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# EVALUATION OF THE CONTRALATERAL TESTIS AFTER UNILATERAL ORCHIDECTOMY BY LACTATE DEHYDROGENASE-C4 ISOENZYME ACTIVITY IN RATS

RATLARDA UNİLATERAL ORKİDEKTOMİ SONRASI KONTRALATERAL TESTİSİN LAKTAT DEHİDROGENAZ-C4 İZOENZİMİ İLE DEĞERLENDİRİLMESİ

Fatma TANELİ Ece ONUR

Ahmet VAR Bekir Sami UYANIK

Cevval ULMAN Can TANELİ

Department of Biochemistry, Faculty of Medicine ,Celal Bayar University, Manisa

Cüneyt GÜNŞAR<sup>2</sup> Zeki ARI

Department of Pediatric Surgery, Faculty of Medicine, Celal Bayar University, Manisa

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### SUMMARY

There are numerous experimental studies on contralateral testicular damage induced by torsion, varicocele and vas deference obstruction. In the present study, we aimed to investigate contralateral testicular function after unilateral orchidectomy. The study comprised 20 Wistar Albino, male rats, 200-250 g in weight. In ten rats unilateral orchidectomy was performed while the remaining were Sham operated and taken as controls. After a period of 30 days, rats were sacrificed and all testes were harvested. Testicular function was evaluated with lactate dehydragenase-C4 (LDH-C4) isoenzyme in the testicular tissue. Testicular LDH-C4 isoenzyme was assessed by agarous gel electrophoresis method. LDH-C4 isoenzyme activities were found significantly (p<0.05) decreased in the study group (7.42±6.69 vs 10.07±6.92 IU/mg wet tissue) compared with Sham controls. In conclusion, LDH-C4 isoenzyme could be a sensitive biochemical parameter showing the injury formed in the contralateral testis. The present experimental study showed that in unilateral orchidectomy the biochemical alterations were demonstrated in the contralateral testis at one month postoperatively. In a standard orchidectomy, spermatic cord ligation, vas deference obstruction, even spermatic cord torsion are inevitable; thus, during orchidectomy operations unintentional twisting of the spermatic cord, accordingly, does not seem to be appropriate.

## ÖZET

Torsiyon, varikosel, ve vas deferens obstruksiyonuna bağlı kontralateral testisdeki akut hasarı inceleyen bir çok araştırma yapılmıştır. Çalışmamızda, tek taraflı orşidektomi operasyonu sonrası kontralateral testiküler fonksiyonunun uzun dönem sonuçlarını incelemeyi amaçladık. Çalışmamızda 20 adet 200-250 gr ağırlığında Wistar albino erkek rat kullanılmıştır. On rat'a unilateral orşidektomi uygulanmış, 10 rat'a ise Sham operasyonu yapılarak control grubu olarak kullanılmıştır. 30 gün sonra ratlar sakrifiye edilmiş ve testisleri çıkarılmıştır. Testiküler fonksiyon testis dokusunda laktat dehidrogenaz izoenzimi aracılığıyla değerlendirilmiştir. Testiküler LDH-C4 izoenzimi agaroz jel elektroforez metoduyla analiz edilmiştir. LDH-C4 izoenzim aktiviteleri çalışma grubunda Sham kontroluna göre anlamlı(p<0.05) bulunmuştur.(7.42±6.69 vs 10.07±6.92 IU/mg yaş doku). Sonuç olarak, LDH-C4 izoenzimin kontralateral hasarı değerlendirebilecek hassas bir biyokimyasal parameter olabileceği sonucuna varılmıştır. Çalışmamızda unilateral orkidektomi operasyonundan 30 gün sonra kontralateral testisde biyokimyasal değişiklikler olduğu gösterilmiştir.

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## INTRODUCTION

Standard orkidektomi operasyonunda, spermatic kordun bağlanması, vas deferens obstruksiyonu ve hatta spermatic kord torsiyonu kaçınılmazdır. Bu nedenle orkidektomi operasyonu sırasında kontrlateral testiste hasara yol açmamak için özellikle spermatic kordu ekseni etrafında döndürmekten kaçınılması uygun olacaktır. Damage of the affected testis is known to cause deterioration not only in the ipsilateral but also in the contralateral testis. Unilateral incarcerated inguinal hernia, testicular torsion, varicocele and undescended testis are examples of causes of contralateral deterioration reported in literature (1-6). Electromagnetic, radioisotopic blood flow measurements in the testicular tissue after unilateral testicular torsion causes a decrease in the contralateral testicular blood flow and it is postulated that hypoxia produced by a decrease in the blood flow might be the cause of the contralateral testicular damage (7,8). It is reported that the presence of ipsilateral testicular tissue is not mandatory for the contralateral changes after unilateral testicular torsion (9, 10). However, the mechanism which causes these changes in the contralateral testis remains obscure. Lactate dehydrogenase isoenzyme C4 (LDH-C4) has been assayed as a well-known parameter of testicular function (11-13). LDH-C4 is the specific isoenzyme of Lactate Dehydrogenase (LDH) produced by germ cells and is involved in the energy metabolism of spermatozoa (14, 15). Thus, in the present study, we planned to investigate the possible effect of unilateral orchidectomy on the contralateral testicular function with LDH-C4 isoenzyme.

