MORPHOMETRIC ANALYSIS OF REGRESSION OF CHANGES IN THE ADRENAL CORTEX DURING EXPERIMENTAL HYPERCHOLESTEROLEMIA

I. D. Tyuryukhanova and B. Ya. Ryzhavskii

UDC 616.153.922-008.61-092.9-07: 616.453-091

KEY WORDS: cholesterol; adrenal cortex; regression.

Changes taking place in atherosclerosis are to a certain degree reversible; the degree of reversibility, moreover, differs considerably in different organs [1, 3, 5, 6, 9]. It was shown in [8] that changes observed in the adrenal cortex of rabbits receiving cholesterol for 14 days undergo regression, although incomplete, after 1.5 months. Accordingly the question arises of whether regression of morphological changes developing in the adrenal gland during prolonged hypercholesterolemia, when definite signs of depression of function of the adrenocorticocytes, trophic changes in these cells [7] and a decrease in the intensity of steroid production by the adrenal tissue [4] are found, can take place. An examination of this problem is interesting not only in connection with the study of the morphogenesis of atherosclerosis, but also as a model of the state of steroid-producing cells at a time of considerable fluctuation of the blood levels of cholesterol — the substrate for synthesis of biologically active steroids.

EXPERIMENTAL METHOD

The adrenals of adult rabbits belonging to three groups were studied: group 1 consisted of intact animals (11 rabbits), group 2 of animals receiving cholesterol for 2 months in a daily dose of 25 mg/100 g body weight (10 rabbits), and group 3 of animals receiving cholesterol for 2 months and killed 5.5 months after its administration ceased (17 rabbits). Rabbits of the different groups were killed simultaneously during the morning by air embolism. The blood cholesterol level was determined by the method described in [10]. The absolute and relative weight of the adrenals was determined. The area of cross section of the medulla and cortex of the adrenals and of the zones of the cortex, and also the area of cross section of the adrenocorticocytes were measured in sections passing through the central part of the gland, stained with hematoxylin and eosin. In preparations stained with gallocyanin and bromphenol blue the RNA and protein concentrations in the cytoplasm of these cells were determined cytophotometrically as descirbed in [8]. The numerical results were subjected to statistical analysis.

EXPERIMENTAL RESULTS

Administration of cholesterol to the rabbits for 2 months led to a sharp rise in its blood concentration (from a normal 50 ± 6.6 mg % to 582 ± 90.6 mg %). The blood cholesterol level 5.5 months after the end of administration of this substance was virtually the same as in intact animals (49 ± 4.1 mg %), in agreement with data given in [3]. Marked hypercholesterolemia, observed in the animals of group 2, led to a moare than twofold increase in the absolute and relative weight of the adrenal (Table 1). As a result of comparison of the area of cross section of the adrenal medulla and cortex and of the separate zones of the cortex in intact rabbits (group 1) and rabbits receiving cholesterol (group 2) it was concluded that the increase in weight of the gland took place mainly on account of marked hypertrophy of the zona fasciculata. The area of cross section of the zona reticularis also increased significantly. The dimensions of the adrenocorticocytes increased significantly under these circumstances in the zona fasciculata (Fig. 1a, b, Table 1) and zona reticularis (Table 1). This increase could be largely responsible for the hypertrophy of these zones and of the cortex as a whole in the animals of group 2. The concentrations of RNA and total protein in the cytoplasm of the adrenocorticocytes were significantly lower in this case, especially in the

Department of Histology and Central Research Laboratory, Khabarovsk Medical Institute.
(Presented by Academician of the Academy of Medical Sciences of the USSR V. P. Kaznacheev). Translated from Byulleten' Eksperimental'noi Biologii i Meditsiny, Vol. 101, No. 3, pp. 365-367, March, 1986. Original article submitted June 3, 1985.

0007-4888/86/0003-0385$12.50

TABLE 1. Changes in Adrenal Cortex of Rabbits during Administration of Cholesterol for 2 Months and 5.5 Months after Its Discontinuation

