Testicular growth, sperm concentration, percent motility, and pregnancy outcome after varicocelectomy based on testicular histology

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Objective: To evaluate the outcome following surgery in two different patterns of testicular histology in patients with varicocele.
Design: Prospective study.
Setting: Academic medical center.
Patient(s): Sixty patients underwent varicocelectomy and had a testicular histology diagnosis of germ-cell aplasia (group I, n = 28) or maturation arrest (group II, n = 32).
Intervention(s): Varicocelectomy.
Main Outcome Measure(s): Preoperative hormone levels, testicular size, and sperm parameters. After varicocelectomy, variations in testicular size, semen parameters, and pregnancy rates were evaluated.
Result(s): The mean volume of the right and left testicle was smaller in group I patients than in group II. In addition, the mean sperm concentration before treatment was lower in group I than in group II. The mean volume of the left and right testicle increased in group I after the intervention. The mean postoperative sperm concentration and motility in group II showed no increase, whereas the mean sperm concentration in group I did increase. The pregnancy rate was higher in group II (14/26, 53.8%) than in group I (4/16, 25%) (P=.02).
Conclusion(s): Patients with germ-cell aplasia present a postoperative increase in testicular size but the pregnancy rate is higher in patients with maturation arrest following surgery. (Fertil Steril® 2005;83:362–6. ©2005 by American Society for Reproductive Medicine.)

Key Words: Testis, semen, varicocele, infertility, histology

Infertility may be the only clinical feature of several disorders which are accompanied by some level of spermatogenesis damage or impairment of sperm delivery (1). Male infertility treatment is certainly better carried out if the reasons for it are understood. Although the association between male infertility and varicoceles has been known for the last century (1–8), varicocele is only complementary to most forms of male infertility (2–3).

It is well known that induction of left varicocele in the rat and rabbit results in bilateral testicular damage (4, 5). Semen quality uniformly declines in animals with induced varicoceles, even when only a left varicocele is produced (6). However, a major problem resides in the fact that varicoceles can have a highly variable influence on testicular function, leaving it apparently unaltered in some cases but causing a partial or total arrest of spermatogenesis and/or deficient androgen production in others (6, 10–14).

Progressive deterioration of the Sertoli cells may occur as a result of varicocele, leading to vacuolization and the release of the spermatogenic cells before their maturation is complete (15–18). In addition, Sertoli cell secretory dysfunction is detected in patients with varicocele. Intratesticular androgen binding protein concentrations in subjects with varicoceles are significantly smaller than in controls (5). Subjects with varicocele have Leydig cell secretory dysfunction, and intratesticular testosterone profiles are significantly smaller than in controls (19). In addition, secretory deficiency of Leydig cells in animals with varicocele has been demonstrated (20). That well designed experimental study demonstrated an inhibition of testicular testosterone biosynthesis after experimental varicocele in rats. The inhibition of testosterone synthesis in rats with surgically induced varicoceles was shown to be due mainly to a reduction in the activity of the enzyme 17,20-desmolase (20).

Although the pathogenesis of the varicocele remains enigmatic, gross testicular alterations associated with varicocele are well documented (14). The effects of the varicocele vary but may often result in a generalized impairment of sperm

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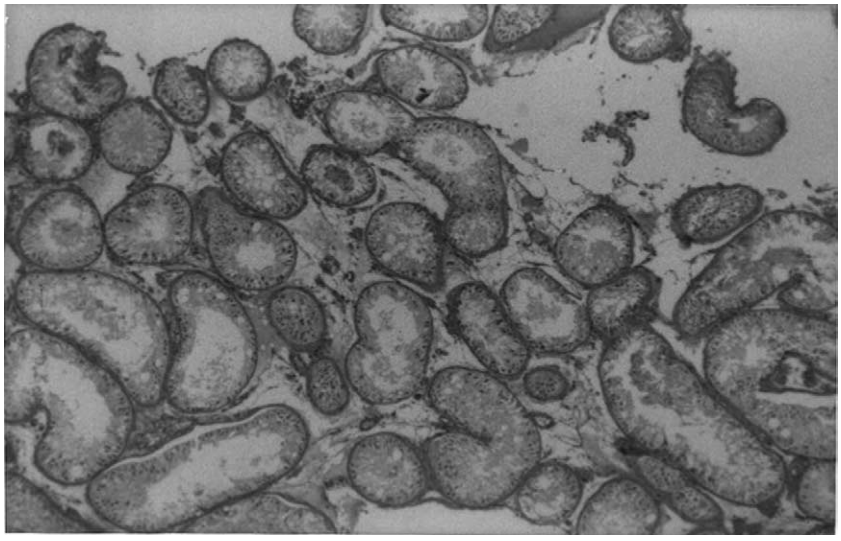
FIGURE 1

Histology showing germ-cell aplasia.



