 21].

In previous studies [20], we revealed a hypertrophy of ventricular cardiomyocytes and an increase in the area occupied by the connective tissue in the interstitial space. According to the researchers, these structural changes in the myocardium eventually provoke myocardial remodeling [4]. Taking into account the data of the study, ANPs secrete in response to the increase in the synthetic activity of fibroblasts and/or hypertrophy of cardiac myocytes [6, 11].

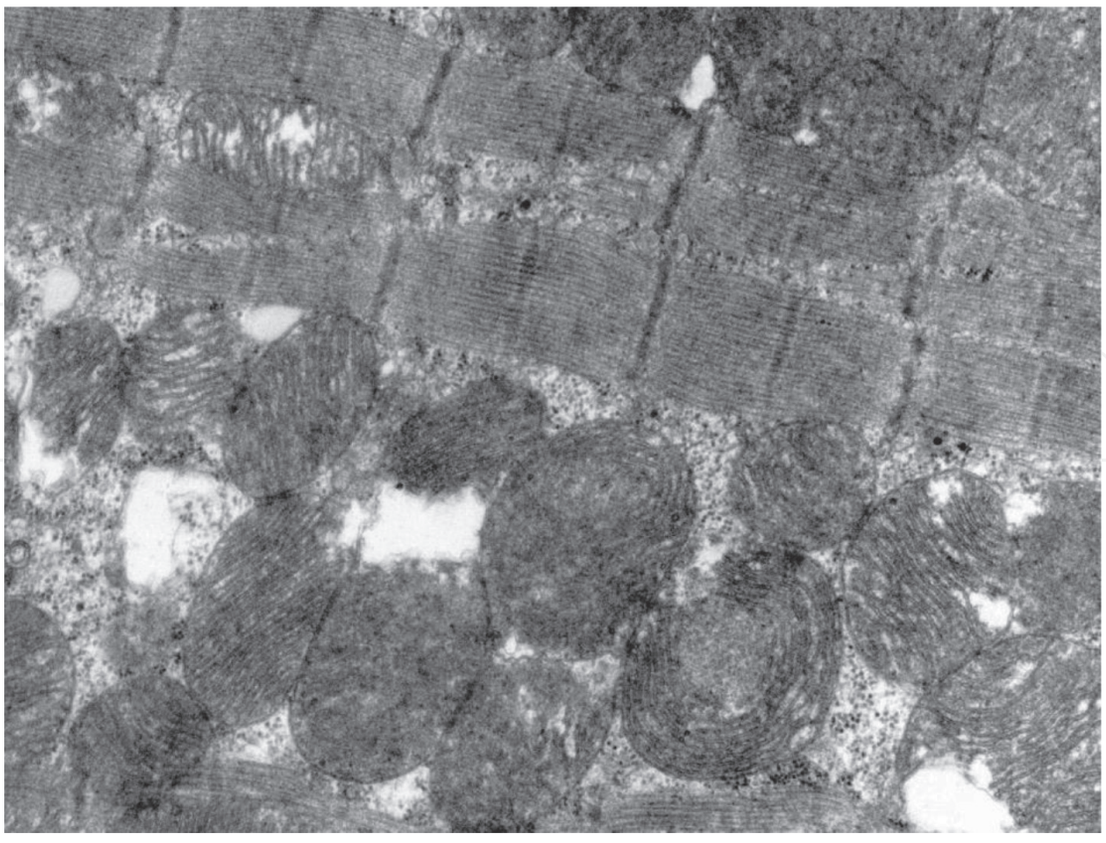

Figure 10. Enlightenments of the matrix and disorientation of cristae of mitochondria in the myocyte of the right atrium after 60 days of post-reperfusion period. ×22,000.

Thus, after 60 days of post-reperfusion period, we observed some pronounced changes in the ultrastructure of the secretory atrial cardiac myocytes and an increase in the accumulation and excretion of ANP in their granules. These processes are accompanied by a high blood pressure, an increase in the area occupied by the connective tissue in the myocardium, and the hypertrophy of the ventricular myocytes.

3.6. Secretory atrial myocytes in different types of the arterial hypertension

Some scientists believe the ambivalence of the role of secretory cardiac myocytes and ANP in the development of cardiovascular diseases accompanied by an increased blood pressure to be present [11]. A scientific interest to the comparison of the content of peptide in the endocrine myocytes in hypertension being formed on different experimental models appeared. We investigated the structure and granulopoiesis in myocytes with renovascular hypertension, developed in 30 days after ligation of the left renal artery and compared the data of hypertensive animals after 60 days of post-reperfusion period.