Parameters studied	Experimental conditions					
	intact animals	Rabbits receiving cholesterol	P_{1-2}	5.5 months after administration of cholesterol	P_{1-3}	P_{2-3}
Blood cholesterol, mg %	50±6,6	582±90,6	<0,001	49±4,1	>0,2	<0,001
Weight of adrenal						
absolute, mg	257±25	590±94,9	<0,01	288±21,9	>0,2	<0,01
relative, mg/100 g	7±0,7	15±2,1	<0,001	7,1±0,6	>0,2	<0,001
Area of cross section, mm^2 of						
whole adrenal	14,3±0,66	28,4±3,9	<0,001	16,9±0,93	<0,05	<0,001
cortex	13,3±0,66	27,4±3,85	<0,001	15,8±0,91	<0,05	<0,01
zona glomerulosa	2,1±0,22	2,4±0,14	>0,1	2,3±0,07	>0,2	>0,2
zona fasciculata	10±0,61	22,9±3,62	<0,002	11,4±0,89	>0,1	<0,01
zona reticularis	1,4±0,05	2±0,22	<0,01	1,9±0,22	<0,05	>0,2
medulla	1±0,08	1±0,08	—	1,2±0,09	>0,05	>0,05
Area of cross section of adrenocorticocytes, μ^2						
zona glomerulosa	50±2,7	59±4,4	<0,1	51±3,0	>0,2	<0,1
zona fasciculata	115±11,3	238±18,4	<0,001	122±9,5	>0,2	<0,001
zona reticularis	74±7,9	146±11,8	<0,001	80±6,0	>0,2	<0,001
Concentration in cytoplasm, conventional units:						
protein						
zona glomerulosa	343±40,7	218±34,5	<0,05	410±39,3	>0,1	<0,001
zona fasciculata	271±34,5	120±22,1	<0,001	255±21,4	>0,2	<0,001
zona reticularis	251±41,9	146±25,4	<0,05	258±32,4	>0,2	<0,01
RNA						
zona glomerulosa	148±17,8	96±9,6	<0,02	145±11,5	>0,2	<0,01
zona fasciculata	89±10,4	50±5,7	<0,01	83±10,8	>0,2	<0,01
zona reticularis	94±11,7	58±6,6	<0,02	86±9,2	>0,2	<0,02

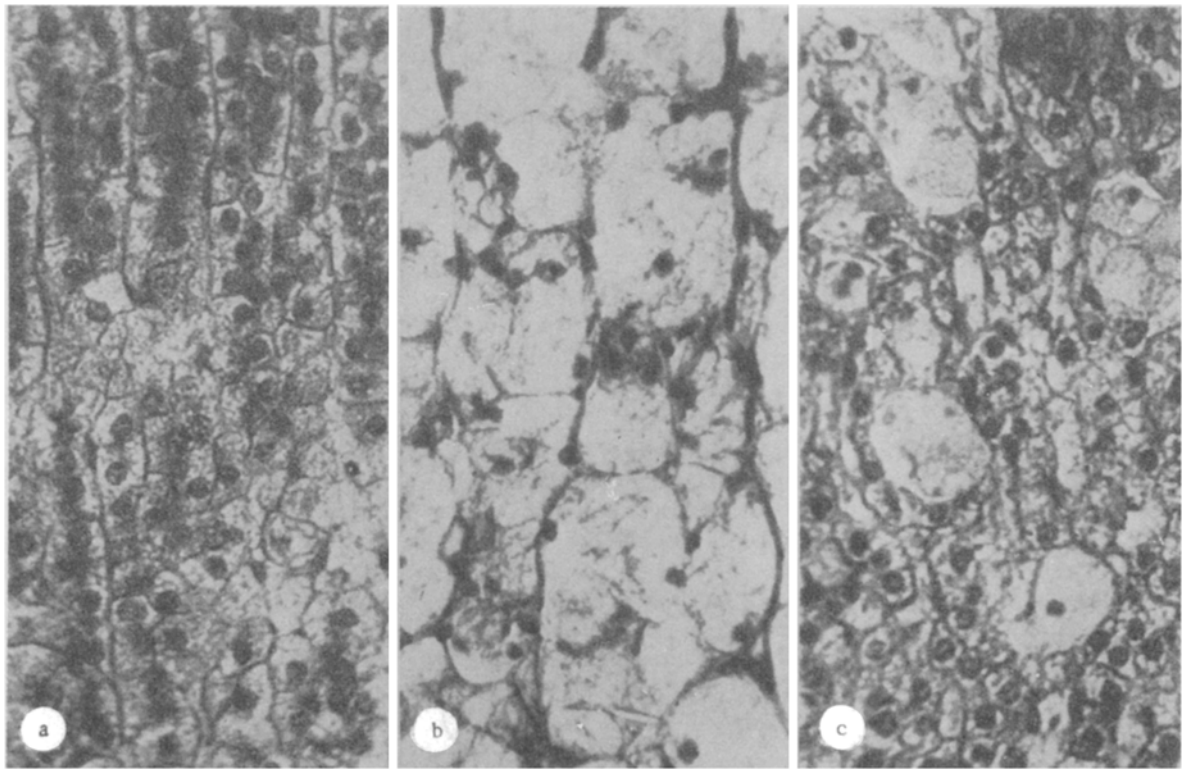

Fig. 1. Zona fasciculata of adrenal cortex of an intact rabbit (a), and of rabbits receiving cholesterol for 60 days (b) and receiving cholesterol for 60 days killed 5.5 months after its discontinuation (c). Hematoxylin and eosin. 500 ×.

