

Sperm analysis of the vas deferens fluid after a long interval of unilateral percutaneous epididymal sperm aspiration in vasectomized patients

Fernando Lorenzini, Eduardo Zanchet, Mariana Lorenzini

Department of Urology, Federal University of Parana and Department of Urology, Hospital Cruz Vermelha, Curitiba, Brazil

ABSTRACT

Objectives: Evaluation of the presence of spermatozoa in vas deferens fluid after a long interval of unilateral and homolateral percutaneous epididymal sperm aspiration (PESA) in vasectomized men. When found, the spermatozoa were evaluated including concentration and motility, in order to verify the patency of the epididymal tubules.

Materials and Methods: Four patients, numbered in a progressive order, from one to four, with 38, 40, 48 and 51 years old and vasectomy interval of 10, 10, 25 and 11 years, respectively, whose wives did not get pregnant using intracytoplasmic sperm injection of sperm obtained by unilateral PESA and decided to try only natural conception, were submitted to intrasurgical sperm analysis of the vas deferens fluid (ISAVDF) during microsurgery for reconstruction of the seminal tract.

Results: Time interval between PESA and ISAVDF was 13.75 \pm 11.12 months (x \pm s) varying from 3 to 29 months. Homolateral ISAVDF and PESA showed the presence of spermatozoa. Patients 1, 2 and 4 had a high concentration of 10 x 10 6 , 64 x 10 6 and 45 x 10 6 spermatozoa/ mL; the first two had motile sperms and patient 3 had no sperms. Conclusions: Three of four patients showed spermatozoa in the vas deferens fluid after a long interval of unilateral and homolateral PESA with high concentration, including motile forms. These findings support the concept that PESA may not result in late epipidymal tubule obstruction in vasectomized patients.

ARTICLE INFO

Key words:

PESA; Epididymis; Vasovasostomy; Spermatozoa; Vas Deferens

Int Braz J Urol. 2013: 39: 720-6

Submitted for publication: May 24, 2013

Accepted after revision: August 13, 2013

INTRODUCTION

Although most men at the time of vasectomy are sure of the choice of surgical contraception, around 4 to 6% will desire posteriorly to father children using their own spermatozoa (1). In those cases the treatment options include microsurgical reconstruction of seminal tract and intracytoplasmic sperm injection (ICSI).

In order to obtain spermatozoa to ICSI some surgical techniques are employed, as percu-

taneous epididymal sperm aspiration (PESA), testicular sperm aspiration (TESA), testicular sperm extraction (TESE) and microsurgical dissection of seminiferous tubules (microTESE) (2-4). It is still possible to use microsurgical epididymal sperm aspiration (MESA) (5-7), during which it is performed a microsurgical repair of the epididymal tubule in order to avoid posterior obstructive scarring of the tubules keeping them patent.

Classically, PESA is considered a recovery technique that causes obstruction of epididymal

tubules (3,8). The mechanism of obstruction is related to scarring after the use of the aspiration needle and leakage of spermatozoa.

In the vasectomized patient, the spermato-zoa flow freely inside the epididymal tubules until the place of the vasectomy with natural processes of production, death and absorption of sperm. On the other hand, it was believed that in the vasectomized patients submitted to PESA, there would be an air-tight space between the place of PESA and the site of vasectomy where spermatozoa would only suffer death and absorption, and after a period of time, this site would not present sperms.

The presence of high concentration of sperms including motile forms during intrasurgical analysis of vas deferens fluid (ISAVDF) after a long interval of time of PESA, homolateral to ISAVDF, would definitely demonstrate that PESA do not cause epididymal tubule obstruction. From a practical point of view, these ISAVDF findings would allow homolateral vasovasoanatomosis (VV) to PESA instead of vasoepididymal anastomosis (VE), that is more troublesome and more complex, with the need of great microsurgical ability and results of pregnancy and patency inferior to VV.

The objetive of the present study was to determine the presence of spermatozoa in the vas deferens fluid after a long period of the unilateral and homolateral PESA in vasectomized patientes, and, if affirmative, to determine their concentration and motility in order to infer epididymal tubule patency.

MATERIALS AND METHODS

The study was performed in four patients with azoospermia due to bilateral vasectomy who were submitted to unilateral PESA and ICSI, whose wives did not get pregnant. These patients further decided to father children spontaneously and decided to be submitted to ISAVDF during bilateral microsurgical reconstruction of seminal tract. It was proposed vasoepididymal anastomosis in the homolateral side submitted to PESA and VV or VE on the contralateral side of unilateral PESA, according to the results of ISAVDF.