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production, characterized by abnormal semen quality ranging from oligospermia to complete azoospermia. Furthermore, the varicocele affects not only the physiology and the reproductive potential of the spermatozoa but also the fertilizing capacity of the haploid male gamete (21).

The degree of histologic impairment seems to be independent of the clinical stage of the varicocele (11, 16, 17). In general, testicular derangement is more pronounced on the side of the varicocele, though the difference between the two sides disappears as the histologic impairment increases (14). Clinical evidence suggests that spermatogenesis may vary within the same side in a damaged or failing testis, resulting in focal areas or "patches" of sperm production within an organ largely devoid of germinative cells (22). Therefore, it is assumed that an infertile man with low sperm count might have testicular damage reflected by abnormal histology patterns such as germ-cell aplasia or maturation arrest before becoming azoospermic.

The purpose of this study was to evaluate two patterns of testicular histology, namely germ-cell aplasia and maturation arrest and the treatment outcome following varicocele repair. This information could be used to inform our patients weather a varicocelectomy performed in a man with a specific testicular histology pattern may result in better sperm parameters or pregnancy rates compared to others.

MATERIALS AND METHODS

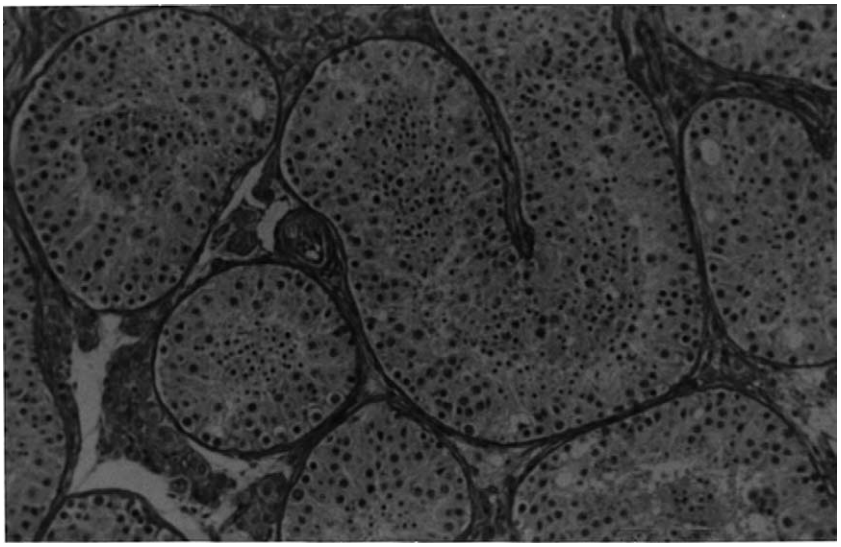
Our study was approved by our Institution Review Board, and all patients granted informed consent. From July 1999 to November 2001, 107 patients underwent varicocelectomy. All of them had primary infertility. All women were normal, based on history, hormonal levels, and hysterosalpingogram. Infertility was defined by failure to establish pregnancy within 1 year of unprotected intercourse. A basic infertility evaluation including a detailed history and a complete physical examination was undertaken. Testicular size was evaluated in all patients with a caliper. Only patients with varicocele identified on physical examination were included. Patients who were taking antioxidants such as vitamins C and E were excluded from the study.

