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Vasectomy reversal using a microsurgical three-layer technique: one surgeon's experience over 18 years with 1300 patients

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Summary

The technique and the results of microsurgical vasectomy reversal in a single-centre study over 18 years are presented. Both vasovasostomy (VV) and epididymovasostomy (EV) were carried out in a three-layer technique. With strict adherence to the strategy, end-to-end VV was only performed if spermatozoa had been demonstrated at the epididymal stump of the vas. In all other cases, EV was carried out in a preocclusive region of the epididymal tubule. The outpatient procedure of refertilization was associated with a very low complication rate, which underlines its minimal-invasive character. The follow-up rate was 71%, the overall patency rate was 89% and the pregnancy rate was 59%. Secondary azoospermia was only observed in 1% of the patients. In relation to the intervals of obstruction, the patency and pregnancy rates were higher after short-term obstruction than after long-term obstruction. Correspondingly, higher success rates were found after VV than after EV. This is understandable because the probability for indication of EV increases with longer periods of obstruction. There is a significant discrepancy between patency and pregnancy rates that is likely to be caused by a relevant number of patients with postoperative asthenozoospermia. The duration of obstruction is an important factor concerning epididymal damage, but it only disproportionately affects the results of refertilization if the technology of EV is implemented consistently in case of an epididymal granuloma. Good clinical results are achieved with this strategy, as evidenced by pregnancy rates and semen analyses.

Introduction

Obstructive azoospermia (OA) is a possible cause of male infertility. OA is mainly caused by vasectomy. The therapy of OA requires microsurgical refertilization (MR) or intracytoplasmic sperm injection (ICSI) with epididymal or testicular spermatozoa.

In cases of obstructive azoospermia after vasectomy, MR is performed by end-to-end or side-to-end anastomosis between vas and vas or vas and epididymis. A one- or two-layer technique is generally used for anastomosis. We consequently applied a microsurgical three-layer technique for the end-to-end and side-to-end anastomoses and present this technique and our results over 18 years in terms of semen analyses and clinical outcomes, such as rates of patency and pregnancy.

Materials and methods

All microsurgical interventions were carried out on an outpatient basis under general anaesthesia. A single shot of Ciprofloxacin 500 mg or Cefuroxim 500 mg was given perioperatively. The use of an operating microscope was obligatory in all cases. Through a surgical approach of two lateral scrotal incisions (only in a few cases of inguinal approach because of inguinal vasectomy) both scrotal cavities are explored. The tunica vaginalis is only opened when epididymal surgery is carried out. The further operative strategy consists in attempting an end-to-end vasovasostomy (VV) whenever possible (see below). If there is no sperm outflow from the epididymal stump of the vas (which is mainly the case after long obstructive intervals) an end-to-side anastomosis between vas and epididymis is

required [epididymovasostomy (EV)]. Both procedures are carried out using a three-layer technique. The wound is closed with self-dissolving sutures and a pressure dressing is applied for 1 day.

Intraoperative strategy

At first both ligated stumps of the vas deferens are identified, prepared and trimmed. If liquid comes out from the epididymal stump, there is apparently no additional obstruction in the epididymis, caused by the formation of an epididymal granuloma. The fluid gushing out of the vas deferens is examined intraoperatively by microscopic analysis for the presence of spermatozoa and its viscosity. If spermatozoa are demonstrated, VV is realizable. Sperm motility and morphology is of minor importance for the further surgical strategy according to the authors own experience and the literature (Belker *et al.*, 1991).

In addition to the presence of spermatozoa, low viscosity of the fluid is a positive prognostic factor for the outcome of the procedure. (Belker *et al.*, 1991; Silber & Grotjan, 2004; Schlegel & Margreiter, 2007; Hinz *et al.*, 2009).

If the fluid has a toothpaste like consistence, normally no or only a few fragments of spermatozoa are found. In this case, as in the case of missing epididymal fluid, an anastomosis at the epididymal stump of the vas deferens does not make sense – a view that is largely non-controversial (Silber & Grotjan, 2004; Parekattil *et al.*, 2005; Schlegel & Margreiter, 2007; Hinz *et al.*, 2009; Nagler & Jund, 2009). Instead, an EV between pre-occlusive epididymal tubule and abdominal stump of the vas deferens should be carried out. If spermatozoa cannot be demonstrated only in case of water clear fluid from the epididymal stump, it is indicated to carry out a VV.

