COMPARATIVE CHARACTERISTICS OF HIGH-POLYMER RNA FROM TISSUES AND CELL FRACTIONS FROM DIFFERENT PARTS OF THE DOG HEART

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The content and composition of high-polymer RNA obtained from whole tissue and isolated nuclear and mitochondrial fractions of the right and left ventricles of the dog's heart were studied. The concentration of high-polymer RNA both in the tissues and cell fractions of the left ventricle was 20-25% higher than in the right. The nucleotide composition of high-polymer RNA of tissue and cell fractions in the left and right ventricles was the same. A marked difference was found between the nucleotide composition of high-polymer RNA from the nuclei, mitochondria, and tissue of dog heart muscle.

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Modern methods of cell fractionation [2-4, 6, 8, 9] have made possible the isolation of individual fractions of intracellular structures and elucidation of the principles governing RNA distribution between the organoids of individual cells. Results obtained by different workers [11-13, 17, 20, 21] investigating the RNA composition of cell fractions reveal considerable differences between them. In these investigations the isolation of cell structures and subsequent study of the RNA distribution in them and its composition were carried out on total RNA isolated from the whole organ.

Having regard to the fact that the ventricles perform contractile work of different intensities, and having previously established that the content of both total and high-polymer RNA differs in the tissues of the right and left ventricles, the next step was to determine whether this difference is characteristic of individual cell structures of the myocardium and whether or not it depends on the state of its different parts.

EXPERIMENTAL METHOD

The nucleus and mitochondria were isolated from the left and right ventricular walls of dogs. Removal of the heart, the methods used to isolate the mitochondria, and methods of testing the purity of the preparation obtained were described previously [5]. The nucleus was isolated by Marshak's method [19]. Purity of the preparation was determined by microscopic examination of a nuclear suspension stained with methylene blue. To isolate high-polymer RNA, the cell fractions and tissue homogenate were subjected to phenolic deproteinization followed by salt fractionation [1, 15, 16]. The concentration of high-polymer RNA was determined spectrophotometrically [1, 7]. The results are expressed as μg RNA/mg protein [7, 18].

The RNA nucleotides were separated on columns of the cation-exchange resin Dowex $5OH^+$ [14]. The content of each nucleotide was expressed in molar percentages.

EXPERIMENTAL RESULTS AND DISCUSSION

The results given in Table 1 show that the concentration of high-polymer RNA in the tissue, mitochondria, and nuclei of the left ventricle was higher than in the right. The difference for nuclei and mitochondria was about 25%, and for tissue about 20%. The quantitative distribution of high-polymer RNA between the studied fractions of heart muscle was as follows: the nuclei contained a little more than 10% of the RNA and the mitochondria about 22%. This is rather higher than the RNA content in the nuclei and mitochondria of parenchymatous tissue, where the nuclei contain about 7% and the mitochondria 17%.

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TABLE 1. Concentration (in μg RNA/mg protein) of High-Polymer RNA in Tissue and Cell Fractions of Left and Right Ventricles of Intact Dogs (M ± m)

Test object	Left ventricle	Right ventricle	P
Cell nucleus	2,42± 0,25	1,81± 0,16	<0,001
Mitochondria	5,30± 0,102	3,69± 0,209	<0,001
Tissue	21,6± 0,705	17,7± 0,399	<0,001

TABLE 2. Nucleotide Composition (in moles %) of High-Polymer RNA from Tissue and Cell Fractions of Left and Right Ventricles of Intact Dogs (M ± m)

Test object	Left ventricle					
	G	A	C	U	$\frac{G+C}{A+U}$	$\frac{Pu}{Py}$
Cell nucleus	29,6± 0,324	20,4± 0,132	30,3± 0,38	19,7± 0,570	1,52± 0,025	1,0+0,165
Mitochondria	27,4± 0,89	21,5± 0,89	28,0± 0,56	23,1± 0,78	1,25± 0,071	0,96± 0,018
Tissue	31,2± 0,509	19,03± 0,60	28,06± 0,35	21,7± 0,185	1,45± 0,031	1,01± 0,028

Test object	Right Ventricle					
	G	A	C	U	$\frac{G+C}{A+U}$	$\frac{Pu}{Py}$
Cell nucleus	30,9± 0,645	19,5± 0,397	29,9± 0,585	19,7± 0,394	1,55± 0,0012	1,01± 0,0145
Mitochondria	27,5± 0,35	21,1± 0,945	28,5± 0,255	22,9± 0,745	1,27± 0,022	0,95± 0,0025
Tissue	30,73± 0,405	19,21± 0,415	28,24± 0,503	21,82± 0,360	1,44± 0,0345	1,0± 0,022

Note: g, guanine. A, adenine. L, cytosine, U, uracil. Pu, purines; Py, pyrimidines.