From the 107 operated men, 78 underwent open diagnostic testis biopsy at the same time as varicocele repair under general anesthesia. Forty-three of the 78 biopsied men had a left varicocelectomy performed, and 35 had bilateral varicocelectomies performed. Biopsies were undertaken on both testes, irrespective of which appeared healthier by size or consistency. Testicular biopsy was performed in these patients only for research purposes. Of these, 60 had a testicular biopsy showing germ-cell aplasia or maturation arrest (at the primary spermatocyte stage) and only 18 men had a diagnosis of normal spermatogenesis or hypospermatogenesis. Therefore, patients were divided into 2 groups according to the testicular histology pattern: group I (germ-cell aplasia, n = 28) and group II (early maturation arrest, n = 32) (Figs. 1 and 2). No differences were detected in the age of the wives between patients in group I (32.2 ± 4.1) and group II (31.7 ± 3.7) (P=.09).

All patients were assessed for testicular size, serum testosterone, LH, and FSH levels. Two semen analyses were obtained by masturbation after 2 to 5 days of abstinence before the operation. If the difference between the 2 semen analyses was higher than 10%, a third one was requested.

FIGURE 2

Histology showing maturation arrest.



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Sperm concentration and motility were evaluated according to the World Health Organization criteria. Patients with pyospermia were treated before varicocele repair. All patients were treated with the microsurgical, subinguinal approach. Using the microsurgical approach, we ligated the pampiniform plexus and the cremasteric veins, leaving the gubernaculum veins intact.

Preoperative hormone levels, testicular size, and sperm parameters were compared between the two groups. After varicocelectomy, the variations in testicular size, sperm parameters, and pregnancy rates were compared between the two groups. Values are presented as the mean ± SD, with Student's *t*-test and the Wilcoxon paired test used for statistical evaluation. A χ^2 test was used in order to compare pregnancy rates between groups I and II. A value of $P<.05$ was considered statistically significant. Statistical calculations were performed with computer software (Statistical Package for Social Sciences, version 10.0; SPSS, Chicago, IL).

RESULTS

No differences were seen between the two groups in the mean age, mean grade of varicocele, and levels of LH, FSH, and testosterone ($P>.05$) (Table 1). However, the mean volume of the right testicle was smaller in group I patients (16.62 ± 1.87 mL) than in group II (20.37 ± 8.02 mL) ($P=.02$). Also, the mean volume of the left testicle was smaller in group I patients (13.62 ± 1.91 mL) than in group II (19.46 ± 9.65 mL) ($P=.01$). The mean sperm concentration before treatment was lower in group I ($9.82 \pm 5.9 \times 10^6$) than in group II ($19.97 \pm 22.34 \times 10^6$) ($P=.03$). No differences were seen in sperm motility before treatment (44.3 ± 25.6% vs. 38.5 ± 23.1%) ($P>.05$). Pregnancy data were available for 16 patients in group I and for 26 in group II.

Although no increase in testicular volume was seen in group II (right, 20.3 ± 6.9 mL; left, 20.5 ± 6.8 mL), the mean volume of the right testicles of group I increased (19.6 ± 9.1 mL; $P=.02$) as did that of the left testicles (16.02 ± 4.8 mL; $P=.03$). The mean postoperative sperm concentration ($24.76 \pm 16.9 \times 10^6$) and motility (42.4 ± 22.6%) in group II showed no increase, whereas the mean sperm concentration in group I did increase ($12.05 \pm 4.8 \times 10^6$; $P=.04$). The pregnancy rate was higher in group II (14/26; 53.8%) than in group I (4/16; 25%) ($P=.02$) (Table 1).

DISCUSSION

The effect of varicocelectomy on testicular histology was well documented by Johnsen and Agger (15) who compared preoperative and postoperative testes biopsies in patients with varicoceles who were either fertile or infertile and showed that the infertile group presented a significant increase in the number of seminiferous tubules containing numerous spermatozoa after varicocelectomy. Other investigators also have demonstrated that gross and microscopic alterations are associated with the presence of clinical varicoceles (1–3, 6–13, 15–17). It is, therefore, important to know whether a particular testicular histology may predict which patients will benefit from surgical treatment.

Few studies have shown that nonobstructive azoospermic patients with varicocele identified on physical examination

TABLE 1

Age, hormonal levels, testicular size, and sperm concentration and motility before and after surgery in patients with germ-cell aplasia (group I) and with maturation arrest (group II).

