Torsion of the Spermatic Cord – A Long Term Study of the Contralateral Testis

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Summary. The object of the present investigation was to study the long-term effects of the unilateral torsion of the spermatic cord on the contralateral testis. Eighteen guinea pigs were divided into 3 groups. In group I of six animals, unilateral torsion of the spermatic cords was maintained until the time of sacrifice. In group II of six animals, torsion of the spermatic cords was maintained for 8-12 h, then the spermatic cords were untwisted and the animals were maintained until the day of sacrifice. Group III six animals, received an injection of pentobarbital, which served as control. All animals were sacrificed after 16 months. Extensive light and electron microscopic studies were carried out. In the contralateral testes of the experimental group of animals, several degenerative changes were noted, which included excessive intraepithelial vacuolization, a loss of germ cells and the presence of tubules containing only Sertoli cells and a few spermatogonia. 10.6% and 19.5% seminiferous tubules were damaged in the contralateral testes of torsion maintained and the torsion reversed groups of animals, respectively in comparison to 3.1% tubular damage (indicated only by occassional presence of intraepithelial vacuoles and necrotic germ cells), in the control testis. It was concluded that long-term effect of unilateral torsion of the spermatic cord is permanent and irreversible in nature.

Key words: Testis, Torsion, Spermatic cord, Germ cells, Seminiferous tubules.

Introduction

We have previously reported our studies on the effects of unilateral torsion of the spermatic cord on the contralateral testes of guinea pigs [2-4, 7]. Those studies were based on our observations using the animals, from 1 week to 8 months after surgical induction of unilateral torsion of the spermatic cord.

During the present investigation, we have further extended our studies to examine the effects of unilateral torsion of the spermatic cord on the contralateral testis in animals which had undergone unilateral torsion of the spermatic cord, 16 months previously. The objective of the present project was to evaluate whether the damage to the contralateral testis, as a result of the unilateral torsion of the spermatic cord was transitory and reversible or permanent and irreversible in nature.

Materials and Methods

Eighteen male Hartley strain guinea pigs weighing 600-700 g were used for the present study. All animals were maintained in the animal facility with controlled temperature and light and dark cycle. Animals were divided into three equal groups. In Group I and Group II of 12 animals, unilateral torsion of the spermatic cord was surgically induced by twisting the spermatic cord through 540° [2], under pentobarbital anesthesia and in sterile conditions. In 6 out of these 12 animals, unilateral torsion of the spermatic cord was maintained until the day of sacrifice. Orchiopexy was not done to avoid additional trauma to the testis. This group was designated as Group I or torsion maintained (TM) group of animals. In each of the remaining 6 animals, spermatic cord was untwisted 8 to 12 hours after the induction of unilateral torsion. This group was designated as Group II or torsion untwist (TU) group of animals. The rest of the 6 animals received an intraperitoneal injection of pentobarbitol. This group served as control and designated as Group III animals.

All animals were sacrificed 16 months after the experimental procedure. At the time of autopsy, spermatic cords of each animal were examined carefully. All animals of torsion maintained group (Group I) retained the spermatic cord torsion on the experimental side. Each testis was removed and divided into 5 parts. One piece of tissue from each part, i.e. a total number of five pieces of tissues from different regions of each testis were fixed with 2.5% glutaral-dehyde by immersion and were post fixed in 1% O_sO_4 . The fixed tissue specimens were then processed for epon embedding using a routine procedure [2]. 1 μ m semithin sections were cut with a LKB ultramicrotome, stained with 1% toluidine blue and examined under a light microscope. 90–100 nm thin sections were used for the electron microscopic observation. A Philips EM300 electron microscope was used for that purpose.