VE was proposed using termino-lateral microsurgical anastomosis with single stitches of mononylon 10-0 and VV termino-terminal anastomosis with single stitches of mononylon 9-0, single plan under surgical microscope. The fluid from the proximal vas deferens stumps in relation to the epididymus was aspirated in natura with a 26G needle attached to a 1 mL syringe that was send to the clinical laboratory. After surgery, patients were evaluated every three months. Table-1 shows the patients age as well as their wives age, time since vasectomy, side, place and number of aspirative punctures with 26G needles during unilateral PESA and the time between unilateral PESA and the microsurgical reconstructive procedure.

RESULTS

The median age of the patients was 44.25 ± 6.24 years (medium \pm standard deviation) and of the wives 33.00 ± 1.63 years. The median time of interval between vasectomy and microsurgical reconstruction was 14.00 ± 7.35 years and between unilateral PESA and microsurgical reconstruction was 13.75 ± 11.12 months, varying from 3 to 29 months.

In all patients, the fluid from the vas deferens stump during ISAVDF homolateral and contralateral to unilateral PESA was whitish or transparent serous.

ISAVDF homolateral to unilateral PESA showed the presence of a high number of spermatozoa: 10×10^6 , 64×10^6 and 45×10^6 sperms/mL, with motility of 30%, 1% and 0% in patients 1, 2 and 4, respectively. Patient 3 showed no sperms.

Contralateral ISAVDF in relation to unilateral PESA showed the presence of high number of spermatozoa, of 8 x 10^6 and 54 x 10^6 sperms/mL and motility of 0% and 32% in patients 1 and 2, respectively; patients 3 and 4 had no sperms.

Tables 2 and 3 show the results of homolateral and contralateral ISAVDF in relation to PESA, respectively. All patients were submitted to VV. The follow-ups every three months of patients are described in Table-4.

DISCUSSION

After the report of the first ICSI (7), PESA became progressively used to recover sperm in vasectomized patients, since it is a very simple techni-

Table 1 - Distribution of patients, time since vasectomy and unilateral PESA data.

| Patient # | Age of patient/age of wife (years) | Time since vasectomy (years) | Side of unilateral PESA/num- ber of punctures and place at the epididymus | Time between unilateral PESA and microscopic reconstruction of seminal tract (months) |
|-----------|---------------------------------------|---------------------------------|---|---|
| 1 | 38/35 | 10 | Right/4Ca and 2Co | 3 |
| 2 | 40/31 | 10 | Right/5Ca and 2Co | 29 |
| 3 | 48/33 | 25 | Right/6Ca and 4Co | 14 |
| 4 | 51/33 | 11 | Left/5Ca and 2Co | 9 |

PESA: percutaneous epididymal sperm aspiration; **nCa:** number of aspirative punctures on the head of the epididymus with a 26G diameter needle, **nCo:** number of aspirative punctures of the body of the epididymus with a 26G diameter needle.

Table 2 - Results of intrasurgical analysis of vas deferens fluid homolateral to unilateral PESA.

| Patient # | Macroscopic aspect of the vas deferens fluid: consistency/color | Microscopic analysis of the vas deferens fluid |
|-----------|--|---|
| 1 | Serous/whitish | 10 x 10 ⁶ sperms/mL 30% motile |
| 2 | Serous/transparent | 64 x 10 ⁶ sperms/mL 1% motile |
| 3 | Serous/whitish | Abasence of spermatozoa |
| 4 | Serous/transparent | 45 x 10 ⁶ sperms/mL 100% non-motile |

que, but with the inconvenience of contraindicating a future VV (3,8).

Marmar et al. (9) described the first VV after bilateral PESA in eight vasectomized patients, and during the intrasurgical microsurgical procedure they evaluated the presence of spermatozoa in the vas deferens fluid proximal stump in relation to epididymis. They reported that these patients had been submitted to one to four bilateral PESA; seven patients showed spermatozoa at least in one of the stumps intrasurgically and among

three patients submitted to bilateral VV two of their wives got pregnant naturally.

Posteriorly Van Roijen (10) described two vasectomized patients submitted to bilateral VE that presented post-surgical azoospermia and that were next submitted to bilateral PESA. Three years after the bilateral PESA in one patient and one year after the other, patients presented spermatozoa in the semen, and they concluded that VE could in a late period result in epididymal tubule patency even after PESA.