We have identified both similarities and differences in the experimental groups. Heterogeneity of cardiac myocytes was found in both groups, but after 60 days of post-reperfusion period, we noted morphological signs of apoptosis (**Figure 9**). In renovascular hypertension, we found mitochondria with vacuoles and myofibrils lysis in myocytes (**Figure 11**).

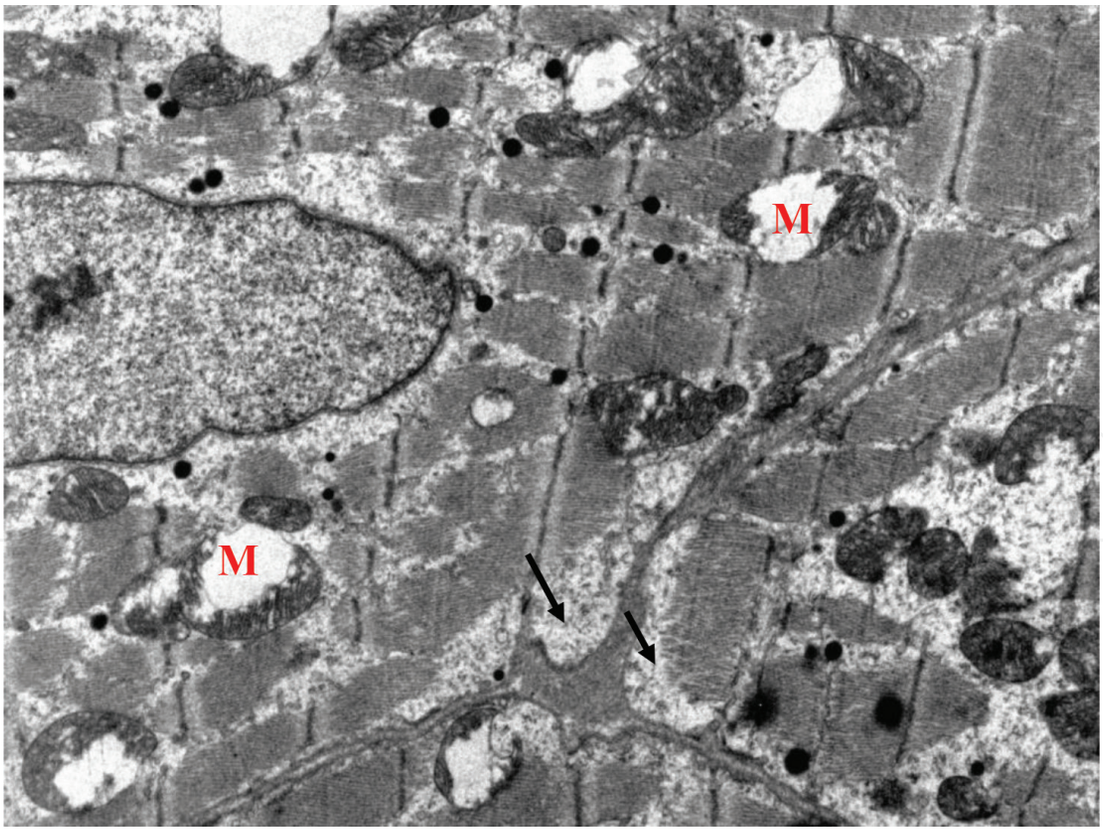

Figure 11. Cardiac myocytes of the right atrium in renovascular hypertension: mitochondria with vacuoles (M) and myofibrils lysis (arrows). ×7100.

Quantitative characteristics of the ultrastructure changes were different (**Table 2**). The table shows the area occupied by mitochondria, myofibrils, the sarcoplasmic reticulum, and the sarcoplasm to be not significantly different from the values of the intact animals.

There is hyperplasia of mitochondria in cardiomyocytes after 60 days of post-reperfusion period. We revealed mitochondrion destruction in renovascular hypertension (**Figure 11**). Vacuoles were noticed in cardiomyocytes of both groups. According to the study, these ultrastructural changes indicate a destabilization of the energy metabolism in the myocytes of rats in renovascular hypertension [3]. Morphological picture indicates compensatory processes in cells after 60 days of post-reperfusion period [4]. The area occupied by the sarcoplasmic reticulum of cardiomyocytes is more increased after 60 days of post-reperfusion period than in the group with renovascular hypertension (**Table 2**).

There is a difference of granulopoiesis in atrial myocytes of experimental groups (**Table 1**, **Figure 12**). In renovascular hypertension, the number of A-granules and the total content of granules were the same as the intact values. B-granules decreased by 39%.

