

same as in samples containing adenosine. Addition of progesterone or PVP-HC alone to the incubation medium in the above concentrations did not change the cAMP level in the cells. Consequently, progesterone completely abolishes the effect of PVP-HC on the adenosine-mediated increase in the cAMP concentration, or in other words, the effect of PVP-HC was specific for the glucocorticoid hormone. There is as yet no clear idea of the molecular mechanism of action of progesterone as a glucocorticoid antagonist [8]. The data suggest that progesterone can interact directly with thymocyte membrane receptors for glucocorticoids.

It can be concluded from these results that the modulating effect of cortisol on the cAMP concentration in the thymocytes is one way whereby the hormonal activity of glucocorticoids is realized. The results of the study of the effect of progesterone on potentiation of the action of the adenylate cyclase activator, adenosine, by PVP-HC are evidence that the plasma membrane of thymus lymphocytes can be regarded as the first stage in interaction of glucocorticoids and their antagonists with target cells.

LITERATURE CITED

1. E.-E. Baulieu, in: *Cell Membrane Receptors for Drugs and Hormones*, R. W. Straub and L. Bolis (eds.), New York (1978), pp. 129-149.
2. T. M. Morozova, V. E. Volchkov, T. I. Merkulova, and N. N. Nagibneva, *Dokl. Akad. Nauk SSSR*, 272, No. 6, 1494 (1983).
3. T. G. Pukhal'skaya and P. V. Sergeev, *Zh. Mikrobiol.*, No. 10, 56 (1983).
4. V. B. Rozen, *Principles of Endocrinology* [in Russian], Moscow (1984).
5. P. V. Sergeev, G. V. Kalinin, and A. S. Dukhanin, *Byull. Eksp. Biol. Med.*, No. 8, 192 (1986).
6. P. V. Sergeev, G. V. Kalinin, A. S. Dukhanin, and A. V. Semeikin, *Neurohumoral Regulation of Immune Homeostasis* [in Russian], Leningrad (1986), pp. 64-65.
7. M. L. Elks, V. C. Manganiello, and M. Vaughan, *Endocrinology*, 115, 1350 (1984).
8. S. S. Simons, R. E. Shlenbaker, and H. J. Eisen, *J. Biol. Chem.*, 258, 2229 (1983).
9. Y. Zick, R. Cesla, and S. Shaltiel, *Biochim. Biophys. Acta*, 762, 355 (1983).

PROTEIN SYNTHESIS AND FREE AMINO ACID LEVELS IN ORGANS OF RATS WITH EXPERIMENTAL PERITONITIS

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Mechanisms of the disturbance of homeostasis which accompanies the development of peritonitis remain partly unexplained, and are largely associated with the character of the metabolic disturbances in this state. The development of peritonitis is characterized, in particular, by a marked catabolic reaction and a negative protein balance, a leading role in the genesis of which is ascribed to irreversible losses of proteins [5]. Meanwhile the disturbance of synthesis and processing of various classes of RNA [1, 2], and also disturbance of the structural integrity of the polysomes of the liver and spleen [4] are evidence that the disturbance of protein metabolism in peritonitis is also linked with damage to the protein-synthesizing apparatus of the cells in the organ studied. Protein synthesis, as we know, includes several consecutive stages, each of which may become the limiting stage in the time course of development of peritonitis.

The aim of this investigation was to study protein synthesis in different organs of rats and to compare it with changes in the reserves of precursors, namely free amino acids, during the development of experimental peritonitis.