Variables	Group I	Group II	P^a	P^b	P^c
Female age	32.2 ± 4.1	31.7 ± 3.7	.09		
Male age	35.5 ± 6.1	34.9 ± 6.1	.09		
FSH (mIU/mL)	7.8 ± 7.6	6.9 ± 6.9	.08		
LH (mIU/mL)	4.3 ± 1.9	4.7 ± 2.1	.08		
Testosterone (ng/dL)	540.36 ± 94.5	590.72 ± 112.6	.08		
Left testicle (mL) (before surgery)	13.62 ± 1.91	19.46 ± 9.65	.01		
Right testicle (mL) (before surgery)	16.62 ± 1.87	20.37 ± 8.02	.02		
Left testicle (mL) (after surgery)	16.02 ± 4.8	20.5 ± 6.8	.04	.03	.08
Right testicle (mL) (after surgery)	19.6 ± 9.1	20.3 ± 6.9	.07	.02	.09
Sperm concentration ($\times 10^6$/mL) (before surgery)	9.82 ± 5.9	19.97 ± 22.34	.03		
Sperm motility (%) (before surgery)	44.3 ± 25.6	38.5 ± 23.1	.07		
Sperm concentration ($\times 10^6$/mL) (after surgery)	12.05 ± 4.8	24.76 ± 16.9	.06	.04	.08
Sperm motility (%) (after surgery)	41.3 ± 19.6	42.4 ± 22.6	.09	.09	.08
Pregnancy rate (%)	25%	53.8%	.02		

Note: P values <.05 in bold type were considered significant.
[a]Comparisons between the two groups.
[b]Comparisons of variables before and after surgery in group I.
[c]Comparisons of variables before and after surgery in group II.

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may benefit from varicocele repair (2, 23, 24). The few studies published on completely azoospermic men have consistently shown improvement of semen parameters in up to 50% and of spontaneous pregnancies after microsurgical inguinal varicocelectomy (2, 23, 24). Kim et al., studying 28 patients, demonstrated that testicular histopathology was the most important predictive factor of outcome (24). They conclude that for patients with germ-cell aplasia pattern and maturation arrest at the spermatocyte stage, varicocelectomy does not improve semen quality. However, 50% of the completely azoospermic men with maturation arrest at the spermatid stage and 55.6% of completely azoospermic men with hypospermatogenesis achieved postoperative improvement in semen quality (24). Recently, it was demonstrated that azoospermic patients who presented germ cell aplasia in a single large testis biopsy may have an improvement in semen quality following varicocelectomy (2). In addition, due to the great possibility of their relapsing into azoospermia after an initial improvement in semen quality following varicocelectomy, patients should be informed of the possibility of sperm cryopreservation (2).

Interestingly, in our series of patients, both testicular volumes in patients with germ cell aplasia increased, whereas the same increase was not seen in patients with maturation arrest. In addition, sperm concentration increased only in patients with germ cell aplasia. One of the possible reasons for this contrast is that the varicocele is the physiopathologic mechanism associated with infertility in the group of patients with germ cell aplasia but not in those with maturation arrest.

Of utmost importance was the higher pregnancy rates seen in the wives of patients with maturation arrest when compared to those with germ cell aplasia. Even though no increase in sperm concentration or percent motility was seen in patients with maturation arrest, more than 50% of the respective wives became pregnant after the varicocelectomy. A recent study has shown that in infertile couples undergoing intrauterine insemination whose respective female evaluation was normal, pregnancy and live birth rates were significantly higher if the man had varicocele treatment (25). The authors conclude that men should be screened for varicocele before intrauterine insemination for male factor infertility is initiated, because a functional factor not measured in routine semen analysis may affect pregnancy rates in this setting. In our opinion, the same principle might be applied here in our study. In other words, even if a varicocelectomy does not improve conventional semen parameters, it may improve some biochemical or molecular patterns which might be why patients with maturation arrest have a high pregnancy rate without improvement in sperm concentration or motility.

We can therefore conclude that patients with germ-cell aplasia present an increase in testicular size following varicocele repair, whereas those with maturation arrest do not.

However, the pregnancy rate was higher for those patients with maturation arrest than in those with germ-cell aplasia.

