REVERSIBILITY OF EXPERIMENTAL HYPERTROPHY OF THE MYOCARDIUM OF THE RIGHT VENTRICLE

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Hypertrophy of the right ventricle produced in rabbits by transient experimental pulmonary hypertension is accompanied by thickening of the muscle fibers, an increase in the volume of their nuclei,and an increase in the cytophotometric index of their DNA content. Removal of the conditions contributing to the development of cor pulmonale is followed by a decrease in the weight of the myocardium of the right ventricle, in the volume of its nuclei,and in their DNA content.

Operative and conservative treatment of many chronic lung diseases has recently become much more effective. It is therefore a matter of considerable interest to study the reversibility of hypertrophy of the myocardium of the right ventricle after removal of the cause of cor pulmonale. No systematic morphological investigations in this field could be discovered in the literature, with the exception of a few isolated papers [8, 9, 11].

A previous communication [4] examined the character of hypertrophy of the right ventricle in several types of chronic experimental lung pathology, the possibility of restoration of the normal weight of the myocardium in the recovery period was demonstrated, and these findings were compared with the results of histological examination of the lungs.

The object of the present investigation was to continue the morphometric analysis of the dynamics of changes in the myocardium of the right ventricle, including cytophotometry of the muscle cell nuclei during hypertrophy resulting from a primary lesion of the pulmonary vessels, and also during disappearance of the hypertrophy of the ventricular wall.

EXPERIMENTAL

Chronic pulmonary arteritis was produced in 23 rabbits by intravenous injections of a 1% solution of carboxymethylcellulose (CMC)* by the method of Benkös and co-workers [7]. Purified preparations of CMC (degree of polymerization 400 and 600, coefficient of esterification 87 and 75) were used in the experiments. The results of histological investigation of the lungs (this is the subject of a separate report) showed that CMC produces granulomatosin and destruction of the walls of small arteries, angiosclerosis followed by constriction of the arterial system, and the development of pulmonary hypertension. Five rabbits receiving an intravenous injection of the same volume of physiological saline as of CMC solution acted as controls.

In all cases the walls of the heart were weighed separately by the method of Fulton and co-workers [10] as modified by Köhn and Berg [11], and the ventricular index (weight of myocardium of right ventricle/ weight of myocardium of left ventricle) was calculated. Control values were obtained by the study of 74 hearts of healthy rabbits.

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TABLE 1. Changes in Morphometric Indices and Relative Content of DNA during Hypertrophy of Myocardium of Right Ventricle and Subsequent Decrease in its Weight (M ±m)

	Times of observation (in weeks)	Number of experiments	Weight of myocardium of right ventricle (in g)	Ventricular index	Diameter of muscle fibers (in μ)	Volume of myocardial nuclei (in μ^3)	Cytophotometric index
Control							
I		5	0,95±0,02	0,44±0,01	10,0±0,12	53,0±1,52	1,0
Experiment (development of hypertrophy)							
II	4	4	1,08±0,08	0,59±0,06	11,0±0,21	104,0±2,7	1,16±0,08
III	10	4	1,14±0,14	0,52±0,04	12,6±0,16	100,0±3,9	1,35±0,04
IV	14	4	1,28±0,06	0,58±0,07	13,1±0,15	135,0±3,9	1,66±0,09
End of experiment and decrease in weight of myocardium							
V	4	5	1,21±0,05	0,52±0,03	12,8±0,15	108,0±2,9	1,23±0,08
VI	8	3	1,21±0,03	0,54±0,02	13,1±0,17	111,0±3,3	1,46±0,07
VII	16	3	0,96±0,01	0,48±0,02	10,1±0,15	84,0±3,0	1,21±0,06
		P	I—IV<0,001 V—VII<0,01 IV—V>0,2 I—VII>0,5	I—IV<0,05 V—VII>0,2 IV—V>0,2 I—VII>0,2	I—IV<0,01 V—VII<0,05 IV—V>0,1 I—VII>0,5	II—IV<0,001 V—VII<0,001 V—VI>0,5 IV—V<0,001	II—IV<0,05 VI—VII<0,05 IV—VII<0,01