Patency of the inguinal stump of the vas deferens is checked by injection of 3 mL saline solution.

Operative technique of vasovasostomy

The anastomosis is performed with an end-to-end technique. An absolute precondition for a successful anastomosis is the possibility of preparing both stumps of the vas deferens without any tension, so that they can be fixed in an approximator.

At first the interior (mucosal) layer is sutured with 10–12 non-absorbable single-armed 10-0 stitches with a round needle. So many stitches are necessary to compensate for the different lumina of both vasal stumps, to ensure a conical lumen at the point of anastomosis and to avoid a step-like intraluminal formation and any shifting of the mucosal layer. This adaptation of the different lumina is crucial for subsequent patency of the anastomosis. The interior layer is a water tight adaptation of the mucosa, however, without any tensile strength (Figs 1 & 2).

The second layer comprises suturing the muscle walls of both vasal stumps, which have the same diameter despite different lumina, if both stumps were cut in the straight part of the vas deferens. If the vasectomy site is in the convoluted vas deferens very close to the epididymis, the muscular layer of the epididymal duct stump becomes significantly thinner with increasing nearness to the epididymis. About ten 9-0 single stitches are placed with non-absorbable threads. A sharp spatula needle is necessary for optimal passage through the compact muscular layer.

The closer the cut is to the epididymis, the thinner is the muscular layer, so that stitches should not be placed too deeply. The muscularis suture provides tension relief to the fragile internal layer (Fig. 3).

The third layer consists of adventitial connective tissue surrounding the duct. About ten 8-0 stitches are placed, preventing any tensile stress to the internal mucosal layer.

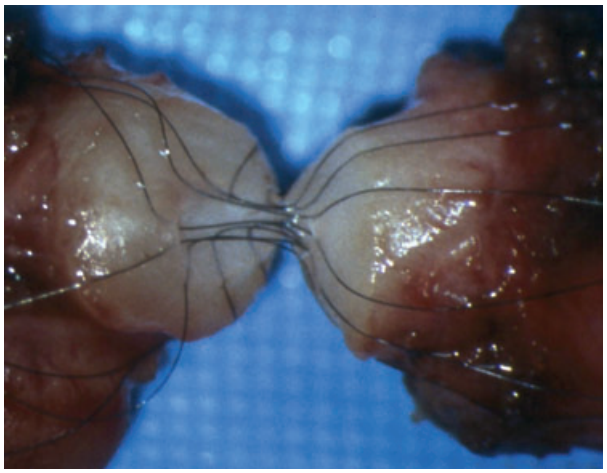

Figure 1 Vasovasostomy: internal layer between the mucosa of both stumps of the vas deferens, typically presenting relevant luminar difference.

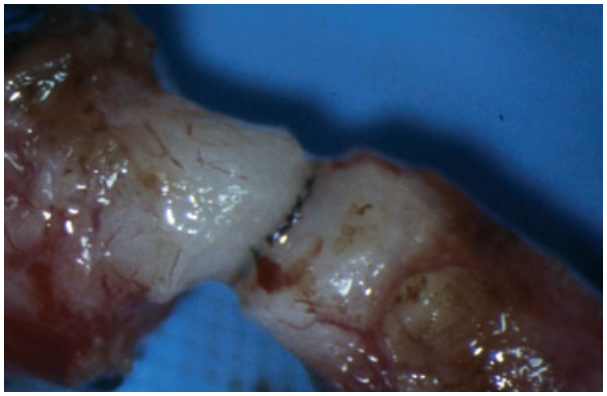

Figure 2 Vasovasostomy: finished internal layer.

When preparing the stump of the epididymal duct it is most important to make sure that the connective tissue layer of the duct is preserved because excessive denudation involves the risk of secondary hypotrophy (Fig. 4).

Operative technique of epididymovasostomy

If there is no outflow or only creamy fluid from the epididymal stump of the vas deferens, the tunica vaginalis must be opened for microsurgical exploration of the epididymis. The strategy consists in looking for the duct obstruction which in most cases is located in the cauda epididymis.