Thus, the content of ANP is not increased in myocytes of rats in renovascular hypertension. After 60 days of post-reperfusion period, we revealed the intensive synthesis and secretion of ANP and have shown a positive correlation between the area occupied by the sarcoplasmic reticulum and the total number of granules (r = 0.36).

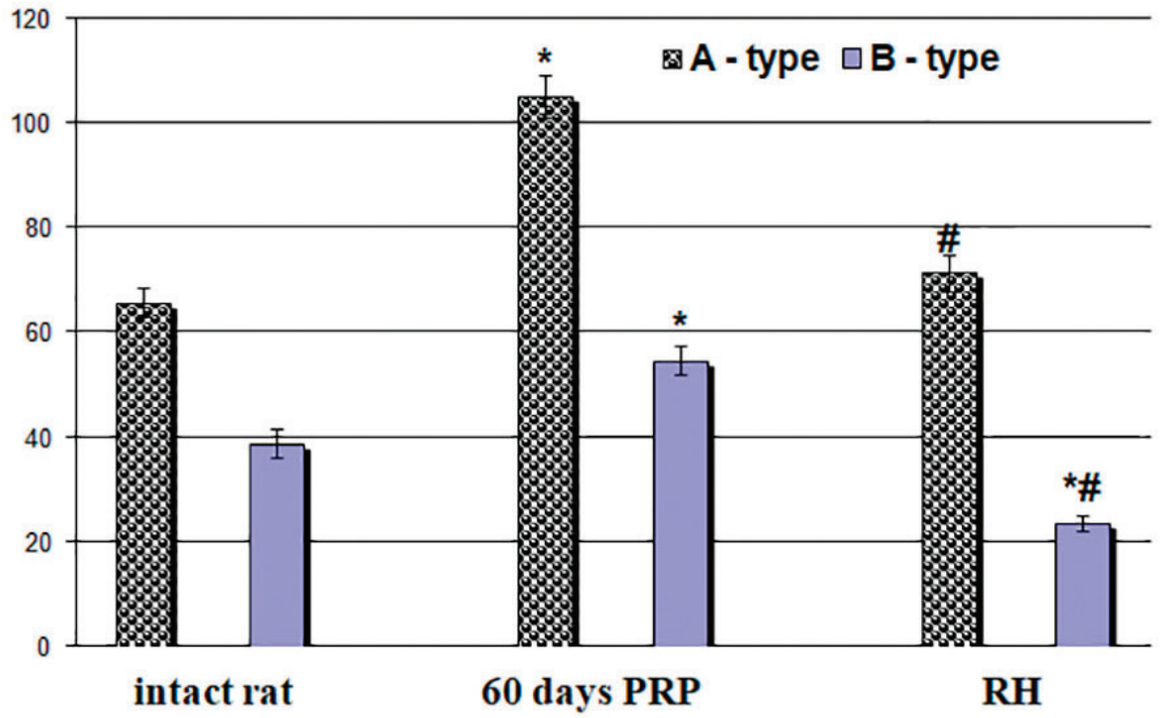

Figure 12. Quantitative distribution of the granules with ANP in the intact rat hearts, rat after 60 days of post-reperfusion period (PRP) and rat with renovascular hypertension (RH). Asterisk indicates significant differences from the intact animals; hash indicates significant differences from the rats after 60 days of PRP; $p < 0.05$ (according to Mann-Whitney test).

The hypertrophy of cardiomyocytes was detected in both experimental groups [32], but the area occupied by the connective tissue does not increase in renovascular hypertension in contrast to the 60 days of the post-reperfusion period (**Figures 6** and **7**).

Thus, the comparison of models of renovascular hypertension and 60 days of post-reperfusion period shows various ultrastructural changes of secretory myocytes and the content of ANP in their granules. The granulopoiesis in atrial myocytes depends on the combination of factors, such as a high blood pressure, the hypertrophy of cardiac myocytes of the left ventricle, and the area occupied by the connective tissue in the myocardium.

4. Conclusion

The study identified the morphological characteristics of secretory cardiac myocytes of the right atrium in male Wistar rats in norm and in experimental cardiovascular pathology.

The certain regularity of localization of this type of myocytes in the right atrium is not detected, so we assume that all atrial myocytes have the potential ability for secretory function.

Experiments on models of clinical death, renovascular hypertension, and in Langendorff-perfused rat heart allowed detecting features of granulopoiesis in atrial myocytes under the influence of pathological factors of ischemia/reperfusion and in high blood pressure.

We found a direct correlation between increasing the area occupied by the sarcoplasmic reticulum or mitochondria and the increased number of granules with ANP. The increase in blood pressure is not always the main stimulus for the formation and secretion of the peptide in myocytes of the right atrium. The granulopoiesis can be activated by a certain combination of factors influencing the ultrastructure of the secretory myocytes.