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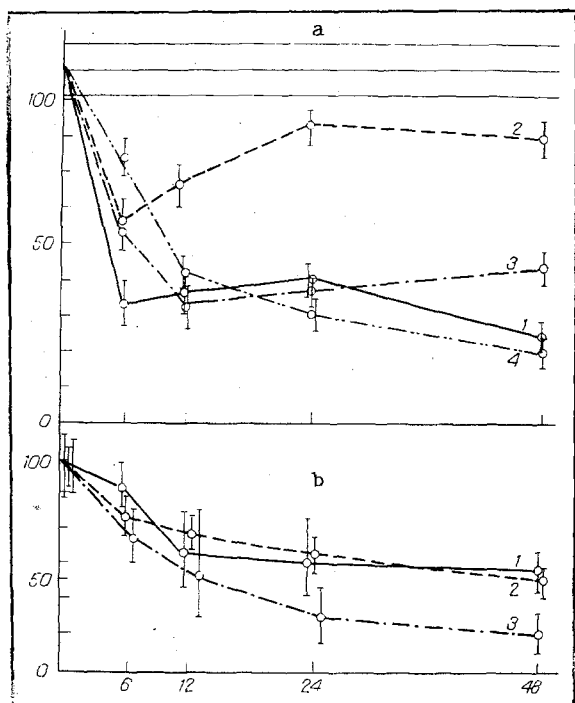


Fig. 1. Incorporation of ^{14}C -digest of chlorella into proteins of rat organs (a) and change in visceral coefficient during development of peritonitis (b). Abscissa, time after operation (in h); ordinate, values of parameters studied (in percent of normal). 1) Liver, 2) kidneys, 3) spleen, 4) skeletal muscles.

EXPERIMENTAL METHOD

Experiments were carried out on 120 male Wistar rats weighing 200-300 g, in which peritonitis was induced by the method described previously [6]. The animals were killed 6, 12, 24, and 48 h after the initiation of peritonitis and changes in weight of the liver, kidneys, and spleen, expressed relative to the total body weight (visceral coefficient) were recorded. Parallel with the above, incorporation of labeled amino acids into total protein of these organs was studied in the liver, kidneys, spleen, and skeletal muscles. The labeled precursors used consisted of a ^{14}C -protein digest of chlorella (specific radioactivity 37 MBq/g, Czechoslovakia), which was injected intraperitoneally in a dose of 1.85 MBq 1 h before sacrifice. Specific radioactivity of the acid-soluble fraction was determined as the ratio of radioactivity of the total volume of the fraction to absorption of light by the same sample at 280 nm (SRASF/E_{280}). Total radioactivity was measured on an Ultra-Beta counter (Sweden) in toluene scintillator. The total concentration of free amino acids in the tissues and blood serum was determined chromatographically [3].

EXPERIMENTAL RESULTS

The development of peritonitis was accompanied by a fall in the visceral coefficient and a decrease in the uptake of radioactive precursors (^{14}C -amino acids) into tissue proteins (Fig. 1). A decrease in the relative weight of the kidneys and spleen was observed as early as 12 h after the beginning of peritonitis, whereas significant changes for the liver were not observed until after 48 h. Meanwhile reduced incorporation of the label was observed as early as 6 h after the beginning of the experiment, and was most marked in the case of the liver and spleen. By 48 h after the beginning of peritonitis the maximal reduction of incorporation of radioactive precursors was recorded in tissues of the liver and skeletal muscles (Fig. 1).

After 12 h of peritonitis an increase in the total concentration of free amino acids was observed in tissues of the kidneys, spleen, and skeletal muscles (Table 1). This trend of changes in the blood serum was significant only after 24 h of the experiment, and in the liver tissues after 48 h of peritonitis. In the absence of an intake of exogenous amino acids, this time course is evidence that the development of peritonitis leads to rapid intensification of catabolism, mainly in skeletal muscle tissue, and also in the spleen and kidneys. Protein breakdown in the liver or the arrival of an excess of tissue amino acids of extrahepatic origin in the liver did not take place until 48 h after the beginning of the experiment. Comparison of the time course of changes in incorporation of the label into tissue proteins with changes in the free amino acid concentrations in these organs indicate that the primary response of the liver to the development of a suppurative inflammatory process is a decrease in the intensity of protein biosynthesis, whereas tissue catabolism becomes manifested much later. Not until 48 h after the beginning of peritonitis did the free