Table 3 - Results of intrasurgical analysis of the vas deferens fluid contralateral to unilateral PESA.

| Patient # | Macroscopic aspect of the vas deferens fluid: consistency/color | Microscopic analysis of the vas deferens fluid |
|-----------|---|--|
| 1 | Serous/whitish | 8 x 10 ⁶ sperms/mL 100% non-motile |
| 2 | Serous/transparent | 54 x 10° sperms/mL 32% motile |
| 3 | Serous/whitish | Absence of spermatozoa |
| 4 | Serous/transparent | Absence of spermatozoa |

Table 4 - Post-surgical results of microscopic reconstruction of seminal tract (bilateral microsurgical vasovasoanastomosis).

| Patient # | Post-operatory sperm analysis* | Natural pregnancy of wife |
|-----------|--|---|
| 1 | 25,6 x 10 ⁶ sperms/mL Progressive motility = 20% Strict forms = 14% | Present: after 5 months of microsurgery Born of a healthy baby |
| 2 | 99,0 x 10 ⁶ sperms/mL Progressive motility = 48% Strict forms = 19% | Present: after 6 months of microsurgery Born of a healthy baby |
| 3 | Rare spermatozoa evolving to azoospermia | Absence |
| 4 | 6,8 x 10 ⁶ spems/mL Progressive motility = 13% Strict forms = 11% | Absence |

^{*} Arithmetic media of three post-operatory sperm analysis performed every three months.

This study presents four vasectomized patients submitted to unilateral PESA that posteriorly were submitted to microsurgical reconstruction of the seminal tract. In three patients it was observed the presence of spermatozoa with high concentration at the vas deferens stumps homolateral to unilateral PESA, with motility in two patients. Since the interval time between PESA and ISAVDF of all patients was equal or superior to three months, enough time for death and tubular absorption of remaining sperms between the

PESA and vasectomy sites, based on the results of ISAVDF, PESA did not cause late obstruction of the epididymal tubules. Macroscopic evaluations of the fluids collected during ISAVDF at the time of the microscopic reconstruction was coincident with the sperm analysis, since the fluids were serous, whitish or transparent, as shown in Tables 2 and 3.

Some additional observations must be made. First, in this study, there were a very limited number of patients, due to the rarity of vasectomized patients described in the literature that

were submitted to microscopic reconstruction after PESA and with ISAVDF results showing spermatozoa (9). Second, patient 3 did not have spermatozoa in both sides during ISAVDF. However, due to the bilateral macroscopic characteristics of the fluids in the vas deferens stumps (Tables 2 and 3), and although with a long interval time between vasectomy and reconstruction, it was performed bilateral VV. Table-4 shows that this patient presented rare spermatozoa in the post-operatory period, that evolved to azoospermia, meaning that at least in one side there was epididymal patency. This patient was maintained in the study since he matched the inclusion methodological criteria. Third, patient 4 had spermatozoa present during ISAVDF at the homolateral side of PESA and no sperms on the contralateral side. This aspect could be explained by the characteristics of the vas deferens fluid at the contralateral side of PESA. Maybe it did not represent all tubular fluid of the vas deferens stump and the epididymis, being only the most proximal fluid at the site of vasectomy where possibly there were no sperms. At the homolateral side in relation to PESA, all fluid or most of it was present. Fourth, although the results of sperm analysis and pregnancies at Table-4 could not be related exclusively to microsurgical reconstruction homo or contralateral to PESA, these data were presented since they were part of the post--operatory follow-up of patients.

In the present study it was also quoted the diameter of the needle and the number of aspirative punctures of epididymus during unilateral PESAs (Table-1). Accordingly, Saade et al. (11) after histologic analysis of epididymal tissue, after one to five percutaneous aspirative punctures of sperm of epididymal rats, with a 25G needle, they concluded that there is a cumulative effect of the amount of lymphoplasmocitary infiltrate, local fibrosis and volumetric augmentation of the connective tissue, directly proportional to the number of punctures.

The hypothesis why PESA do not cause epididymal tubule obstruction after PESA had already been quoted by Marmar et at (9). They are based on the possibility of self-repair of epi-

didymal tissue and that there are anatomically 10 to 15 efferent ducts that converge to a single epididymal tubule.

Finally, we suggest that multicentric and prospective studies with the same characteristics of the present must be realized, as well as studies in animals, in order to obtain a greater population analysis.