The process of formation and secretion of ANP in the granules of endocrine cardiac myocytes occurs without the involvement of extracardiac factors after 60-min post-reperfusion period.

Factors of ischemia/reperfusion stimulate the accumulation and secretion of ANP in the granules of the myocytes in isolated rat hearts.

In the research, we revealed a significant positive effect of Mexidol on the ultrastructure, granulopoiesis, and secretion of ANP from granules into the sarcoplasm of secretory cardiac myocytes of the right atrium in Langendorff-perfused hearts and in rats at the early post-reperfusion period. The cardioprotective property of Mexidol can be realized indirectly by activating the synthesis and secretion of ANP in the myocytes of the right atrium.

The changes of the ultrastructure of secretory myocytes of the right atrium and the intensity of the accumulation and secretion of ANP vary considerably in hypertension of different genesis. After 60 days of post-reperfusion period, increased granulopoiesis and secretion of the peptide are associated with an increase in the area of sarcoplasmic reticulum and integrity of mitochondria. These processes are accompanied by the increase of the total area occupied by the connective tissue in the intercellular space in the myocardium.

However, the functional activity of myocytes of the right atrium does not increase despite high blood pressure and hypertrophy of ventricle cardiac myocytes in renovascular hypertension. Based on these data, we put forward the concept of the dominant role of structural reorganization of the myocardium resulting in a change of the ultrastructure of secretory myocytes and the secretion of atrial natriuretic peptide, localized in the granules, in hypertension.

The study makes a significant contribution to the understanding of the peculiarities of ultrastructural organization of endocrine cardiac myocytes of the right atrium containing atrial natriuretic peptide. It is necessary to underline the role of their granulopoiesis in the regulation of complex mechanisms of the heart in normal and pathological conditions.

Author details

Marina L. Bugrova

Address all correspondence to: marysmir@mail.ru

Federal State Budgetary Educational Institution of Higher Education "Privolzhsky Research Medical University" of the Ministry of Health of the Russian Federation, Nizhniy Novgorod, Russian Federation

References

[1] Galoyan A. Concepts of neuroendocrine cardiology and neuroendocrine immunology, chemistry and biology of signal molecules. Neurochemical Research. 2010;**35**(12): 2001-2017

[2] Flynn T, de Bold M, de Bold A. The amino acid sequence of an 594 atrial peptide with potent diuretic and natriuretic properties. Biochemical and Biophysical Research Communications. 1983;**117**:859-865

[3] Gavrish A, Paucov V. The Ischemic Cardiomyopathy. Moscow: GEOTAR-Media; 2015. p. 536

[4] Sapin M, Nikolenko V, Milyukov V, Dolgov E, Rakhimov A. Morphofunctional organization of the myocytes of the atria and ventricles of the heart. Journal of Anatomy and Histopathology. 2012;**1**:11-17

[5] Hermanov V. Modern drug treatment of acute heart failure decompensation. Researches and guidance in recent years. Clinical Pharmacology and Therapy. 2015;**3**:15-33

[6] Kuhn M. Cardiac actions of atrial natriuretic peptide: new visions of an old friend. Circulation Research. 2015;**116**(8):1278-1280

[7] de Bold A. Thirty years of research on atrial natriuretic factor: Historical background and emerging concepts. Canadian Journal of Physiology and Pharmacology. 2011;**89**:527-531

[8] Ivanova S, Nesterova E. Family of natriuretic peptides: Possibilities of application in polyclinic practice. Medical Council. 2014;**2**:77-81

[9] Okamoto A, Nojiri T, Konishi K, Tokudome T, Miura K, Hosoda H, Hino J, Miyazato M, Kyomoto Y, Asai K, Hirata K, Kangawa K. Atrial natriuretic peptide protects against bleomycin-induced pulmonary fibrosis via vascular endothelial cells in mice: ANP for pulmonary fibrosis. Respiratory Research. 2017;**18**(1):1. DOI: 10.1186/s12931-016-0492-7

[10] Hotchkiss A, Feridooni T, Baguma-Nibasheka M, McNeil K, Chinni S, Pasumarthi K. Atrial natriuretic peptide inhibits cell cycle activity of embryonic cardiac progenitor cells via its NPRA receptor signaling axis. American Journal of Physiology. 2015;**308**(7):C557-C569

[11] Banerjee P, Bandyopadhyay A. Cytosolic dynamics of annexin A6 trigger feedback regulation of hypertrophy via atrial natriuretic peptide in cardiomyocytes. Journal of Biological Chemistry. 2014;**289**(9):5371-5385