#### MATERIALS AND METHODS

The study comprised 20 Wistar Albino, male rats, 200-250 g in weight. The rats were housed in a temperature and light controlled environment with ad libitum access to

water and rat pellet for three days before the study. All experimental procedures were approved by the Experimental Animal Research Ethics Committee of the Faculty of Medicine. The surgical procedure was performed under general anesthesia administered by intramuscular injection of ketamine and xylasine hydrochloride, employing sterile technique. In ten rats unilateral orchidectomy was performed while the remaing rats were Sham operated and taken as controls. After 30 days, rats were sacrificed and all testes were harvested. Tunica albuginae were stripped off and all tissue were homogenised (IKA T25, England) at a cold milieu in a phosphate buffer solution of pH 8.8 and centrifuged. The testicular tissue total LDH activity were assessed by enzymatic methods on autoanalyzer (Integra Roche Diagnostics Corporation, Indianapolis, USA). The isoenzymes of LDH were separated by agarous gel electrophoresis method by a commercial kit (Sebia Hydrage kit, Sebia Hyrys analyser, France) on the same day. Data were expressed as mean±standard deviation. Statistical analysis was performed using Student-t test and a value of p<0.05 was considered to be significant.

#### RESULTS

LDH-C4 isoenzyme activities were found 7.42  $\pm$  6.69 IU/mg wet tissue in the orchidectomy group and 10.07  $\pm$  6.92 IU/mg wet tissue in the Sham control group. The values are given as mean $\pm$ standard deviation. A significant decrease (p<0.05) was found in the quantitative LDH-C4 activities of the contralateral testis of the orchidectomy group when compared to Sham operated control group. The well-known testicular parameter LDH-C4 isoenzyme which monitors testicular function, demonstrated a statistical difference between the orchidectomy and the Sham control group as shown in Figure 1.

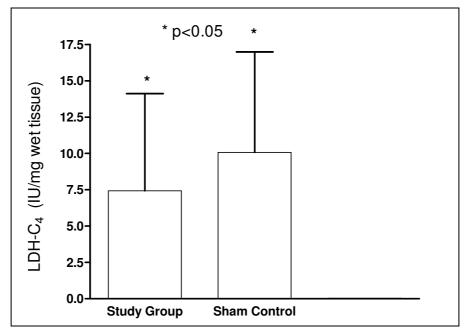


Figure 1. Testicular tissue LDH-C4 isoenzyme activities in the orchidectomy and Sham control groups.

### DISCUSSION

Unilateral spermatic cord torsions are accepted to damage both the ipsilateral and the contralateral testes, and diminish spermatogenesis (6-8). Although early reports proposed auto-immunization as the mechanism of contralateral testicular injury; recently, it is thought that autoimmune mechanism does not play a role in contralateral testicular damage (9, 10, 16). Akgür et al (9), suggested that unilateral spermatic cord torsion both in the presence and absence of the ipsilateral testicular tissue ends with increases of lactic acid, hypoxanthine and thiobarbituric acid reactive products (TBARs) of lipid peroxidation in the contralateral testis, thus they reported that the presence of the ipsilateral testis is not mandatory for the ischemic injury of the contralateral testis. They also, mentioned that testis does not produce a substancetrigger-pathway affecting contralateral testicular blood flow, however, they thought that the impulse-triggering contralateral changes might arise from spermatic vessels or nerves (9). Karagüzel et al (10), suggested that in the spermatic cord torsion model, the presence of ipsilateral testicular tissue seems not to be essential in causing the contralateral testicular histological deterioration, but seems to augment the damage. In contrast to our study, they stated that orchidectomy does not cause any adverse effects on the contralateral testis by way of the mean seminiferous tubular diameters and the mean testicular biopsy scores. The results of the above mentioned studies point to the fact that the testicular torsion, more specifically the torsion of spermatic cord, is the pathologic condition that renders the contralateral testicular damage.

Altay et al (16), suggested that spermatic cord ligation or vassal ligation only, cause a marked increase in the hydrostatic pressure and oedema of both the ipsilateral and the contralateral testes. Savas et al (17), showed that similar electron microscopic findings observed in the torsion and the ligation (spermatic cord ligation including vas deference) groups indicate that testicular ischemia is responsible for the ultrastructural changes on the contralateral side. Savas et al (18) studied acute (48 hours) oxidative stress mechanism on experimental unilateral orchidectomy in rats. They reported that the oxidative marker malondialdehyde, was significantly (p<0.05) reduced in the contralateral testis, and antioxidant enzymes such as superoxide dismutase and catalase were also significantly (p<0.05) decreased; whereas glutathione peroxidase was significantly (p<0.05) increased in the unilateral orchidectomy. These 48-hourresults of unilateral orchidectomy coincide with our 30day-results.

The point of view is that, in spermatic cord ligation or torsion, the contralateral damage is shown to be independent of the presence of the ipsilateral testis. In a standard orchidectomy, spermatic cord ligation, vas deference obstruction, even spermatic cord torsion is inevitable; thus, we are of the opinion that, it is not surprising to observe measurable alterations in the contralateral testis after unilateral orchidectomy. Nevertheless, there still remain some points to be enlightened, e.g., whether the biochemical alterations would be permanent or not, what would be the long term effect of these results on testicular function and how long would they last, need to be elucidated by further studies.

### CONCLUSION

The present experimental study showed that in unilateral orchidectomy, the biochemical alterations occur in the contralateral testis one month postoperatively. We are of the opinion that LDH-C4 isoenzyme could be a sensitive biochemical parameter showing the deteriorations formed in the contralateral testis. Thus, during orchidectomy operations unintentional twisting of the spermatic cord, accordingly, does not seem to be appropriate.

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