TABLE 3. Nucleotide Composition (in moles %) of High-Polymer RNA of Isolated Mitochondrial Fraction from Dog's Heart (our data) and Rat's Liver (data of Mil'man and Kozyaeva)

Dog's heart					Rat's liver				
A	G	C	U	$\frac{G+C}{A+U}$	A	G	C	U	$\frac{G+C}{A+U}$
21,5	27,4	28,0	23,1	1,25	20,8	27,7	27,7	23,8	1,24

The nucleotide composition of high-polymer RNA of the tissue, nuclei, and mitochondria of the left and right ventricles was identical (Table 2). A marked difference was observed between the nucleotide composition of high-polymer RNA of the mitochondrial and nuclear fractions. The base ratio (G + C/A + U) of high-polymer RNA of the mitochondrial fraction was 1.25 ± 0.071, and for the nuclear fraction it was much higher, i.e., 1.50 ± 0.024. These results for the nucleotide composition of the high-polymer RNA of the isolated mitochondrial fraction are in agreement with those obtained by Mil'man and Kozyaeva [6] for the nucleotide composition of high-polymer RNA of the mitochondrial fraction of rat liver (Table 3).

The comparative data for the nucleotide composition of high-polymer RNA of the dog's heart and rat's liver given above suggest that similarity of nucleotide composition is observed not only for ribosomal RNA of one class of animals, but that it also extends to the high-polymer mitochondrial RNA of this class.

LITERATURE CITED

1. G. P. Georgiev, Biokhimiya, No. 6, 1095 (1961).
2. A. L. Dounce, in: The Nucleic Acids [Russian translation], Moscow (1957), p. 51.
3. I. B. Zbarskii and G. P. Georgiev, Biokhimiya, No. 2, 192 (1959).
4. I. B. Zbarskii and G. P. Georgiev, Tsitologiya, No. 2, 95 (1960).
5. L. A. Kopteva and V. I. Biryuzova, Byull. Éksperim. Biol. i Med., No. 7, 19 (1968).
6. L. S. Mil'man and V. A. Kozyaeva, Tsitologiya, No. 1, 42 (1962).
7. A. S. Spirin and A. I. Belozerskii, Dokl. Akad. Nauk SSSR, 113, No. 3, 650 (1957).
8. V. V. Allfrey, A. E. Mirsky, and H. Stern, Advances Enzymol., 16, 114 (1954).
9. J. Chauweau, Y. Moulé, and C. Rouiller, Bull. Soc. Chim. Biol. (Paris), 39, 1521 (1957).
10. J. N. Davidson and M. S. Smellie, Biochem. J., 52, 594 (1952).
11. D. Elson and E. Chargaff, Nature, 173, 1073 (1954).
12. D. Elson and E. Chargaff, Biochim. Biophys. Acta, 17, 362 (1955).
13. G. H. Hogeboom and W. E. Schneider, J. Biol. Chem., 172, 619 (1948).
14. S. Katz and D. Comb, J. Biol. Chem., 238, 3065 (1963).
15. K. S. Kirby, Biochem. J., 64, 405 (1956).
16. K. S. Kirby, Biochem. J., 66, 495 (1957).
17. G. De Lamirande, C. Allard,, and A. Cantero, Biochim. Biophys. Acta, 27, 395 (1958).
18. O. H. Lowry et al., J. Biol. Chem., 93, 265 (1951).
19. A. Marshak, J. Gen. Physiol., 25, 275 (1941).
20. S. Osawa and Y. Hotta, Biochim. Biophys. Acta, 25, 656 (1957).
21. T. D. Weill, M. Ledig, and P. Mandel, Biochim. Biophys. Acta, 20, 430 (1956).