Blood-Testis Barrier after Transplantation of Pituitary Cell into Dystopic Testicle

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A total of 87 allotransplantations of the pituitary were carried out: into intact testicle with or without subsequent transposition into the abdominal cavity and on a previously created model of unilateral cryptorchidism. Histological study showed that pituitary cells retained viability for up to 3 months in all three series. The main structures of the blood-testis barrier (membranes of testicular tubules, tunica albuginea testis, vascular walls, and interstitial tissue) were not damaged, which determined advantages of dystopic testicle in pituitary cell allotransplantation.

Key Words: *testicle; hematotesticular barrier; pituitary; transplantation; rats; cryptorchidism*

Barrier properties of the testicle depends on the blood-testis barrier (BTB) formed from vascular walls, membrane of testicular tubules, tunica albuginea testis, and interstitial tissue [4].

The number of fibroblast-like cells in the BTB structures, e.g. in the tunica albuginea testis decreases with aging, but these age-specific changes do not notably affect the permeability of BTB [7].

Another important component of BTB is interstitial tissue consisting mainly of Leydig cells (LC) producing testosterone. LC are resistant to ischemia [2]. LC retain their functional activity for up to 14 days when cultured in appropriate nutrient medium [1], and after allotransplantation into rat testicle remain viable for up to 6 months [3].

Interstitial endocrinocytes (spermatogenic epithelial cells) produce hormones mediating paracrine and autocrine regulation [2,6,7].

Apart from hormones, the gonads secrete neurotrophins, the best known of which are glial-cellular purifying neurotrophic factor and neurotourine affecting cell differentiation in testicles [8,9]. All neurotrophic factors affect all metabolic processes in the gonads.

Testicular atrophy in various andrological diseases (including cryptorchidism) is caused by deficiency of neurotrophine regulating the synthesis of germinative cells.

Pronounced morphological changes develop in the dystopic testicle during experimental cryptorchidism [5]. The structure of seminal tubules is impaired primarily because of changes in spermatogenic epithelial cells, which constantly die, this leading to gonadal atrophy. The membrane of testicular tubules and blood vessels architectonics in the testicle are not changed in cryptorchidism. The number of LC is not decreased in dystopic testicle, but the cells can be hypertrophic [5]. It remains unclear whether immune response changes and whether dystopic testicle retains its advantages after transplantation of pituitary cells. We found no reports answering these questions. Here we studied the state of BTB after allotransplantation of pituitary cells into dystopic testicle.

MATERIALS AND METHODS

Experiments were carried out on 87 adult rats. The pituitary for allotransplantation was taken from adult rats. After opening the skull, the pituitary was re-

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moved from the sella turcica and prepared; adenohypophysis was washed in Hanks' solution and minced with ophthalmic scissors in Eurocollins medium. Pituitary cells were transplanted under the tunica albuginea testis.

The testicle and BTB after allotransplantation of pituitary cells into dystopic testicle were studied in 3 experimental series. In the control (n=39), allogenic pituitary cells were transplanted under the tunica albuginea testis of orthotopic testicles. In series II (n=25) the testicle with transplanted pituitary cells was placed into the abdominal cavity and fixed to the parietal peritoneum. In series III (n=23), allogenic pituitary cells were transplanted under tunica albuginea testis of dystopic testicle 1 month after modeling of unilateral cryptorchidism.

In all series pituitary cells were transplanted using microsurgical technique. Suspension of allogenic pituitary cell through a 1-mm incision in tunica albuginea, was introduced into special pouch created using a kit of microbougies. The tunica albuginea was sutured with 8/0-9/0 atraumatic needle.

No immunosuppressive therapy was used. The results of transplantation were evaluated 1 and 2 weeks and 1 and 3 months after surgery. Histological preparations were stained with hematoxylin and eosin, by Mallory's and Van Gieson's methods, and using periodic acid-Schiff reaction.

RESULTS

In the control, pituitary cells were found under the tunica albuginea testis at all terms of observation. The structure of the adjacent seminal tubules remained unchanged (Fig. 1) and spermatogenesis was normal at all terms. No lymphoid infiltration was observed. Allogenic pituitary cells were seen under the tunica albuginea and were viable at all terms.

One week after allotransplantation and transposition of the testicle into the abdominal cavity pituitary cells were loosely distributed under the tunica albuginea. Their nuclei were clearly differentiated, no lymphoid infiltration was observed. After 2 weeks allo-

