PARTICIPATION OF THE EPITHELIUM OF THE RETE TESTIS IN THE REPARATIVE REGENERATION OF THE TESTIS IN GUINEA PIGS

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In 1911, Kyrle [10] described the reactive proliferation of the epithelium of the rete testis and the appearance of convoluted tubules of small diameter and lined with Sertoli cells near it in the testes of irradiated dogs and also of human patients dying after severe general diseases. Kyrle concluded from his observations that regeneration of the testis may take place from the epithelium of the rete testis, just as the regeneration of certain other glands takes place from the tissue of the ducts. However, in no subsequent investigations of the regeneration of the testis after various injuries [3, 5, 8, 9] was the epithelium of the rete testis indicated as a possible source of its regeneration, and Kyrle's hypothesis was disputed by some authorities but ignored by most.

The present article summarizes results obtained during the study of regeneration of the testis in guinea pigs after partial resection, which can be regarded as confirming Kyrle's views of the possible participation of the epithelium of the rete testis in regeneration of the gland.

EXPERIMENTAL METHOD

The results of several experiments performed on guinea pigs, weighing from 200 to 550 g, for various purposes were analyzed. Altogether 59 animals underwent the operation. In most guinea pigs part of the parenchyma of the left testis was removed from the side of it's free surface, without any major blood vessels, through an incision in the tunica albuginea, which was later sutured. In some animals half the left testis was removed together with the tunica albuginea covering it. The operations were performed with observance of the rules of asepsis under superficial ether anesthesia. The animals were sacrificed 2, 7, 14, and 60 days after the operation. The testes were weighed and fixed in Bouin's or Carnoy's fluid. Paraffin sections cut to a thickness of 7 μ, and in some cases serial sections, were stained with hematoxylin-eosin and with iron hematoxylin by Heidenhain's method.

EXPERIMENTAL RESULTS

The first experiments giving results which suggested participation of the rete testis in the regeneration process were carried out on guinea pigs weighing 300–400 g. Two months after removal of half the left testis followed by suture of the incision in the tunica albuginea, an area was found in the injured testes with the characteristic structure of the embryonic testis, and situated in the region of the hilum close to the intensively proliferating rete testis (Fig. 1, a). These experiments were repeated three times, and altogether 38 animals underwent the operation; in half of them convoluted tubules of immature type were found in the injured testes. The microscopic structure of these testes was very similar. Most of the organ consisted of convoluted tubules 180–200 μ in diameter, bounded by a complex tunica propria, with a fibrous structure, and of elongated cells with rod-shaped nuclei, arranged some distance from each other.

Spermatogonia and nuclei of Sertoli cells lay in the walls of the tubules, and nearer the lumen of the tubules spermatogenic cells could be seen in various stages of spermatogenesis. Near the place of injury, which was easily determined from the remains of the silk ligature in the tunica albuginea, the convoluted tubules were almost completely free from spermatogenic cells. Their diameter varied from 120 to 150 μ, and they were lined by Sertoli cells with a cytoplasm of ragged appearance and by solitary spermatogonia and spermatocytes with pycnotic nuclei. Finally, near the hilum of the testis an area was observed, varying from 1.5 to 3 mm in diameter, consisting of very narrow convoluted

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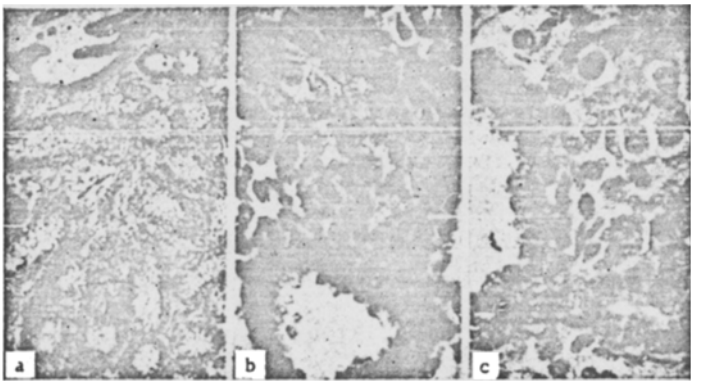