TABLE 1. Changes in SRASF/E₂₈₀ of Organs (A) and in Concentrations of Free Amino Acids (B) during Development of Peritonitis (M ± m)

Parameter	Organ	Time course of experimental peritonitis				
		normal state	6 h	12 h	24 h	48 h
SRASF/E ₂₈₀	Liver	4424,5±213,2 (10) 0,001	3781,0±233,1 (10) 0,001	3143,0±224,1 (10) 0,001	3354,4±198,8 (10) 0,001	4238,0±243,6 (7) 0,001
	<i>p</i>	—	0,001	0,001	0,01	0,05
	Kidneys	2969,0±208,1 (9) 0,001	2800,0±121,2 (10) 0,001	2587,0±166,7 (10) 0,001	2743,8±300,1 (8) 0,001	2928,1±218,1 (8) 0,001
	<i>p</i>	—	0,05	0,05	0,05	0,05
	Spleen	5320,0±158,2 (10) 0,001	3731,0±311,7 (10) 0,001	3402,0±180,7 (10) 0,001	3654,4±197,6 (9) 0,001	4865,0±97,1 (8) 0,001
	<i>p</i>	—	0,001	0,001	0,001	0,05
Concentration of free amino acids, mg%	Liver	211,9±3,79 (7) 0,001	—	219,4±8,42 (7) 0,001	272,2±11,48 (7) 0,001	389,7±23,57 (7) 0,001
	<i>p</i>	—	—	0,05	0,05	0,01
	Kidneys	449,6±20,83 (7) 0,001	—	553,9±24,02 (7) 0,001	628,8±24,66 (7) 0,001	547,6±7,78 (7) 0,001
	<i>p</i>	—	—	0,01	0,001	0,01
	Spleen	208,7±23,22 (7) 0,001	—	348,2±22,78 (7) 0,001	336,2±17,56 (7) 0,001	672,2±22,91 (7) 0,001
	<i>p</i>	—	—	0,001	0,001	0,001
	Skeletal muscle	159,5±1,71 (7) 0,001	—	240,0±5,06 (7) 0,001	331,8±1,83 (7) 0,001	329,9±8,0 (7) 0,001
	<i>p</i>	—	—	0,001	0,001	0,001
	Blood serum	187,8±9,59 (7) 0,001	—	196,6±4,18 (7) 0,001	212,1±5,11 (7) 0,001	303,5±9,46 (7) 0,001
	<i>p</i>	—	—	0,05	0,05	0,001
	<i>p</i>	—	—	—	—	—
	<i>p</i>	—	—	—	—	—

amino acid level rise, accompanied by a fall in relative weight of the liver. The reduced rate of synthesis of protein and the more rapid rate of their breakdown took place simultaneously in the kidneys, spleen, and skeletal muscles. Under these circumstances an increase in the free amino acid concentration in the blood serum, which did not take place until 24 h after the beginning of peritonitis, is evidence of relative retention of amino acids released from proteins in the organs, and of their nonarrival in the systemic blood flow during the first 12 h of peritonitis. Meanwhile the value of SRASF/E₂₈₀ fell in the liver and spleen, but not in the kidneys ($p < 0.05$). In the absence of any increase in the level of free amino acids in the liver, this change in this particular parameter mainly reflects the reduced content of label in the organ, whereas the decrease in SRASF/E₂₈₀ in the spleen is evidently connected with an increase in the concentration of aromatic amino acids, more especially of phenylalanine and tryptophan. After 48 h of peritonitis, when free amino acids have arrived in the blood stream, changes in the value of SRASF/E₂₈₀ cancelled each other out.