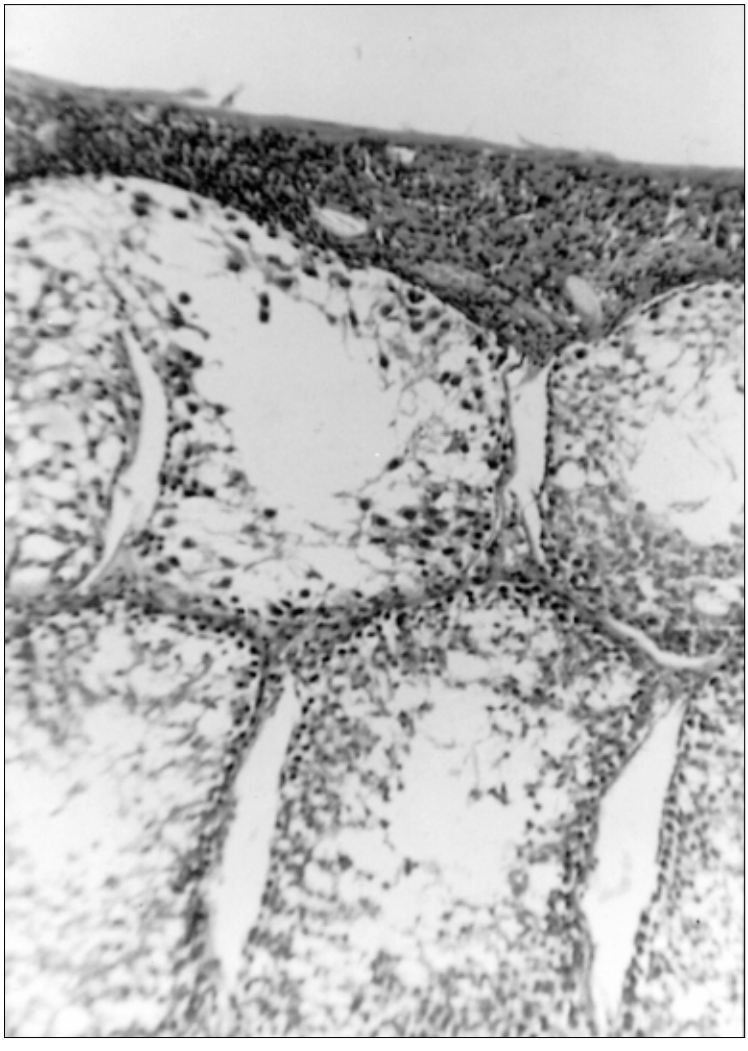

Fig. 1. Rat testicle 2 weeks after allotransplantation of pituitary cells to testicle. Hematoxylin-eosin staining, ×80. Allogenic pituitary cells lie compactly under the tunica albuginea. Seminal tubules are intact. Spermatogenic epithelial cells are intact.

genic pituitary cells lay more compactly. The testicular capsule was thickened. Spermatogenesis in seminal tubules was impaired: 1st-2nd order spermatocytes were present. Seminal tubule membranes were intact, no lymphoid infiltration was seen. One-three months after transplantation the testicle shrank in size, its parenchyma was loosened. Histological examination showed that testicular capsule was thickened. Allogenic pituitary cells were in a satisfactory state, their nuclei were clearly differentiated, small capillaries grew between pituitary cells (Fig. 2). Pronounced changes in seminal tubules were noted: supporting cells and Sertoli cells were seen. Seminal tubule membrane was intact. No spermatogenesis was observed. LC were viable at all terms.

One month after cryptorchidism modeling, the testicle shrank in size and became loose. Histological analysis showed supporting cells and solitary Sertoli cells in the seminal tubules. The shape of seminal tubules was irregular, most often oval; their membranes were unchanged.

One week after pituitary transplantation to rats with experimental cryptorchidism, allogenic pituitary cells lay under the tunica albuginea, their nuclei were clearly differentiated, no signs of lymphoid infiltration were seen, seminal tubules were oval-shaped and contained supporting and solitary Sertoli cells (Fig. 3). Later pituitary cells more compactly lay under the tunica albuginea, vascularization of the transplant was seen. Seminal tubules still contained supporting cells and solitary Sertoli cells; the area occupied by LC increased in comparison with seminal tubule cells.

High concentrations of steroid hormones and neurotrophins in the testicle maintained mitosis, which strengthened BTB and maintained the privileged position of the testicle as a site for endocrine cell and tissue transplantation.

The structures of the testicle, in particular, spermatogenic epithelium, are highly sensitive to temperature rise, which manifests by inhibition of neurotourine synthesis and impaired production of spermato-