zona fasciculata (Table 1). Comparison of changes found after feeding with cholesterol for 2 months and for 14 days [8] showed that their direction was the same. However, deviations from normal of all parameters studied were much greater after 2 months of the experiment. Changes in the parenchymatous cells of the adrenal cortex (a decrease in the RNA and protein

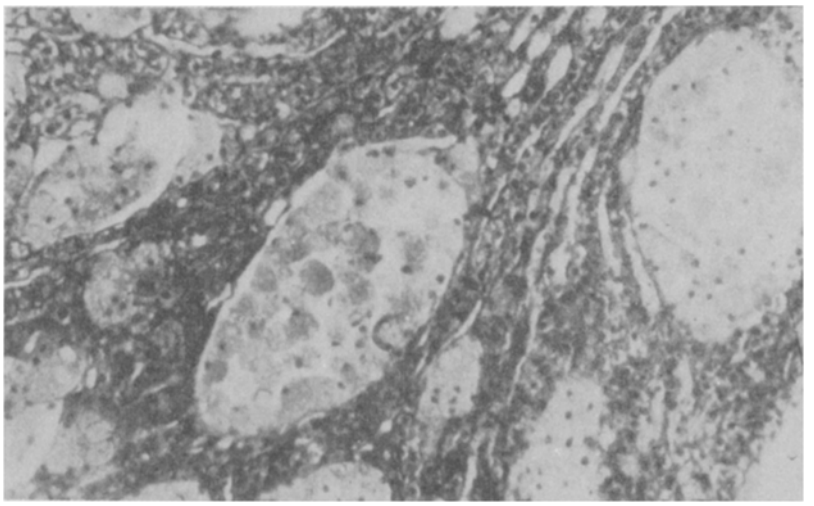

Fig. 2. Adrenal cortex of rabbit receiving cholesterol for 60 days and killed 5.5 months after its discontinuation. Microadenomas in zona fasciculata. Hematoxylin and eosin. 150 ×.

concentrations), found in animals receiving cholesterol for 2 months, can be regarded as factors leading to a decrease in the steroid-producing potential of the adrenal cortex in hypercholesterolemia [2, 4, 11]. Regression of the changes in most of the parameters studied took place 5.5 months after the end of administration of cholesterol to the rabbits (group 3; Fig. 2). No significant differences from normal were observed in the weight of the adrenal, the dimensions of the adrenocorticocytes in all zones, and concentrations of total protein and RNA in their cytoplasm. Meanwhile the area of cross section of the cortex and of the zona reticularias was somewhat larger than in the gland of intact rabbits.

It was shown in [8] that the adrenal cortex of rabbits killed 1.5 months after cholesterol administration for 14 days differed from the gland in intact animals with respect to parameters such as the area of cross-section of the cortex and of the zona fasciculata, and occupied in intermediate position with respect to these parameters between that of intact rabbits and that of rabbits receiving cholesterol for 14 days and killed immediately after its administration ceased. This is evidence of incomplete regression of the changes in these parameters 1.5 months after the end of excessive cholesterol intake. Under the experimental conditions used the adrenals of the rabbits of group 3 also differed with respect to these parameters from the adrenals of intact animals (group 1), although in this case a much longer time had elapsed (5.5 months) after the end of cholesterol administration. Besides the above-mentioned deviations from normal, revealed by the use of morphometric methods, the adrenal cortex of the animals of group 3 also differed from the gland of intact rabbits on examination survey sections. For instance, cells were found in the gland which were similar to those in the animals of group 2, for they were much larger and had a lower nucleo-cytoplasmic ratio and pale cytoplasm. These cells were often arranged in small groups (Fig. 1c). A characteristic feature of the adrenal cortex of the animals of this group was the presence (in 13 of 17 cases) of multiple pale-cell adenomas [11], consisting of large adrenocorticocytes with pale cytoplasm, located in the zona fasciculata and zona reticularis, and standing out clearly against the background of their surrounding tissue. These microadenomas can be regarded as having developed after the end of cholesterol administration, for in the animals of group 2 they were found in only one of ten cases.