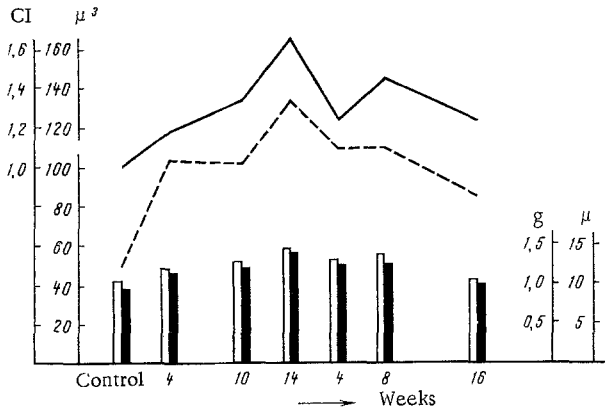

Fig. 1. Changes in morphometric and cytophotometric parameters (mean values) of right ventricle during hypertrophy and its disappearance. Abscissa: duration of experiments (up to 14 weeks) and period after their end (from 4 to 16 weeks). Unshaded columns represent diameter of muscle fibers (in μ), shaded columns represent weight of right ventricle (in g); continuous line indicates cytophotometric index (CI), broken line shows volume of nuclei (in μ^3).

The heart and lungs were fixed in Carnoy's fluid and formalin, and paraffin sections were stained with hematoxylin-eosin, picrofuchsin, and by Feulgen's method for DNA. In each case the diameter of 50-100 transversely divided fibers of the myocardium of the right ventricle, and the transverse diameter and length of 50 nuclei in bundles of muscle fibers divided longitudinally, were measured with a screw-adjusted ocular micrometer. The volume of the nuclei was calculated from the formula for the volume of a cylinder:

$$V = 1/4\ D^2L,$$

where D is the diameter and L the length of the nucleus in microns [12]. Sections of the myocardium cut from pieces of the right ventricle of experimental and healthy rabbits, embedded in the same paraffin block, and stained by Feulgen's method, were used for cytophotometry. By cutting the sections to equal thickness, a comparative cytophotometric determination of the optical density of the nuclei could be carried out, and some idea obtained of the relative DNA content in each nucleus [1]. Cytophotometry was carried out on a integrating microspectrophotometer built in the laboratory of Morphometry and Biophysics of the Institute of Human Morphology, Academy of Medical Sciences of the USSR.

All the numerical data were subjected to statistical analysis.

EXPERIMENTAL RESULTS

During the first weeks of the experiment the ventricular index rose by almost 35% as a result of the development of cor pulmonale (Table 1, Fig. 1). The probable reason for this was a rapid increase in resistance of the vascular system of the lungs due both to mechanical blocking of the precapillaries by aggregates of large CMC molecules (mol. wt. 50,000), playing the role of microemboli, and also to constriction of the smaller arteries. Although the increase in absolute weight of the right ventricle at this period was slight and within the limits of random variation of weight, nevertheless the muscle fibers were appreciably thickened and the volume of the nuclei increased on the average by more than twice (whereas in the control most—65%—nuclei had a volume of 40-60 μ^3, after 4 weeks of the experiment 50-75% of nuclei had a volume of 100 μ^3 and more). Meanwhile an increase in the optical density of the nuclei was observed, indicating some increase in the DNA content. This increase in size of the nuclei was probably a reaction to stretching of the muscle fibers of the right ventricle through a rapid increase in the load. Similar changes in the nuclei have been described in smooth-muscle cells of the inferior vena cava in rats [2].

During the next 4-6 weeks there was a marked increase in absolute weight of the right ventricle accompanied by very slight changes in the ventricular index. The diameter of the muscle fibers and the cytophotometric index continued to increase, whereas the volume changed only slightly compared with that found previously. Toward the end of the 14th week, an increase in the cytophotometric index was again observed. There was a considerable increase in the number of thick muscle fibers (more than 60% of fibers were 13-15 μ in diameter) with large nuclei, among which more than 50% had a volume of 120-180 μ. Calculations showed a positive correlation between the volume of the nuclei and weight of the myocardium (coefficient of correlation $r = 0.66$, mean error $m_r = 0.09$).