The changes described above indicate as a whole that during peritonitis disturbances of protein metabolism in the organs and tissues are characterized both by a decrease in the rate of protein synthesis and by a simultaneous increase in the intensity of tissue protein catabolism, reflected in an increase in the concentration of free amino acids. Consequently the disturbances of protein synthesis are not the direct consequence of a deficiency of precursors (free amino acids). Meanwhile, during the first 24 h of peritonitis, the normal exchange of amino acids between the organs and the systemic circulation is disturbed, evidently because of the centralization of the circulation that is characteristic of this state [7].

LITERATURE CITED

1. E. V. Pasternak, V. V. Prosvirin, and I. F. Paskevich, Vopr. Med. Khimii, No. 2, 57 (1984).
2. E. V. Pasternak, V. V. Prosvirin, and I. F. Paskevich, Biochemical Mechanisms of Regulation of Genetic Activity [in Russian], Kiev (1984), p. 101.
3. S. V. Pashkina, Modern Methods in Biochemistry [in Russian], Moscow (1964), p. 137.
4. V. V. Prosvirin, N. M. Martynyuk, and I. F. Paskevich, Vopr. Med. Khimii, No. 1, 85 (1986).

5. B. D. Savchuk, Suppurative Peritonitis [in Russian], Moscow (1979).
6. F. F. Usikov, L. D. Romanova, L. S. Goncharova, et al., Khirurgiya, No. 8, 127 (1984).
7. R. Wilson, Intens. Care Med., 6, 89 (1980).

PHYSICOCHEMICAL PROCESSES IN A PHOTOEXCITED GLYCINE SOLUTION

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Data obtained with a photomultiplier (PM), during recording of the radiation from an excited glycine solution, add to the views formed previously regarding its after-luminescence and based on a study of the mitogenetic radiation of glycine by a method of biological detection.

We may perhaps first summarize briefly the original data, full details of which were given in [1-4]. Soon after short-term irradiation (seconds) of a 0.5% glycine solution with light from a mercury-quartz lamp, the intensity of which was reduced by about 4 orders of magnitude, long-term (several hours) radiation arises in the solution in the UV region of the spectrum. This radiation disappears after the solution is heated to 80°C. After dialysis of the irradiated amino acid through collodion film, the radiation is preserved only in the solution in the inner compartment of the container. It was suggested that this radiation is connected with the active influence on the glycine molecule of a polypeptide formed during the first few minutes after photoexcitation. Its concentration, however, is maintained at a very low level, due to the considerable reversibility of the condensation process.

In fact, addition of a small portion of emitting glycine to a much larger volume of fresh solution led to the appearance of radiation in the latter, which behaved in the same way as the radiation of the original solution. These "transfers" could be repeated many times.

It was also shown that during the first minutes after photoexcitation of glycine, its emission spectrum contains bands characteristic of free NH_2 , CO, and OH radicals, which later were replaced by the appearance of a band in the 230 nm region, corresponding to fluorescence of NH_3 . It can thus very probably be concluded that a chemically active polypeptide, an analog of the prosthetic group of the enzyme deaminase, capable of inducing a mild degree of oxidative deamination of the glycine molecule, is synthesized from the initial photodissociation products.

Later facts obtained by the use of a photomultiplier, recording in the visible region of the spectrum, confirmed and, at the same time, supplemented the previous data.

EXPERIMENTAL METHOD

To investigate the fluorescence of glycine, the apparatus illustrated in Fig. 1 was set up. The FÉU-106 PM, with multialkali photocathode, capable of recording light rays within the spectral range from 170 to 830 nm, was used as radiation receiver. The PM was tuned to operate on the pulsed mode (counting photons). The output signal, after appropriate amplification, was recorded by the pulse counters 1 and 2. To reduce thermoemission noise the PM was placed in the chamber 4, packed with dry ice, and in this case the frequency of counting of dark pulses on the plateau of the counting curve did not exceed 1 Hz. To increase the effective area of the photocathode the quartz photon 7 was fixed on the end window of the PM by means of optical glue. Radiation from the cuvette 13 to be investigated was focused on

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