The pre-occlusive epididymal duct can be identified under the microscope. Then the dilated pre-occlusive tubule is tangentially incised in a selective way, which requires a very subtle operating technique. The outflow of epididymal fluid indicates the preocclusive location. The outflowing fluid is analysed by the operating surgeon using a lab microscope, with the aim of demonstrating spermatozoa. If spermatozoa are identified, a side-to-end anastomosis between epididymal tubule and abdominal stump of the vas deferens is carried out in a three-layer technique. Crucial to the outcome is an operative procedure without any tissue tension.

For the internal layer between the wall of the laterally opened epididymal tubule and the mucosa of the vas deferens 8–10 non-absorbable single-armed 10-0 stitches are placed with a round needle.

This internal layer, including the easily tearable structure of the tubular wall, requires 20–30× magnification with the operating microscope as well as extensive microsurgical experience and utmost concentration of the surgeon (Fig. 5).

The second layer is closed between the muscularis of the vas and the epididymal serosa with about ten 9-0 stitches with spatula needle. It provides substantial tension relief to the tearable internal layer (Fig. 6).

Complete tension relief is then achieved by suture of the third layer, which is performed between the adventitia of the vas and the epididymal serosa with about ten 8-0 single stitches (Fig. 7).

For completion of the third layer it is most important that the connective tissue around the vas deferens is well-preserved; excessive denudation should therefore be avoided (see operative technique of VV).

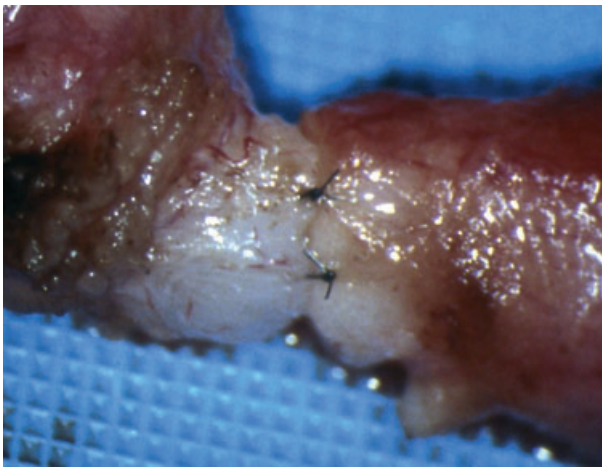

Figure 3 Vasovasostomy: middle layer between the muscular layer of both stumps of the vas deferens.

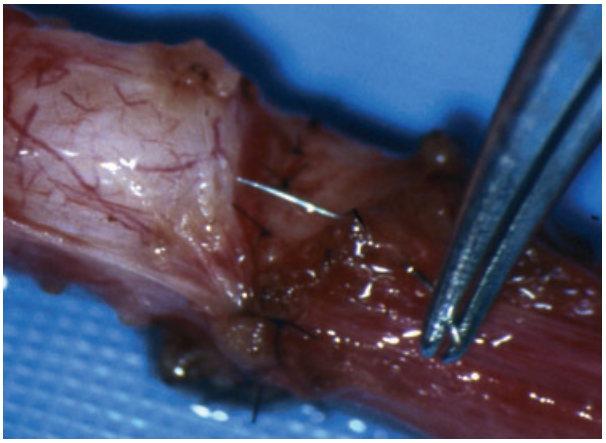

Figure 4 Vasovasostomy: outer layer between the adventitia of both stumps of the vas deferens.

Patients

From 10/93 to 06/11, 1429 patients underwent MR by one surgeon in a single centre for genital microsurgery. Between 1987 and 1993 the author used a two-layer

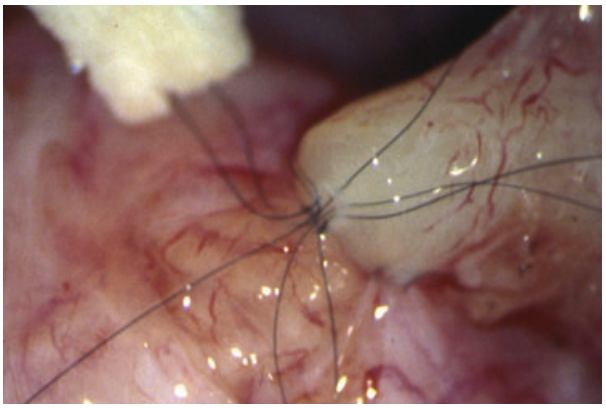

Figure 5 Epididymovasostomy: internal layer between mucosa of the vas deferens and wall of the epididymal tubule.

