STRUCTURAL CHANGES IN THE TESTIS AFTER TOTAL OR PARTIAL PANCREATECTOMY

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Hormonal regulation of the functions of the testis and its relations with other endocrine glands is not in dispute [6]. Spermatogenesis is indissolubly connected with the general state of the body and is completely dependent on it [5]. The effect of disturbances of different parts of the digestive system and of individual endocrine organs and, in particular, blocking or reduction of the endocrine and exocrine activity of the pancreas, on the state of testicular function has not been investigated.

The reaction of the testes to various forms of stress differs from reactions of other endocrine glands [1-3]. Hence the importance of a study of changes in the testis and its role in the general response of the organism to insufficiency of the external and internal secretion of the pancreas, and the investigation described below was carried out for this purpose.

EXPERIMENTAL METHOD

Experiments were carried out on 54 dogs, and another 10 intact animals served as the control. The dogs undergoing operations were divided into three groups: 1) 19 dogs undergoing resection of one or two terminal portions of the pancreas, equivalent to resection of the tail or tail and body of the human pancreas; 2) 16 dogs undergoing resection of the central part of the pancreas, with ligation and division of the principal efferent ducts; 3) 19 dogs in which the pancreas was totally removed. The operations were performed by the method described previously [4]. The dogs were withdrawn from the experiment after intervals of 3 days to 1 year. Sections through the testis 7 μm thick were stained with hematoxylin and eosin, by the methods of Van Gieson, McManus, Feulgen, and Brachet, by Gomori's sulfidine method, with Sudan III, and with Sudan Black B. The longitudinal and transverse diameters of the convoluted seminiferous tubules, the height of the spermatogenic epithelium, and the thickness of the limiting membrane were measured histometrically, the index of spermatogenesis calculated, and the number of sustentacular cells in one seminiferous tubule and the number of intestitial endocrinocytes per seminiferous tubule was determined. The volume of the nuclei of the interstitial endocrinocytes was calculated separately. The nuclei of these cells were subdivided, on the basis of the logarithm of their volume, into small, medium, and large, and their relative proportions were determined. All numerical parameters were subjected to statistical analysis and compared with the corresponding parameters of testicular structure in intact dogs (Fig. 1a).

EXPERIMENTAL RESULTS

Analysis of the data showed that the changes discovered depended on the type of operation on the pancreas and on the time elapsing after operation. Removal of one or two terminal portions (one to two thirds of the weight of the pancreas) led to minor and inconstant changes in the testis. On the first days some reduction in thickness of the spermatogenic epithelium and partial blockade of spermatogenesis were observed. By 1 month after the operation the convoluted seminiferous tubules of the dogs had reacquired their usual shape. Their longitudinal diameter was 162.3 ± 6.1 μm and their transverse diameter 130.6 ± 5.3 μm. The spermatogenic epithelium was wide, measuring 51.5 ± 2.4 μm. The process of spermatogenesis continued until maturation of spermatozoa, and the index of spermatogenesis was 3.5 points. Swelling and edema of the intertubular interstitial tissue, and plasma inhibition of the vessels walls were observed in sections through the testes. Hemodynamic disturbances

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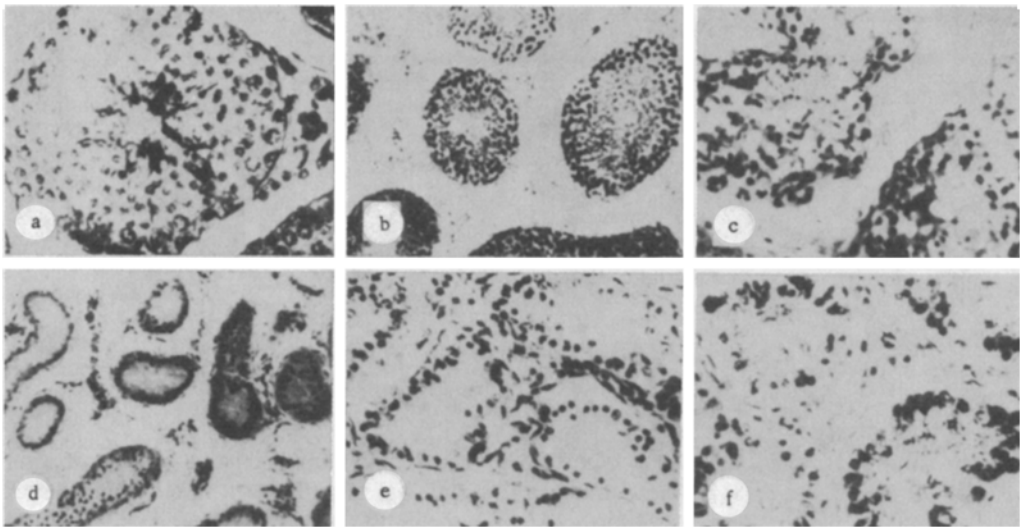