Fig. 1. Testis of a sexually mature guinea pig two months after partial resection. a) Immature convoluted tubules near the reactively modified rete testis. Hematoxylin-eosin. Magnification 100×; b) proliferation of the epithelium of the rete testis with the formation of a projecting process in the center of which may be seen typical follicular cells. Iron hematoxylin. Magnification 500×; c) Pattern of formation of genital cords: primary sex cells surrounded by follicular cells (indicated by an arrow), appear close to the proliferating rete testis. Iron hematoxylin. Magnification 500 ×.

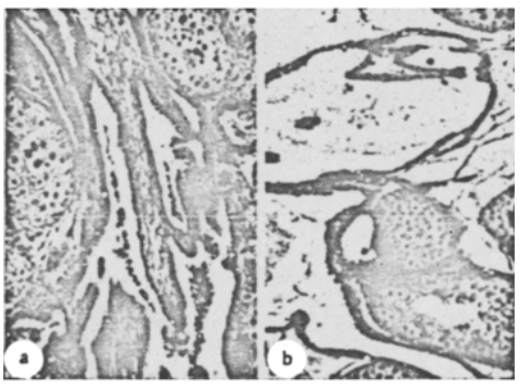

Fig. 2. a) Reactive changes in the rete testis 2 days after operation. Hematoxylin-eosin. Magnification 100×; b) rete testis of the intact testis of the same guinea pig: low, flattened epithelium. Hematoxylin-eosin. Magnification 100×.

tubules with a diameter of 50-60 μ, not yet possesing a lumen and separated from the surrounding tissue by a well marked tunica propria, possessing the typical cellular structure of immature tubules [11]. The spermatogonia formed a single continuous layer in the tunica propria of these tubules, whereas the nuclei of the Sertoli cells, varied in shape and containing one or two comparatively small nucleoli, were situated above the spermatogonia. At the periphery of the area of immature tissue, tubules of larger diameter (100-200 μ) were seen, containing many spermatocytes and providing for the gradual transformation from tissue of embryonic type to tubules of normal structure. Consequently, two months after the operation, three types of convoluted tubules were found in the injured testes, differing in their localization, their structure, and their morphogenetic potential.

In every case near the immature seminiferous tubules, characteristic changes were observed in the rete testis, for its epithelium, normally low, cubical and considerably flattened, had a high, cylindrical appearance, and in certain areas it was stratified (Fig. 1, b and c). It formed processes, buds, and bands of cells which, because of their oval nucleus with one or two clearly outlined nucleoli, were very reminescent of the follicular cells or immature Sertoli cells. These processes of the rete in some cases continued as typical genital cords, without a lumen or tunica propria, consisting of follicular cells and primary sex cells with a large nucleus and clear cytoplasm.

The changes described above in the rete testis were evidence supporting the view that its epithelium plays an active part in the formation of genital cords and convoluted tubules by transformation of the rete cells into follicular cells, although they did not explain the origin of the primary sex cells in the fully formed testis of sexually mature animals. To discover their origin, experiments were carried out in which the animals were sacrificed at various times after the operation. These showed that on the 2nd day after the operation the epithelium of the rete testis over a wide area, but more especially in the region where it leaves the testis, assumed the characteristic appearance of the rete of the immature organ (Fig. 2, a),