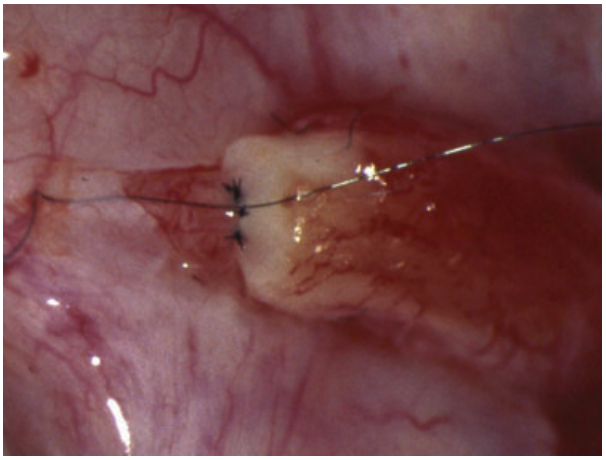

Figure 6 Epididymovasostomy: middle layer between muscular layer of vas deferens and serosal layer of epididymis.

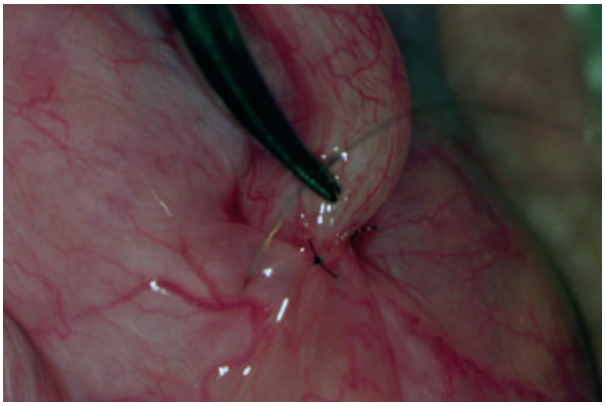

Figure 7 Epididymovasostomy: outer layer between adventitia of vas deferens and serosal layer of epididymis.

technique in several hundred patients, who are not considered in the database and therefore are not included in this article. Also excluded are 126 patients with seminal tract obstruction caused by infection or iatrogenic factors who were operated during the study period. The study thus comprises 1303 patients who underwent vasectomy reversal. Of these, 172 (13.2%) required repeat intervention after a previous attempt of refertilization.

All patients were physically examined with palpation of the scrotum, especially for identification of the vasal stump, and a scrotal sonography.

The age of the patients ranged from 24 to 67 years, with an average of 41 years. The age of the female partners ranged from 21 to 45 years, with an average of 34.6 years.

The periods of obstruction ranged between 18 h and 32 years (average 8.2 years). One patient with 18-h obstructive interval needed immediate reversal because of a non-accepted vasectomy ('agreement error'). The study followed ethical guidelines that are established for human subjects by the Department of Urology of the Technische Universität München.

Results

Perioperative course

Nine-hundred and fifty-eight patients underwent bilateral VV, 214 patients unilateral VV in combination with contralateral EV. Another 36 patients underwent unilateral VV, 84 patients EV bilaterally and 11 patients EV unilaterally (Table 1). So in 24% of the patients EV had to be carried out at least at one side according to our strategy as mentioned above.

The operation time ranged from 90 to 150 min, 110 min on average.

The complication rate was 0.3% ($n = 4$) for scrotal haematoma, only one patient had to be reoperated for evacuation of haematoma. Ten (0.8%) had a superficial wound infection, no case of epididymitis was seen. Apart from two cases of allergic reaction to antibiotics, no side effects or complications were ever seen.

Post-operative course

The follow-up was characterized by special problems, e.g. that many patients changed their place of residence and

Table 1 Vasectomy reversal by microsurgical technique: type of anastomosis in relation to the period of obstruction (total number of patients $n = 1303$): epididymovasostomy at least on one side in 24% of patients