Table 1. Percentage of the affected seminiferous tubules and the tubular diameter in the contralateral testes of guinea pigs after longterm unilateral spermatic cord torsion

Experimental group	% of the affected tubules ^a	Tubule diameter (µm)
Control	3.1 ± 1.1 (0-6) ^b	341.7 ± 4.8
Torsion-maintained	$10.6 \pm 3.1^{\circ}$ $(5-22)^{\circ}$	345.0 ± 6.8
Torsion-untwist	19.5 ± 5.2^{d} $(7-38)^{b}$	334.8 ± 6.2

a Values are given as mean ± SEM

Estimation of Seminiferous Tubular Damage

1 μ m semithin sections stained with toluidine blue were used for estimation of the damage to the seminiferous tubules in the affected as well as in the contralateral and control testicular tissue samples. For each animal, at least 40–50 sections of seminiferous tubules were studied, with a total number of 240–250 tubule sections for each group of animals. The following criteria were used to determine the extent of damage in the seminiferous tubules: i) pronounced intraepithelial vacuoles; ii) presence of necrotic germ cells; iii) loss of germ cells, i.e., tubules containing some spermatogonia, spermatocytes and Sertoli cells; iv) severe germ cell hypoplasia, i.e. tubules without any type of germ cells except the spermatogonia.

Seminiferous tubules showing any one or more than one of the above criteria were considered as affected tubules. The number of affected tubules for each group of animals was determined and the percentage of the affected tubules was calculated (Table 1). The diameters of the seminiferous tubules were directly measured under the light microscope, using an ocular micrometer fitted to a 20x objective lens. At least 10 randomly cut round seminiferous tubules were used to obtain the seminiferous tubular diameter [5, 7].

Results

Sixteen months after torsion of the spermatic cord, the contralateral testis of guinea pigs showed various signs of degenerative change in the seminiferous tubules. These degenerative changes included clusters of vacuoles within the seminiferous epithelium. When vacuoles were detected in the control specimen, their appearance was different than those in experimental groups (Group II and III). In the experimental groups, the vacuoles occurred in small or large clusters with occassional presence of large vacuoles from the basal to the apical area of the tubules (Figs. 1 and 2). Whereas in the control animals, the majority of tubules were compact (Fig. 3) with occassional presence of small, isolated vacuoles. Moderate to severe loss of germ cells leading to the vacuolated, single layered germinal epithelium (Figs. 4 and 5) were other characteristics of the contralateral testis of the experimental group of animal. This type of moderate to severe loss of germ cells was never detected in the control testis. Many necrotic germ cells (Fig. 6) were present in the affected tubules of the contralaterial testis of the experimental group of animals. A greater number (19.5%) of seminiferous tubules were affected in the contralateral testis of the torsion untwist group in comparison to the torsion maintained group (10.6%). Control animals always had fewer (3.1%) damaged seminiferous tubules indicated only by the occassional presence of small vacuoles or isolated necrotic germ cells (Table 1). No recovery was noticed in the contralateral testis of both torsion maintained and torsion untwist groups of animals, 16 months after the torsion of the spermatic cord.

Discussion

It has now been well established that unilateral torsion of the spermatic cord has a detrimental effect on the contralateral testis [1–9]. These detrimental effects range from the exfoliation of large number of germ cells leading to the vacuolization in the seminiferous epithelium to the complete absence of developing and differentiating germ cells. However, it is not known whether these contralateral changes are transitory and reversible or permanent and irreversible in nature.

10.6% and 19.5% (Table 1) of the seminiferous tubules were damaged in the contralateral testis of the torsion maintained and torsion untwist groups, in comparison to 3.1% abnormal seminiferous tubules in the control group. Besides, this 3.1% abnormal seminiferous tubules were comprised of epithelial vacuolization and occassional presence of necrotic germ cells only. Other types of damage i.e. many necrotic cells, loss of layers of germ cells, Sertoli cell only type of tubule were never seen in the control specimens. Another characteristic was that the contralateral testis of the untwist group were significantly more damaged (19.5%) in comparison to the torsion maintained (10.6%) group. The reason for this remains unknown at this time. However, it should be noted that although the spermatic cord remained twisted in the torsion maintained group of animals, variable degree of damage was noted in the testes of the experimental side. In fact, the testes of the experimental side of the torsion untwist group (Group II), showed more damage than the ones in the torsion maintained group (Group I). This excess damage in the testis of the torsion untwist group (Group II) may be the result of two successive operations at 8-12 h intervals. Therefore, it can be speculated that in the torsion maintained group, the germ cell degeneration in the contralateral testis was less, related to the variable degree of damage in the testis of the experimental side [2, 3]. Since the torsion untwist group (Group II) of animals had more damage to the ipsilateral testis, a comparatively larger number of seminiferous tubules were affected in the contralateral testis.