CONCLUSIONS

Three out of four patients showed sperm at the vas deferens fluid after a long period of the unilateral and homolateral PESA procedure, with high concentration and motile forms in two patients. It is possible to infer that PESA may not cause epididymal tubule obstruction in vasectomized patients.

CONFLICT OF INTEREST

None declared.

REFERENCES

- Belker AM, Thomas AJ Jr, Fuchs EF, Konnak JW, Sharlip ID: Results of 1,469 microsurgical vasectomy reversals by the Vasovasostomy Study Group. J Urol. 1991; 145: 505-11.
- 2. Glina S, Fragoso JB, Martins FG, Soares JB, Galuppo AG, Wonchockier R: Percutaneous epididymal sperm aspiration (PESA) in men with obstructive azoospermia. Int Braz J Urol. 2003; 29: 141-5; discussion 145-6.
- 3. Esteves SC, Miyaoka R, Agarwal A: Sperm retrieval techniques for assisted reproduction. Int Braz J Urol. 2011; 37: 570-83.
- Schlegel PN: Testicular sperm extraction: microdissection improves sperm yield with minimal tissue excision. Hum Reprod. 1999; 14: 131-5.
- Temple-Smith PD, Southwick GJ, Yates CA, Trounson AO, de Kretser DM: Human pregnancy by in vitro fertilization (IVF) using sperm aspirated from the epididymis. J In Vitro Fert Embryo Transf. 1985; 2: 119-22.
- 6. Silber SJ, Ord T, Balmaceda J, Patrizio P, Asch RH: Congenital absence of the vas deferens. The fertilizing capacity of human epididymal sperm. N Engl J Med. 1990; 323: 1788-92.

- 7. Palermo G, Joris H, Devroey P, Van Steirteghem AC: Pregnancies after intracytoplasmic injection of single spermatozoon into an oocyte. Lancet. 1992; 340: 17-8.
- 8. Chan PT, Libman J: Feasibility of microsurgical reconstruction of the male reproductive tract after percutaneous epididymal sperm aspiration(PESA). Can J Urol. 2003; 10: 2070-3.
- 9. Marmar JL, Sharlip I, Goldstein M: Results of vasovasostomy or vasoepididymostomy after failed percutaneous epididymal sperm aspirations. J Urol. 2008; 179: 1506-9.
- van Roijen JH: Two cases of delayed patency following "failed" epididymovasostomy and subsequent percutaneous epididymal spermaspiration. Can J Urol. 2010; 17: 5022-5; discussion 5025.
- Saade RD, Neves PA, Glina S, D'Ancona CA, Dambros M, Lúcio MA: Quantitative (stereological) and qualitative study of rat epididymis after vasectomy and percutaneous epididymal spermaspiration. J Urol. 2008; 179: 381-4.

Correspondence address:

Dr. Fernando Lorenzini, MD, PhD Universidade Federal do Paraná Serviço de Urologia do Hospital de Clínicas Rua General Carneiro, 181 Curitiba, PR, 80060-900, Brazil Fax: + 55 41 3244-1111 E-mail: lorenzini.ff@gmail.com

EDITORIAL COMMENT

The authors present an interesting study on the presence of viable sperm in the proximal vas deferens end of vasectomized patients who underwent PESA previously. This is not a frequently seen situation, but may be a challenging case in the office of those who treat Male Infertility.

PESA has long been admitted to induce epidydimal obstruction at the puncture site which would demand a vasoepidydimal reconstruction in cases where vas reconstruction was elected.

This work reveals this may not be true and vasovasostomy may be a viable choice in most

cases. Some points deserve to be highlighted: sperm appearance in the ejaculate may not occur immediately after vas reconstruction, specially when sperm is not detected intraoperatively; this doesn't mean, however, it will not appear at all. Besides, sperm may appear only temporarily before vanishing again. So cryopreservation should be considered if ART is still a future possibility which would avoid a novel scrotal intervention.

PESA can be performed using an insulin needle instead of a 25G needle. Our personal experience shows that viable sperm in adequate amount may be retrieved in most cases for IVF/ ICSI treatment. It should be true the thinner the needle the lower the trauma caused in the epidydimis.

Dr. Ricardo Miyaoka Androfert - Referral Center for Male Infertility Av. Dr. Heitor Penteado, 1464 - Taquaral 13075-460, Campinas, Sao Paulo, Brasil FAX: + 55 19 3295-8877 E-mail: rmiyaoka@uol.com.br