Group no.	Obstruction period (years)	Patients (*n*)	Bilateral vasovasostomy	Vasovasostomy + epididymovasostomy	Bilateral epididymovasostomy	Unilateral vasovasostomy	Unilateral epididymovasostomy
1	<5	312	$n = 268$ (86%)	$n = 31$ (10%)	$n = 6$ (2%)	$n = 7$ (2%)	$n = 0$
2	5–9	527	$n = 399$ (76%)	$n = 85$ (16%)	$n = 22$ (4%)	$n = 16$ (3%)	$n = 5$ (1%)
3	10–15	340	$n = 217$ (64%)	$n = 76$ (22%)	$n = 33$ (10%)	$n = 11$ (3%)	$n = 3$ (1%)
4	>15	124	$n = 74$ (59%)	$n = 22$ (18%)	$n = 23$ (18%)	$n = 2$ (2%)	$n = 3$ (3%)
	Total	1303	$n = 958$ (75%)	$n = 214$ (16%)	$n = 84$ (5%)	$n = 36$ (3%)	$n = 11$ (1%)

Table 2 Vasectomy reversal by microsurgical technique: patency and pregnancy rates in relation to the period of obstruction in a follow-up of *n* = 924 out of 1303 patients (71% follow-up rate)

Group no.	Obstruction period (years)	Patients (*n*)	Patency rate (%)	Pregnancy rate (%)	Average age of the partner (years)
1	<5	204	97 (*n* = 197)	66 (*n* = 134)	33.2
2	5–9	361	91 (*n* = 328)	64 (*n* = 231)	34.5
3	10–15	251	84 (*n* = 211)	51 (*n* = 128)	33.8
4	>15	108	81 (*n* = 87)	48 (*n* = 52)	35.1
	Total	924	89 (*n* = 823)	59 (*n* = 545)	34.6
	Statistical significance between group (no/no)		*p* = 0.0103 (1/2) *p* = 0.0155 (2/3) *p* = 0.4449 (3/4)	*p* = 0.7147 (1/2) *p* = 0.0015 (2/3) *p* = 0.4645 (3/4)	*p* < 0.05 between all groups

Table 3 Ejaculate quality after vasectomy reversal in relation to the period of obstruction. Follow-up includes 788 patients with semen analyses according to World Health Organization 2010 (Oligozoospermia: <40 Mio/ejac., Asthenozoospermia: progressive motility <32%). Of 924 patients followed up, 136 reported a pregnancy without having had semen analyses

Group	Obstruction period (years)	Patients (*n*)	Normozoospermia (%)	Oligozoospermia (%)	Asthenozoospermia (%)	Oligoasthenozoospermia (%)	Azoospermia (%)
1	<5	138	70 (*n* = 97)	5 (*n* = 7)	7 (*n* = 9)	13 (*n* = 18)	5 (*n* = 7)
2	5–9	317	67 (*n* = 212)	6 (*n* = 20)	9 (*n* = 27)	8 (*n* = 26)	10 (*n* = 32)
3	10–15	240	55 (*n* = 133)	6 (*n* = 14)	17 (*n* = 42)	5 (*n* = 11)	17 (*n* = 40)
4	>15	93	52 (*n* = 48)	5 (*n* = 5)	14 (*n* = 13)	6 (*n* = 6)	23 (*n* = 21)
	Total	788	62 (*n* = 490)	6 (*n* = 46)	12 (*n* = 91)	8 (*n* = 61)	12(*n* = 100)
	Statistical significance between groups (no/no)		*p* = 0.0016 (1/2) *p* = 0.0063 (2/3) *p* = 0.5422 (3/4)				*p* = 0.1003 (1/2) *p* = 0.0296 (2/3) *p* = 0.2109 (3/4)

were not detectable. Nevertheless, great importance was attached to the follow-up using an individualized database (based on filemaker). Statistical analysis was performed by use of Fisher's exact test.

Patients were urgently asked to have a sperm analysis performed after 3 month and to report the occurrence of a pregnancy. This request was explicitly documented in the medical report to the urologists responsible for further treatment. Unless a response had been received concerning sperm analysis and/or pregnancy, active follow-up consisted of telephone inquiries with patients or urologists 1 year after the operation, strongly recommending a semen analysis.

Forty-eight patients (4%) had actually no desire to have children or wished to have the operation for other reasons, such as chronic pain syndrome or psychic or religious motives. This group of patients was not considered in the follow-up.

The follow-up period comprised at least 3 month, in one case 14 years where the patient presented for repeat vasectomy after having fathered four children.

Altogether, 924 of 1303 cases were followed up, showing an overall patency rate of 89% and a pregnancy rate of 59% without any use of IVF (Table 2).

Patency was demonstrated by semen analyses according to World Health Organization (2010), performed by the referring urologists or in our andrological centre. In 136 patientes who reported a pregnancy without having had semen analyses, patency was assumed.