[12] Vesely D. Heart peptide hormones: Adjunct and primary treatments of cancer. Anticancer Research. 2016;**36**(11):5693-5700

[13] Zhi H, Wang H, Li T, Pin F. Correlated analysis and pathological study on insulin resistance and cardiovascular endocrine hormone in elderly hypertension patients. Diabetology and Metabolic Syndrome. 2015;**9**(2):67-70

[14] Moro C. Targeting cardiac natriuretic peptides in the therapy of diabetes and obesity. Expert Opinion on Therapeutic Targets. 2016;**20**(12):1445-1452

[15] Maksimov V, Korostyshevskaya I, Markel' A, Yakobson G, Rudenko N. Inhibition of secretory activity of atrial myocytes in hypertensive rats after losartan treatment. Bulletin of Experimental Biology and Medicine. 2014;**158**(9):279-281

[16] Mifune H, Nishi Y, Tajiri Y, Yabuki A. Different A-type natriuretic peptide level in five strains of mice. Journal of Veterinary Medical Science. 2012;**74**(4):499-502

[17] Bugrova M, Kharkovskaya E, Yakovleva E. The effect of mexidol on atrial natriuretic peptide in langendorf rat heart preparation. Modern Technologies in Medicine. 2014;**2**:25-31

[18] Bugrova M. Atrial and brain natriuretic peptide of cardiac muscle cells in post-reperfusion period in rats. Cytology. 2016;**6**(2):129-134

[19] Rakhcheeva M, Bugrova M. Changes in the proportion of A- and B-types of granules containing atrial and brain natriuretic peptides in atrial myocytes in vasorenal hypertension in rats. Cytology. 2010;**8**:629-633

[20] Bugrova M, Abrosimov D, Ermolin I. Ultrastructural morphological characterization of right atrial and left ventricular rat cardiomyocytes during postreperfusion period. Bulletin of Experimental Biology and Medicine. 2017;**163**(6):773-777

[21] Ogawa T, de Bold A. The heart as an endocrine organ. Endocrine Connections. 2014;**3**(2): 31-44. DOI: 10.1530/EC-14-0012

[22] Li H, Zhang Y, Wu Q. Role of corin in the regulation of blood pressure. Current Opinion in Nephrology and Hypertension. 2017;**26**(2):67-73

[23] Bugrova M, Abrosimov D, Yakovleva E, Baskina O, Ermolin I. The study on atrial natriuretic peptide of cardiomyocytes in a remote postperfusion period in experiment. Modern Technologies in Medicine. 2013;**5**(4):39-44

[24] Bugrova M, Yakovleva E, Abrosimov D. The relationship of synthesis intensity, accumulation and secretion of natriuretic atrial peptide of cardiac myocytes with cardiac rhythm regulation in rats in early postperfusion period. Modern Technologies in Medicine. 2012;**3**:26-30

[25] Khitrov N. Isolation from nerve effects as a mechanism of adaptation of biological systems in pathology. Bulletin of Experimental Biology and Medicine. 1998;**125**(6):8-14

[26] Arjamaa O, Nikinmaa M. Hypoxia regulates the natriuretic peptide system. International Journal of Physiology, Pathophysiology and Pharmacology. 2011;**3**(3):191-201

[27] Sudarikova Y, Bakeeva L, Tsyplenkova V. Energy-dependent changes in the ultrastructure of mitochondria of human cardiomyocytes in alcoholic heart failure. Archives of Pathology. 1999;**2**:15-20

[28] Klembovsky A, Sukhorukov V. The problem of energy dysfunction of cells in human pathology (pathogenesis and correction). Bulletin of the Russian Academy of natural Sciences. 2007;**4**:62-69

[29] Solov'ev N, Yasnetsov V. Experimental and clinical study of Mexidol effect in some pathology. Determination of possible localization and mechanism of action. Bulletin of Experimental Biology and Medicine. 2006;**1**(Suppl):230-241

[30] Perrin M, Gollob M. The role of atrial natriuretic peptide in modulating cardiac electrophysiology. Heart Rhythm. 2012;**4**:610-615

[31] Imaki R, Niwano S, Niwano H, Satoh D, Yoshida T, Masaki Y, Izumi T. Neutral endopeptidase inhibitor suppresses the early phase of atrial electrical remodeling in a canine rapid atrial pacing model. Indian Pacing and Electrophysiology Journal. 2008;**8**(2):102-113

[32] Bugrova M. The study of atrial natriuretic peptide in arterial hypertension of different genesis in experiment. Morphological Vedomosti. 2015;**2**:28-34

IntechOpen