No significant change was noticed in the diameter of the seminiferous tubules (Table 1) in the contralateral testes of these three groups of animals. Therefore, semini-

b range

c P < 0.05

d P < 0.02 (Student's "t" test)

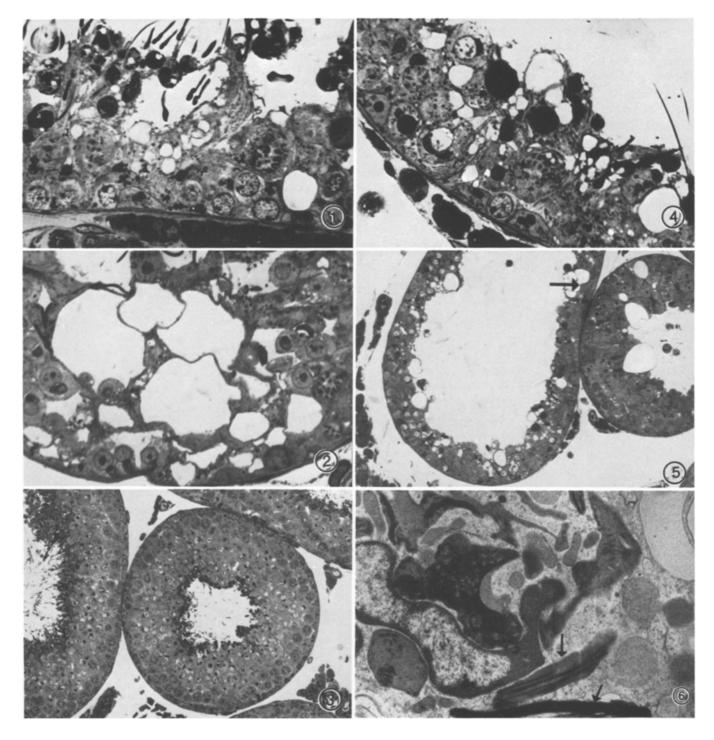


Fig. 1. A portion of an affected seminiferous tubule from a guinea pig of the experimental group of animals, showing vacuolization of the seminiferous epithelium. x1,000

Fig. 2. Another area of the seminiferous tubule of the same animal as in Fig. 1, showing the nature of small clusters of vacuoles in the epithelium. x1,000

- Fig. 3. Control specimen, showing compact seminiferous tubule, without vacuolization. $\times 200$
- Fig. 4. Moderately damaged seminiferous tubule, showing the loss of germ cells, especially the loss of developing spermatids. Only a few spermatids can be seen in this tubule. x1,000
- Fig. 5. Low magnification picture of the same seminiferous tubule of the same animal as in Fig. 4. Note the absence of germ cells (arrow) from some areas of this tubule, showing the severely damaged portions of the same seminiferous tubule. x200

Fig. 6. A representative electron micrograph showing the nature of degenerative germ cells in clusters. Remnants of spermatid head and tail (arrows) can still be identified in this figure. ×15,000

ferous tubular diameter should not be considered as one of the criteria to determine the extent of damage in the contralateral testis due to unilateral torsion of the spermatic cords in guinea pigs.

In conclusion, the present study indicates that in guinea pigs the effect of unilateral torsion of the spermatic cord on the contralateral testis is permanent and irreversible in nature even, 16 months after the induction of torsion. Further, retention of a highly damaged testis in the body is harmful to the contralateral testis and contralateral testicular damage may be related to the extent of damage in the ipsilateral testis.

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