Fig. 1. Dog's testis: a) intact; cell population present in full volume, process of spermatogenesis visible until maturation of spermatozoa; b) 60 days after resection of terminal portions of pancreas: spermatogenic epithelium present as a wide strip, spermatogenesis visible until maturation of spermatids; c) 60th day after resection of central part of pancreas: sex cells chaotically dispersed, cytoplasm of spermatids vacuolated, nuclei of spermatogonia hyperchromic; d) 180 days after resection of central part of pancreas: spermatogenic epithelium present as a dark narrow strip, lysis of nuclei of residual spermatogonia, basically only sustentacular cells can be identified; e) 16 days after pancreatectomy: multiple vacuolation of cytoplasm of sex cells, necrobiotic changes in spermatids, karyolysis of individual spermatogonia; f) 90 days after pancreatectomy with insulin replacement therapy: spermatogenic epithelium separated from limiting membrane, cell population consists almost entirely of sustentacular cells, considerable hyperplasia of intertubular connective tissue. Hematoxylin and eosin. Magnification: a, c) 400 ×, b, d) 140 ×, e, f) 200 ×.

appearing in response to the operation were accompanied by degenerative changes in the spermatids, but spermatogonia of nearly all the seminiferous tubules were well preserved. The steroid-producing structures were indistinguishable from those in the testis of intact dogs. The degree of the hemodynamic and reversible degenerative changes gradually diminished, and signs of compensatory and repair reactions increased. After 4-12 months the structure of the testis was closely similar to that in intact dogs (Fig. 1b).

Although after removal of the central part of the pancreas in dogs, just as after dissection of the head of the human pancreas, the weight of the gland was reduced by only one-third, the operation was accompanied by ligation and division of the principal efferent ducts and by disturbances of digestion. Toward the end of the period of observation the deficiency of body weight amounted to 40% of the weight before the operation, and the weight of the testis was only 50% of the control. The marked reduction in weight of the testes suggests atrophy and depression of function of the testes, and the decrease in body weight [9] was accompanied by disturbances of testosterone production. By the 30th day small, irregularly shaped convoluted seminiferous tubules and dilated intertubular spaces could be observed in the dogs' testis. The connective tissue fibers were loosely arranged. The diamers of the convoluted seminiferous tubules were reduced and measured: longitudinal 140.5 ± 7.7 μm, transverse 112.5 ± 6.9 μm ($P < 0.001$; Fig. 1c). The lumen of the tubules was wider than usually, and they contained leukocytes and conglomerates of desquamated cells. The spermatogenic epithelium was smooth and vacuolated. Its height was reduced to 32.3 ± 2.1 μm ($P < 0.001$). The index of spermatogenesis was 2 points. The number of sustentacular cells was 24.5 ± 0.8 ($P < 0.001$), and some cells with indistinct boundaries were seen. The number of interstitial endocrinocytes was reduced to 2.3 ± 0.02 ($P < 0.001$), but a high percentage (15%) of small nuclei still remained, whereas the numbers of large and medium forms were reduced. The endocrinocytes exhibited marked pseudanophilia, which is regarded as evidence of their adequate level of function. Predominantly focal degenerative disturbances gradually became disseminated after the 3rd month. The dimensions of the convoluted seminiferous tubules were significantly reduced. The capillary lumen was filled with multinuclear cells (spermatids and spermatocytes).