The precapillary pulmonary hypertension arising as the result of inflammatory changes in the small branches of the pulmonary artery thus led after 14 weeks to a marked increase in the absolute and relative weights of the right ventricle, i.e., to hypertrophy. At the same time, thickening of the muscle fibers and an increase in the volume of this nuclei, characteristic of hypertrophy, were observed.

During the first 8 weeks after the last injection of CMC the weight of the myocardium of the right ventricle and diameter of its fibers remained more or less stable (differences between parameters then and previously not significant; Table 1, Fig. 1). Conversely, the mean volume of the nuclei fell appreciably, and the cytometric index of DNA content in the nuclei was reduced (by almost 35%). The relatively rapid decrease in these values can apparently be attributed to reduction of the functional load on the myocardium because of cessation of the microembolism and resolution of the vasoconstriction of the smaller arteries. By the end of the 8th week, the cytometric index was once again slightly increased, corresponding to an increase in the proportion of relatively large nuclei in these cases.

Not until 16 weeks after the end of the experiment were most of the parameters once again close to the control value, although the nuclei still remained larger, on the average, than was normal.

The changes in the ventricular index were not in harmony with the remaining indices because the ventricular index is affected by fluctuations in the weight of the left ventricle through disturbance of the hemodynamics. A detailed analysis of this dependence was not among the objects of this investigation.

The correlation established between the size of the nucleus and the cytophotometric index of its DNA content agrees with the results of those workers [13] who detected an increase in the DNA content in the nuclei of hypertrophied muscle fibers cytophotometrically. No satisfactory explanation of this fact has yet been given. The possible role of polyploidy, which is regarded as an essential link in the mechanism of myocardial hypertrophy [3], has not been confirmed because the increase in DNA content in the hypertrophied

heart has been found to be due to activation of its synthesis in the connective-tissue structures of the heart and not in nuclei of its muscle fibers [5]. On the other hand, the nuclear volume is also dependent to some extent on the content of protein not bound with DNA [6]. The possibility cannot therefore be ruled out that the increase in protein synthesis during hypertrophy may lead to an increase in the volume of the nuclei, while the decrease in intensity of metabolism with a reduction in the functional load on the myocardium during the recovery period may lead to a decrease in the nuclear volume.

LITERATURE CITED

1. G. G. Avtandilov, Great Medical Encyclopedia, Annual Supplement [in Russian], Vol. 2, Moscow (1969), p. 1103.
2. O. Ya. Kaufman, Byull. Éksperim. Biol. i Med., No. 11, 117 (1968).
3. F. Z. Meerson, The Myocardium in Hyperfunction, Hypertrophy, and Failure of the Heart [in Russian], Moscow (1965).
4. E. N. Nesterov, Kardiologiya, No. 12, 120 (1968).
5. P. N. Rumyantsev, G. A. Alekhina, and F. Z. Meerson, Tsitologiya, No. 3, 311 (1967).
6. Ya. E. Khesin, Dimensions of the Nuclei and Functional State of Cells [in Russian], Moscow (1967).
7. S. Benkös, V. Balazs, M. Frolich, et al., Z. Ges. Inn. Med., 17, 542 (1962).
8. C. Büchner and G. Köhn, Beitr. Path. Anat., 121, 170 (1959).
9. D. Ferguson, E. Bernar, and R. Varco, Proc. Soc. Exp. Biol. (New York), 89, 492 (1955).
10. R. Fulton, E. Hutchinson, and A. Jones, Brit. Heart J., 14, 413 (1952).
11. G. Köhn and P. Berg, Beitr. Path. Anat., 132, 86 (1965).
12. M. Palkovits and B. Hajtman, Z. Mikr.-Anat. Forsch., 73, 324 (1965).
13. W. Sandritter and G. Scomazzoni, Nature, 202, 100 (1964).