Among 788 patients who had follow-up semen analyses, 490 (62%) were found to be normozoospermic, 198 (26%) had oligo- and/or asthenozoospermia of different markedness, 100 (12%) were azoospermic (Table 3). Of 823 patients with initially demonstrated patency, 8 (1%) experienced repeated occlusion after 3–13 month (late failure); in six cases, refertilization was successfully performed again.

The patency and pregnancy rates include 136 pregnancies that occurred without follow-up semen analyses. This mainly concerns couples who achieved pregnancy within the first 3–4 month after refertilization so they saw no need (and could not be persuaded) to have a sperm analysis performed.

Discussion

In cases of obstructive azoospermia methods of MR can be used to achieve natural fertility, whereas the alternative

procedure of ICSI is a means of artificial reproduction with a relevant burden to the female partner and higher costs (Lee *et al.*, 2008).

The follow-up rate in our study is higher after long periods of obstruction (>10 years) compared with those <5 years, presumably because the results after short-time occlusion are better anyway and patients did not report the treatment success. This is supported by the fact that the majority of patients who reported pregnancies without having had sperm analyses belong to this group (85 of 136 patients).

Operation technique

Different techniques are used for MR, and since the last 30 years many papers have been published about these techniques and their results (Fischer & Grantmyre, 2000; Holman *et al.*, 2000; Marmar, 2000; Paick *et al.*, 2000; Dohle & Smit, 2005; Ho *et al.*, 2005; Parekattil *et al.*, 2006; Patel & Sigman, 2008; Lipshultz *et al.*, 2009; Jee & Hong, 2010). We introduced and consequently used a three-layer technique for VV and EV, resulting in single-surgeon experience over 18 years.

In our opinion, the three-layer technique is insignificantly more time-consuming than both the previous one- or two-layer techniques (Fischer & Grantmyre, 2000; Marmar, 2000; Paick *et al.*, 2000; Ho *et al.*, 2005; Jee & Hong, 2010) and the robotic techniques currently published (Fleming, 2004; Kuang *et al.*, 2004; Parekattil *et al.*, 2010). One reason for our preference of the three-layer technique over the one- or two-layer technique is the possibility of exact adaptation of the interior layer which typically shows luminal disparity.

Secondly, the third layer of the anastomosis, i.e. the connective tissue coat (adventitia), provides tension relief to the internal layer to a greater extent than does the second layer alone. Furthermore, the third layer ensures vascularization of the duct. According to the author's experience, preservation of the connective tissue layer is of essential importance for a successful operation because it prevents hypotrophy of the duct and ensures complete tension relief to the internal layer – an issue that has so far not been considered in the literature. When the stumps of the epididymal duct are prepared, the third layer of connective tissue should be preserved by all means. Excessive or prolonged denudation and loss of the third layer involve a high risk of secondary hypotrophy and fibrotic occlusion of the anastomosis.

Apart from the operative strategy, the aforementioned technical peculiarities of three-layer anastomosis may explain our favourable results. Although these are not better than many others published before, the 1% rate of secondary reocclusions is significantly lower compared with studies reporting rates up to 12% for VV and up to 21% for EV (Belker *et al.*, 1991; Matthews *et al.*, 1995; Silber & Grotjan, 2004; Kolettis *et al.*, 2005). In our experience, the secondary azoospermia rate is not relevantly underestimated, because patients with an initial positive result in semen analysis whose female partner doesn't become pregnant, will come to reevaluate the ejaculate quality in most cases.

We can present a large single-centre study with a follow-up rate of 71% relating to semen analyses or reported pregnancies.

A comparison of our results and those of others should primarily consider a study by Silber & Grotjan (2004) who published their findings with two-layer VV and EV in 4010 patients, reporting a high follow-up rate of 86.5% and patency rates of 95% for bilateral VV and 78% for bilateral EV. The problem of low follow-up rates is to be found in many studies on refertilization, reflecting insecurity about whether the patients who were not followed up are statistically equal to those who underwent long-term follow-up.

Importance of epididymovasostomy

We suggest that the most important prerequisite for achieving good results even after a long period of obstruction is the consistent implementation of the strategy to perform epididymovasostomy if no spermatozoa is detectable at the epididymal stump of the vas deferens (Sheynkin *et al.*, 2000; Sigman, 2004).