TABLE 1. Histometric Parameters of Testis of Intact and Pancreatectomized Dogs

Group of animals	Time after operation, days	Width of limiting membrane	Linear dimensions, μm			Height of spermatogenic epithelium	Index of spermatogenesis
			diameter of tubules				
			longitudinal	transverse	lumen		
Intact		454,5±9,1	257,2±6,3	179,8±4,2	37,1±6,2	71,4±9,3	3,8±0,04
Pancreatectomized	32	265,3±10,4	167,2±5,9	134,1±5,3	52,4±6,9	41,4±3,2	3,2±0,05
	P	0,001	0,001	0,001		0,003	
	7	264,3±25,1	169,3±9,8	136,2±12,9	66,5±9,7	25,4±2,2	2,2±0,03
	P	0,001	0,001	0,003		0,001	
	13	357,4±28,8	191,2±12,3	134,3±12,3	69,5±11,2	34±1,3	
	P	0,003	0,001	0,002	0,001	0,001	2,4±0,07
	19	453,7±54,5	221,3±10,4	172,3±6,5	65,3±6,4	55,2±5,4	
	P		0,006		0,004	0,002	2,1±0,06
	30	477,2±27,9	136,5±3,9	113,4±6,6	62,8±5,3	29,7±2,5	
	P		0,001	0,001	0,003	0,001	

After 6 months the limiting membrane had become thin. The longitudinal and transverse diameters were 188.4 ± 7.1 and 171.6 ± 4.8 μm respectively. The lumen of the tubules was wide (50.7 ± 7.2) and they were filled with an albuminous mass. The spermatogenic epithelium of most tubules had the appearance of a thin dark band (Fig. 1d). The boundaries of the sex cells were indistinct. In some tubules the remaining sex cells were chaotically dispersed. The height of the spermatogenic epithelium was only 36.5 ± 3.7 μm. In places of massive cell death, an increased content of extracellular glycogen was observed, with redistribution of fat from interstitial cells into spermatogonia. Consequently, after resection of the pancreas and ligation and division of the principal efferent ducts considerable inhibition of the steroid-producing structures gradually developed and changes in the spermatogenic epithelium showed ever-increasing size of irreversible vacuolar and fatty degeneration with an increase in time after the operation. After total pancreatectomy, by the end of only the first week the longitudinal and transverse diameters of the tubules were reduced to 169.3 ± 9.8 and 136.2 ± 12.9 μm respectively. The height of the spermatogenic epithelium was reduced to 25.4 ± 2.2 μm (Table 1). In the capillary lumen there were many desquamated cells, forming conglomerates. The spermatogenic epithelium was separated in places from the limiting membrane, and signs of marked degenerative changes were present in the cells. Some cells were filled with granular cytoplasm and had a hyperchromic nucleus, whereas in others lysis of the nucleus could be seen. The number of sustentacular cells was increased to 23.3 ± 1.3 (Table 2). The cell boundaries were indistinct, their cytoplasm vacuolated, and their stroma highly edematous. Proliferation of adventitial cells was well marked in the vessels. Interstitial endocrinocytes were arranged in groups numbering 3.5 ± 0.1 per seminiferous tubule. The volume of their nuclei was reduced to 101.8 ± 6.1 μm^3. A decrease in the number of small (6.7%) and large (1.7%) forms of nuclei was observed at this time, with a simultaneous increase in the number of medium forms (91.6%) of endocrinocyte nuclei. In the testes of the dogs 1 month after pancreatectomy, the lobules were irregular in shape, and proliferation and fibrosis could be seen in the stroma. The convoluted seminiferous tubules were reduced in size (diameters: longitudinal 136.5 ± 3.9 μm, transverse 113.4 ± 6.6 μm; $P < 0.001$), and varied in shape (Fig. 1e). The capillary lumen was filled with an albuminous mass. The intertubular lymphatic spaces were greatly constricted and the tubules were closely packed together. The limiting membrane was thickened. The spermatogenic epithelium consisted of single spermatogonia, and its height was 29.7 ± 2.5 μm. The number of subtentacular cells was increased to 27.2 ± 1.5 ($P < 0.001$). Some irregularly shaped endocrinocytes could be seen, and many cells had a small nucleus (25%). There was a decrease in number of the principal androgen producers, namely medium (73.3%) and large (1.7%) endocrinocyte nuclei. A sharp decrease in the volume of these cells was characteristic (to 86.9 ± 6.2 μm^3). In general necrotic changes were observed in the spermatogonia with total destruction of the convoluted seminiferous tubules.