The results as a whole are evidence that changes taking place in the adrenal cortex during marked hypercholesterolemia are largely reversible. Meanwhile, it follows from the results that sudden fluctuations in the blood cholesterol concentration leave a definite "structural trace" in the adrenal cortex. These data can be used when the possibility of regression of atherosclerotic changes under the influence of appropriate treatment is being evaluated.

LITERATURE CITED

1. V. Kh. Anestiadi and V. A. Nagornev, Morphogenesis of Atherosclerosis [in Russian], Kishinev (1982).

2. S. N. Babadzhanov, Med. Zh. Uzbekistana, No. 4, 44 (1981).
3. M. A. Krekhova, Vestn. Akad. Med. Nauk SSSR, No. 10, 24 (1965).
4. M. T. Lutsenko, B. Ya. Ryzhavskii, A. D. Chertov, and N. V. Lutsenko, Adaptation of the Organism to an Increased Cholesterol Concentration [in Russian], Blagoveshchensk (1973).
5. R. D. Pal'ma, in: Hormones and Vascular Diseases [in Russian], Moscow (1984), pp. 79-102
6. B. Ya. Ryzhavskii, in: Tissue Metabolism [in Russian], Blagoveshchenski (1978), pp. 108-132.
7. B. Ya. Ryzhavskii, I. D. Tyuryukhanova, N. B. Murzina, and T. P. Markova, Byull. Eksp. Biol. Med., No. 11, 535 (1984).
8. É. M. Tararak and V. M. Ariel', Vestn. Akad. Med. Nauk SSSR, No. 7, 74 (1973).
9. M. Tulczinski, Laboratory Methods in Clinical Research [in Russian], Warsaw (1965).
10. E. V. Uranova, in: Morbid Anatomical Diagnosis of Human Tumors [in Russian], Moscow (1982), pp. 352-354.
11. V. A. Tsiomik, Kardiologiya, No. 8, 70 (1973).

MORPHOLOGICAL AND FUNCTIONAL CHANGES IN THE PANCREAS AFTER EXPERIMENTAL REPAIR OF THE INJURED WALL OF THE CERVICAL PART OF THE ESOPHAGUS

A. S. Zuev, V. V. Zarudin,
A. P. Tyutlikov, A. N. Kartavenko,
R. E. Svistelina, and V. A. Shostak

UDC 616.329-089.844-092.9-07:616.37-07

KEY WORDS: esophagus; injury; pancreas; function; morphology.

There are isolated reports in the literature on the character of disturbances of carbohydrate metabolism after operations on the thoracic part of the esophagus [2]. It has been shown [3], for instance, that resection of the esophagus in its thoracic part causes trophic disturbances of the acinar parenchyma and insular apparatus in the pancreas, accompanied by infiltration of the stroma by polymorphonuclear leukocytes and lymphocytes. This may be connected with injury to small branches of the vagus nerve during the operation, for we know that the paraventricular-vagal mechanism plays an important role in regulation of the endocrine function of the pancreas [1].

The aim of this investigation was to study carbohydrate metabolism and the morphological structure of the pancreas following injury and subsequent repair of a subtotal defect of the wall of the cervical part of the esophagus.

EXPERIMENTAL METHOD

The blood sugar level and the morphological structure of the pancreatic insular apparatus were investigated in experiments on 40 young and middle-aged mongrel dogs. After resection of two-thirds of the circumference of the esophagus for a length of 4-5 cm the operation was concluded by replacement of the subtotal defect in the wall of the cervical part of the esophagus by means of artificial prostheses. The blood sugar concentration was studied by the glucose oxidase method using the "Ames Dextrostix" express system. Glucose loading was at the rate of 1.75 g glucose/kg body weight. Material for histological investigation was taken strictly from definite areas of the tail and body of the pancreas, and the pieces were fixed in 12% neutral formalin solution and embedded in paraffin wax. Sections 5-7 μ thick were stained with hematoxylin and eosin and with aldehyde-fuchsine by the Gabe–Dyban method. The number of islets was counted morphometrically in 100 fields of vision. The investigation was carried out before the operation and on the 3rd, 7th, 14th, 21st, 30th, 40th, 60th, 120th, and 180th days thereafter. Three intact dogs on which access to the cervical part of the esophagus was

Department of Operative Surgery and Topographic Anatomy, Department of Pathological Anatomy, and Course in Endocrinology, Smolensk Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR A. P. Avtsyn.) Translated from Byulleten' Eksperimental'noi Biologii i Meditsiny, Vol. 101, No. 3, pp. 367-370, March, 1986. Original article submitted November 20, 1984

 0007-4888/86/0003-0388$12.50