

On addition of desalinified pancreatic juice (50 μ g protein), heated to 100°C for 3 min, to a sample containing 1 μ g of the enzyme preparation and 1.38 mM cholesteryl oleate, a two-fold increase in enzyme activity was observed. This increase was not connected with the non-specific action of the protein on the enzyme-substrate system, for addition of defatted albumin from human blood serum in a quantity of 400 μ g had no action on the activity of the enzyme. It can be concluded from these results that an activator of cholesterol esterase is present in the pancreatic juice.

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SOME CHARACTERISTICS OF SOLUBLE PROTEINS OF PACINIAN CORPUSCLES

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To study the mechanisms lying at the basis of mechanoreceptor functions information must be obtained on the biochemical processes conducted in them. Yet there is extremely little information in the literature even on the chemical composition of tissue receptors of mechanical sensation. Special attention in this direction is naturally drawn to proteins, more especially because with Pacinian corpuscles such information is limited to data on the total protein content and the content of certain amino acids. Of the proteins it is the soluble proteins that are most accessible for study. They possess the highest rate of turnover and they evidently perform chiefly metabolic functions [2].

In this connection the study of the protein composition of the medium surrounding the mechanically sensitive unmyelinated nerve ending, which has a specific ionic composition, was of definite interest and the investigation described below was aimed at its study.

EXPERIMENTAL METHOD

Pacinian corpuscles were isolated from the mesentery and pancreas of adult cats. To obtain a sufficient quantity of material the receptors were lyophilized. Water- and salt-soluble proteins were successively extracted from the homogenate with 0.9% and 10% NaCl. Gel-filtration of proteins were carried out on Sephadex G-75. The columns were calibrated with proteins of known molecular weight: trypsin, pepsin, ovalbumin, and serum albumin. The protein content in the samples was determined by Lowry's method and spectrophotometrically from absorption in the UV region. The content of nucleotides in the fractions also were

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TABLE 1. Protein Composition of Pacinian Corpuscles

| Carbohydrates | Water-soluble proteins | | | Carbohydrates | Salt-soluble proteins | | |
|---------------|------------------------|------|-----------------------|---------------|-----------------------|------|-----------------------|
| | mol. wt. | % | nucleotide/protein, % | | mol. wt. | % | nucleotide/protein, % |
| 1 | ≥64 000 | 53,5 | 15,9 | 1 | ≥96 000 | 10,0 | 27,4 |
| 2 | 25 000 | 9,3 | 23,8 | 2 | 86 000 | 14,1 | 21,6 |
| 3 | 9 000 | 32,6 | 27,5 | 3 | 64 000 | 4,4 | 28,3 |
| 4 | 6 000 | 4,6 | 35,9 | 4 | 44 000 | 20,8 | 27,0 |
| | | | | 5 | 20 000 | 12,0 | 31,5 |
| | | | | 6 | 6 000 | 39,2 | 25,5 |

TABLE 2. Carbohydrate Content (in µg/mg protein) in Protein Fractions of Pacinian Corpuscles

| Carbohydrates | Total protein | Water-soluble proteins | Salt-soluble proteins |
|-----------------|---------------|------------------------|-----------------------|
| Hexoses | 0,38±0,03 | 0,28 | 0,32 |
| Hexuronic acids | 0,71±0,1 | 0,53 | 0,81 |
| Neuraminic acid | 6,54±0,6 | 4,02 | 7,38 |

determined by the orcin method and sialic acids by the reaction with thiobarbituric acid [1].

EXPERIMENTAL RESULTS

Pacinian corpuscles are similar in shape to an ellipsoid of rotation. The weight of one receptor, about 1 mm long and 0.5 mm thick, varies from 0.14 to 0.43 mg, on average 0.28 mg. The dry residue of the receptors amounted to 6.7-7.7%, i.e., more than 92% of their composition was water. The total protein content, relative to dry weight, averaged $4.5 \pm 0.16\%$, with variation in different animals from 3.4 to 54%; relative to dry weight, the range was from 58.4 to 67.2%.