From the 3rd day after the operation four dogs were given insulin and their blood glucose level was monitored. The survival period of these animals was considerably increased. After 3 months, sections of the testes revealed considerable proliferation of young connective tissue, which compressed and deformed the seminiferous tubules (Fig. 1f). The longitudinal and transverse diameters were reduced to 104.3 ± 4.2 and 71.2 ± 2.6 μm respectively. The limiting membrane was thickened. The cell population of the tubules consisted entirely of sustentacular cells, the number of which was 29.1 ± 1.4 per seminiferous tubule. The nuclei of many sustentacular cells were hyperchromic, and their cytoplasm vacuolated. The lumen of individual capillaries was filled with desquamated spermatocytes and spermatogonia, with small, shrunken nuclei. The number of endocrinocytes was reduced to 2.0 ± 0.01 per seminiferous tubule. The nuclei were irregular in shape and their volume only 63.4 ± 4.6 μm^3. A considerable increase was observed in the number of small (65%), and a decrease in the number of medium (35%) forms of endocrinocytes, and the large forms were completely absent. Fatty degeneration, considerable accumulation of glycogen, a decrease in phosphatase activity, and a decrease in the concentrations

TABLE 2. Principal Data on Sustentacular Cells and Endocrinocytes of Testis of Intact and Pancreatectomized Dogs

Group of animals	Time after operation, days	Number of systentacular cells	Characteristics of interstitial endocrinocytes				
			number of cells per tubule	volume of cell nuclei, μm^3	ratio of cell nuclei (in %) according to logarithm of their volume		
					small	medium	large
Intact		15,3±1,2	4,8±0,2	120,0±9,1	8,5	75	16,5
Pancreatectomized	3	16,4±0,9	3,3±0,1	99,8±8,9	20	70	10
	P		0,001				
	7	23,3±1,3	3,5±0,1	101,8±6,1	6,7	91,6	1,7
	P	0,001	0,001				
	13	20,6±1,3	4,3±0,1	109,5±7,6	10	82,5	7,5
	P	0,004					
	19	21,8±1,3	4,1±0,1	104,3±8,1	10	80	10
	P	0,001	0,001				
	30	27,2±1,5	4,0±0,3	86,9±6,2	21,7	73,3	1,7
	P	0,001		0,001			

of nucleic acid were found. Although death was postponed in the dogs receiving insulin, 3 months after the operation necrobiosis of the epithelium of the seminiferous tubules and germanative aplasia were observed, peritubular fibrosis developed, and the shape, number, and volume of the endocrinocyte nuclei were disturbed.

After resection of the terminal segments of the pancreas, high compensatory potential of the testis was thus preserved and its endocrine function suffered very little. Resection of the head of the pancreas, and even more, total pancreatectomy led to the development of profound and sometimes irreversible changes in the male gonad. Neurovascular and metabolic factors and an endocrine imbalance played a role in their development. Insulin neither completely corrected nor prevented the development of structural changes in the testis. Consequently, these observations do not completely confirm the view that the development of degenerative changes in the testes can be prevented or considerably reduced by administration of insulin [7, 8].

LITERATURE CITED

1. V. V. Endolov, V. F. Sazonov, and A. E. Solov'ev, in: Abstracts of Proceedings of the 9th All-Union Congress of Anatomists, Histologists, and Embryologists [in Russian], Minsk (1981), p. 144.
2. V. V. Zazhaeva, T. E. D'yakova, A. I. Mardal', et al., in: Abstracts of Proceedings of the 9th All-Union Congress of Anatomists, Histologists, and Embryologists [in Russian], Minsk (1981), p. 152.
3. Yu. A. Medvedev and G. A. Vasil'ev, Patol. Fiziol., No. 1, 44 (1972).
4. R. I. Polyak, The Liver and Kidneys after Resection of the Pancreas (Experimental Investigations) [in Russian], Moscow (1969).
5. E. Roosen-Runge, Spermatogenesis in Animals [Russian translation], Moscow (1980).
6. I. V. Stupin, Arkh. Anat., No. 1, 57 (1965).
7. A. Chatterjee, Endocrinologiya (Bucharest), 50, 11 (1966).
8. R. Hunt and W. Donald, Acta Endocrinol. (Copenhagen), 38, 432 (1961).
9. A. Klibanski, J. Beitins, T. Badger, et al., J. Clin. Endocrinol., 53, 258 (1981).