In 24% of our patients, predominantly in those with longer periods of obstruction, we encountered the situation that a bypass anastomosis had to be performed at least on one side. So clearly the indication for epididymovasostomy is statistically correlated with the period of obstruction (see Table 1). This is in accordance with the experiences of many other authors (Goldstein & Girardi, 1997; Eguchi *et al.*, 1999; Matsuda, 2000; Boorjian *et al.*, 2004; Chawla *et al.*, 2004; Silber & Grotjan, 2004; Schiff *et al.*, 2005; Schlegel & Margreiter, 2007; Hinz *et al.*, 2008; Magheli *et al.*, 2010).

Epididymal damage

Similar to other studies (Belker *et al.*, 1991; Kolettis *et al.* 2006; Silber & Grotjan, 2004; Bolduc *et al.*, 2007) we found a significant discrepancy between the patency and the pregnancy rates, independently of the interval of obstruction. In most cases this discrepancy was explained by the demonstration of asthenozoospermia or oligoasthenozoospermia in the post-operative semen analyses. This pathologic finding could be caused by epididymal damage because of a long period of obstruction or

antisperm antibodies (McDonald, 1996; Marconi *et al.*, 2008; Légaré *et al.*, 2010).

In Tables 2 and 3 it is shown that with increasing time of obstruction the sperm quality and pregnancy rates are decreasing. However, the patency rates did not differ significantly with the obstruction period.

In our opinion it can be concluded that the main reason for the decreasing pregnancy rates lies in the decreasing sperm quality. Directly correlating individual pregnancies to individual semen analysis is limited, because for a relevant group of pregnant females (136) no semen analysis of the male partners was available. In our study the distribution of the female age was similar in all the groups independent from the obstructive period. Besides the female factor no other important factor was obvious for us. So we conclude that the sperm quality is the most relevant factor influencing the pregnancy rates.

Spermatogenesis is not altered by the obstruction for at least 20–25 years, which was shown by histological studies in patients with obstructive azoospermia. Only insignificant alterations of spermatogenesis are described, such as interstitial fibrosis (Shiraishi *et al.*, 2002) and increased sperm DNA fragmentation (Smit *et al.*, 2010). However, the epididymis suffers from obstruction in that it decreases with time (Srivastava *et al.*, 2000; Lavers *et al.*, 2006; Yang *et al.*, 2007). Statistically, this deteriorating effect to the epididymis is related to the interval of obstruction, which finds expression in the necessity of EV (Table 1). However, individual differences in the resistance to time-related epididymal damage must be presumed because in single cases of normozoospermia complete recovery of the epididymis after refertilization may occur.

Female fertility factor

Another relevant factor for the difference between patency and pregnancy rates may be the relatively high average age of 34.6 years of the female partners, affecting their fertility. However the age of the female partner at the time of operation was not significantly related to the period of obstruction among the male patients.

Table 2 suggests that the female age being the most important female fertility factor can be considered as an independent variable (Fuchs & Burt, 2002; Gerrard *et al.*, 2007).

Furthermore it should be realized that with increasing female age abortion rates are most likely increasing. This should be considered when birth rates are discussed. So birth rates are probably somewhat lower, as was already shown in other studies (Belker *et al.*, 1991; Silber & Grotjan, 2004).

Importance of the three-layer technique

The advantage of the three-layer technique with a high number of stitches is a perfect seal of the internal layer, preventing leakage with the possible consequence of granuloma. In addition, the third layer with about ten 8-0 stitches is sufficient to prevent any tension to the internal layer of the anastomosis which secures the liquid-tight seal. The third layer is important for vascularization of the deferent duct, so that consequent preservation of this connective tissue coat prevents scarring at the anastomotic site and secondary occlusion. In the present study, this is supported by the low rate of 1% of secondary azoospermia (late failure) and a very low complication rate.

Although excellent results with the two-layer technique are published in the literature we feel that the three-layer offers a promising addition the established surgical techniques.

In our opinion, the three-layer anastomosis is no more time-consuming than a two-layer technique and the patient benefit justifies the higher amount of time compared with the single-layer technique. According to our experience, this sophisticated reconstruction of the seminal tract using at least two-layer-, better three-layer technique, in the framework of a minimally invasive procedure should be the standard of refertilization surgery against which all other techniques, such as the robotic technology, must be measured.

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