Data on the fractional composition of water- and salt-soluble proteins after fractionation on Sephadex are given in Table 1.

As Table 1 shows, about half of the water-soluble proteins was accounted for by proteins with a molecular weight of over 64,000, the rest by proteins of low molecular weight. In the salt-soluble proteins, on the other hand, lighter fractions predominated. Fraction 4 of water-soluble and fraction 6 of salt-soluble proteins probably included polypeptides.

All protein fractions were characterized by a comparatively high content of nucleotides, which play an important role in metabolic reactions. The highest content of nucleotides was found in the lightest fractions of water-soluble proteins, whereas in fractions of salt-soluble proteins they were comparatively uniformly distributed. In the modern view, such proteins are proteins of tissue structures with the most rapid turnover and they are located chiefly in microsomes, cell nuclei, and cytoplasm [2]. Considering that most of the receptor consists of connective-tissue cells of the outer capsule and of intracapsular fluid, it should be pointed out that all these structural elements of the Pacinian corpuscles are characterized by a well-developed nuclear apparatus and endoplasmic reticulum. Many vesicles, the membrane of which is often fused with the plasma membrane of the cell, evidence of the existence of pinocytosis or exocytosis [3], are present in the cytoplasm of the lamellar cells of the outer capsule and inner bulb.

Analysis of the results showed that the presence of protein-carbohydrate complexes, both glycoproteins and proteoglycans, also is characteristic of the water- and salt-soluble proteins of Pacinian corpuscles (Table 2).

The most characteristic components of the glycoproteins were hexoses, hexosamines, and sialic acids. Hexosamines could not be found in the Pacinian corpuscles, whereas hexoses and neuraminic acid were present in both water-soluble and salt-soluble proteins. The high content of neuraminic acid will be noted. The functions of the glycoproteins are varied. One of them maintains the constancy of the microenvironment surrounding the cell. Neuraminic acid, in the form of acyl derivatives, so-called sialic acids, is an essential component of the neutral glycoproteins. Neuraminic acid participates in the formation of collagen and is also present in large quantities in albumins and globulins. Hexuronic (glucuronic and iduronic) acids are specific components of the proteoglycans. They are also present in both groups of proteins. The most important function of the proteoglycans is considered to be regulation of ionic equilibrium and of movement of water in the tissues. In this respect the data showing the increased content of potassium ions in the fluid of Pacinian corpuscles are particularly interesting. This is probably due to the presence of glycosaminoglycans, active anions capable of binding small cations, in the intercellular medium of the external capsule of the receptors.

The characteristics of the soluble proteins of Pacinian corpuscles described above thus indicate that they can perform metabolic functions and provide the specific microenvironment for the tissue mechanoreceptor.

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CHANGES IN SPATIAL ORGANIZATION IN SARCOPLASMIC RETICULUM

MEMBRANES IN RABBITS WITH EXPERIMENTAL THYROTOXICOSIS

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Membrane structures of some biological objects, namely mitochondria, erythrocytes, and the sarcoplasmic reticulum (SR), are a unique target for thyroid hormones [5, 7, 12, 14]. Structural changes in SR membranes in rabbits with hyperthyroidism caused by prolonged administration of thyroxine to the animals have been described [4]. In particular, labilization of protein-lipid bonds and a decrease in the orderliness and viscosity of the lipid bilayer were noted.

In the present investigation the spatial organization of proteins and lipids in SR membranes was studied depending on the thyroid state of the animal by means of the fluorescent probe pyrene.

EXPERIMENTAL METHOD

Fragments of SR were isolated by the method in [10] from skeletal muscle proteins of rabbits weighing from 2 to 3 kg. Thyrotoxicosis was simulated by intraperitoneal injection of L-thyroxine in 0.01 N KOH according to the scheme described in [5]. Control animals received the corresponding volume of 0.01 N KOH solution. Fluorescence of the proteins was measured on a spectrofluorometer used in [3]. The efficiency of energy transfer from tryptophan residues of protein to pyrene molecules was judged by the degree of extinction of the

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