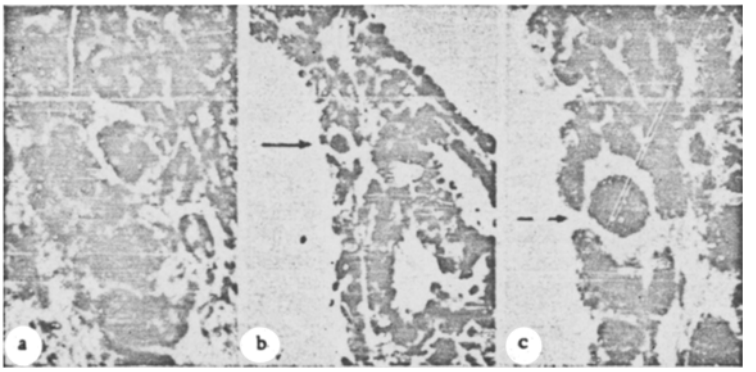

Fig. 3. a) Mitosis in the epithelium of the rete testis one week after operation. Hematoxylin-eosin. Magnification 800×; b) a primary sex cell lying in the epithelium of the rete testis 7 days after operation (indicated by an arrow). Hematoxylin-eosin. Magnification 360×; c) the same primary sex cell. Magnification 800×.

and differed sharply from the flattened epithelium of the rete of the intact testis of the same guinea pig (Fig. 2, b). One week after the operation, near the rete of the injured testis with its reactive changes, genital cords and solitary immature narrow seminiferous tubules could be seen. In the epithelium of the rete mitoses were observed (Fig. 3, a) but were absent in the rete of the intact testis [12].

A particularly interesting finding was that the epithelium of the rete testis one week after the operation contained primary sex cells, one of which is shown in Fig. 3. b and c. Such primary sex cells were also found in the connective tissue surrounding the rete of the uninjured testis of, not only the young, but also of the sexually mature guinea pigs. Two weeks after the operation the general picture of the microscopic changes in the intact testis was the same as at the previous period, but there were more immature tubules. In 10 guinea pigs sacrificed two weeks after removal of part of the testis together with the tunica albuginea, in all cases an area of testicular tissue was formed with a nearly embryonic structure; this area was seen particularly clearly against the background of complete devastation of the convoluted tubules in the rest of the organ.

The results described show that in guinea pigs, in response to partial resection of the testis, the epithelium of the rete testis undergoes characteristic reactive changes expressed by some degree of dedifferentiation, cellular proliferation, and the formation of genital cords. As a whole, these changes evidently cause the appearance of convoluted tubules of embryonic type in the testes of the sexually mature animals. The facts concerning the origin and development of the rete testis demonstrate the perfect feasibility of its participation in the process of formation of the convoluted tubules in the injured testis. Roosen-Runge [13] showed in an extensive and important investigation that the intratesticular part of the rete testis in rats develops from the genital cords which cease to grow and which gradually lose their sex cells, although these latter are sometimes preserved in the rete for a long time after birth. It is interesting that in postnatal oogenesis, described fully by V. Ya. Karmysheva [6], in the normal and regenerating testis of adult cats the oogonia differentiate from the primordial epithelium, mainly in the region of the hilum, as in the case of the injured testis, and the microscopic picture of formation of the follicles is very similar to the picture of formation of the genital cords described above.

It is difficult at present to say to what degree the newly formed immature convoluted tubules are capable of further development and differentiation in adult life, i.e., whether or not they are capable of restoring the part of the parenchyma of the testis lost as a result of injury. The author's observations show that in cases when tubules were formed with the participation of the rete, the initial weight of the organ was not restored even two months after the operation, whereas, on the other hand, in experiments in which one testis was removed completely and the other partly, followed by suture of the tunica albuginea, the weight of the testis was restored after one month, but in this case no reactive changes were observed in the epithelium of the rete testis and tubules of embryonic type were not formed [7]. The impression was created that the character of the regenerative processes in the injured testis was determined by the

hormonal balance, in general agreement with expressed views [2] concerning the existence of an inverse relationship between the formation of new structural units in the injured organ and the amount of functioning tissue removed.

The formation of tubules of embryonic type in the injured testis of sexually mature animals is important not only for assessment of the regenerative power of the testis, but also for understanding the mechanism of development of tumors, frequently arising in the testis after injury [1, 4].

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