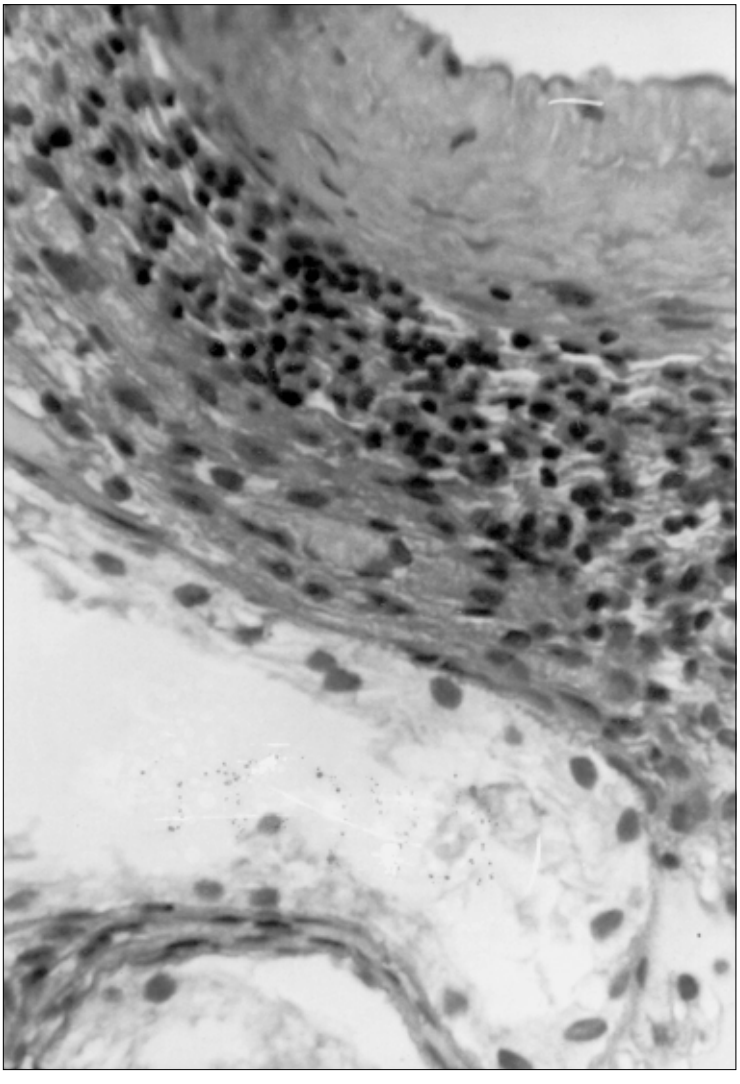

Fig. 2. Rat testicle after allotransplantation of pituitary cells and its transposition into abdominal cavity. Hematoxylin-eosin staining, ×320. Pituitary cells and numerous blood capillaries under thickened tunica albuginea. Solitary supporting cells and Sertoli cells in seminal tubules.

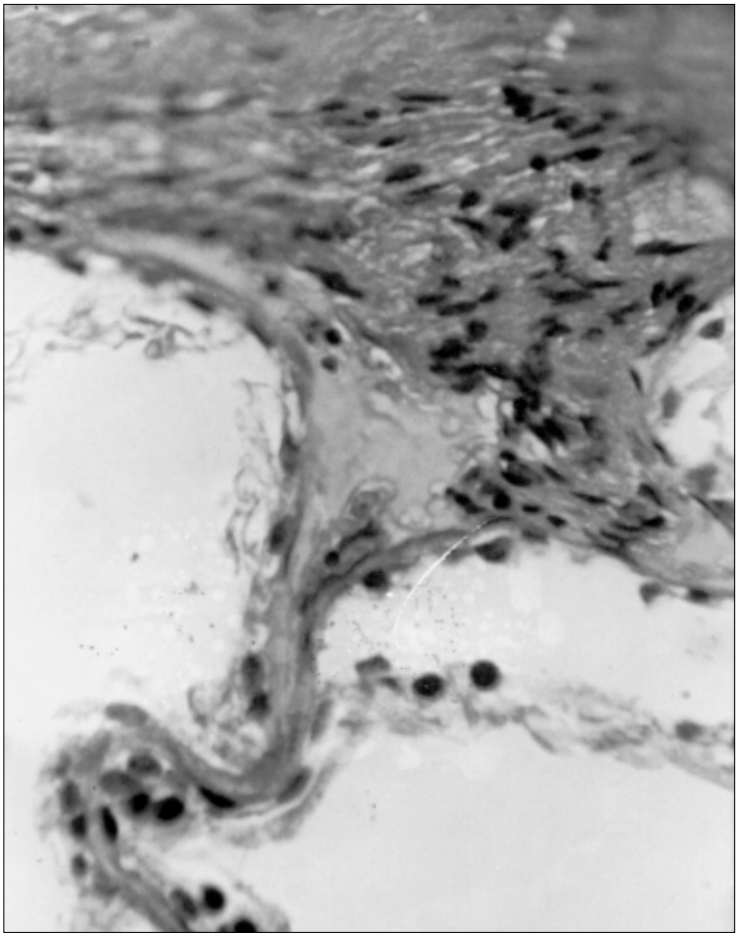

Fig. 3. Allogenic pituitary cells 1 week after allotransplantation to testicle in rat with experimental cryptorchidism, ×320. Edema and swelling at the site of transplantation. Moderate plethora of blood capillaries. Tunica albuginea testis is edematous. Seminal tubules adjacent to the transplant are intact, their shape is abnormal. Spermatogenic epithelium with focal destruction.

genic epithelial cells. However the bulk of neurotrophins functioned satisfactorily maintaining mitoses in BTB cells.

Hence, allotransplantation of pituitary cells (tissue) to the testicle in animals with experimental cryptorchidism damages the spermatogenic epithelium but spares the major structures of BTB: tunica albuginea, seminal tubules membranes, supporting cells, and capillary walls. LC maintaining high concentrations of hormones in the interstitium are preserved, which promotes survival of allogenic pituitary cells without immunosuppressive therapy.

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