REFERENCES

1. Sharlip ID, Jarow JP, Belker AM, Lipshultz LI, Sigman M, Thomas AJ, et al. Best Practice Policy Committee of the American Urological Association, INC. Fertil Steril 2002;77(5):873–8.
2. Pasqualotto FF, Lucon AM, Hallak J, Saldanha LB, Góes PM, Arap S. Induction of spermatogenesis in azoospermic men undergoing varicocele repair. Hum Reprod 2003;18:108–12.
3. Naughthon C, Nangia A, Agarwal A. Pathophysiology of varicoceles in male infertility. Hum Reprod Update 2002;7:473–81.
4. Saypol DC, Howards SS, Turner TT, Miller ED Jr. Influence of surgically induced varicocele on testicular blood flow, temperature, and histology in adult rats and dogs. J Clin Invest 1981;68:39–45.
5. Sofikitis N, Miyagawa I. Effects of surgical repair of experimental left varicocele on testicular temperature, spermatogenesis, endocrine function, and fertility in rabbits. Arch Androl 1992;29:163–75.
6. Sofikitis N, Miyagawa I. Bilateral effect of varicocele on testicular metabolism in the rat. Int J Fertil 1994;39:239–47.
7. Jarow JP. Effects of varicocele on male fertility. Hum Reprod Update 2001:7(1):59–64.
8. Cozzolino DJ, and Lipshultz LI. Varicocele as a progressive lesion: positive effect of varicocele repair. Hum Reprod Update 2001;17:55–8.
9. Schlesinger MH, Wilets IF, Nagler HM. Treatment outcome after varicocelectomy. Urol Clin North Am 1994;21:517–29.
10. Steckel J, Dicker AP, and Goldstein M. Relationship between varicocele size and response to varicocelectomy. J Urol 1993;149:769–71.
11. Redmon JB, Carey P, and Pryor JL. Varicocele—the most common cause of male factor infertility. Hum Reprod Update 2002;8(1):53–8.
12. Witt MA, Lipshultz LI. Varicocele: a progressive or static lesion? Urology 1993;42:541–3.
13. Gorelick J, Goldstein M. Loss of fertility in men with varicocele. Fertil Steril 1993;59:613–6.
14. Madgar I, Weissenberg R, Lunenfeld B, Karasik A, Goldwasser B. Controlled trial of high spermatic vein ligation for varicocele in infertile men. Fertil Steril 1995;63:120–4.
15. Johnsen SG, Agger P. Quantitative evaluation of testicular biopsies before and after operation for varicocele. Fertil Steril 1978;29: 58–63.
16. Zini A, Buckspan M, Jamal M, and Jarvi K. Effect of varicocelectomy on the abnormal retention of residual cytoplasm by human spermatozoa. Hum Reprod 1999;14:1791–3.
17. Heinz HA, Voggenthaler J, Weissbach L. Histological findings in testes with varicocele during childhood and their therapeutic consequences. Eur J Pediatr 1980;133:139–46.
18. Bettochi C, Parkinson MC, Ralph DJ, and Pryor JP. Clinical aspects associated with Sertoli-cell-only histology. BJU 1998;82:534–7.
19. Su LM, Goldstein M, Schlegel PN. The effect of varicocelectomy on serum testosterone levels in infertile men with varicoceles. J Urol 1995;154:1752–5.
20. Rajfer J, Turner TT, Rivera F, Howards SS, Sikka SC. Inhibition of testicular testosterone biosynthesis following experimental varicocele in rats. Biol Reprod 1987;36:933–7.
21. Sofikitis NV, Miyagawa I, Incze P, Andrighetti S. Detrimental effect of left varicocele on the reproductive capacity of the early haploid male gamete. J Urol 1996;156:267–70.
22. Turek PJ, Cha I, Ljung, B-M. Systematic fine-needle aspiration of the testis: correlation to biopsy and results of organ "mapping" for mature sperm in azoospermic men. Urology 1997;49:743–8.
23. Matthews GJ, Matthews ED, Goldstein M. Induction of spermatogenesis and achievement of pregnancy after microsurgical varicocelectomy in men with azoospermia and severe oligoasthenospermia. Fertil Steril 1998;70:71–5.
24. Kim ED, Leibman BB, Grinblat DM, Lipshultz LI. Varicocele repair improves semen parameters in azoospermic men with spermatogenic failure. J Urol 1999;162:737–40.
25. Daitch JA, Bedaiwy MA, Pasqualotto EB, Hendin BN, Hallak J, Falcone T, et al. Varicocelectomy improves Intrauterine Insemination success rates in men with varicocele. J Urol 2001;165:1510–13.