

East African Medical Journal Vol. 93 No. 1 January 2016

SUCCESSFUL IVF-ICSI WITH A LIVE BABY IN AN AZOOSPERMIC PATIENT UTILIZING CRYOPRESERVED TESTICULAR RETRIEVED SPERMS: CASE REPORT

J. Wanyoike-Gichuhi MBChB, MMed (O/G), S. N. K. Waweru, MBChB; Med. Surg. (Nbi), Cert. Urol. (EA), FCS, ESCA, Consultant Surgeon/Urologist, B. Kihara, MBChB, MMed (O/G), Department of Obstetrics/ Gynaecology, College of Health Services, University of Nairobi, P. O. Box 19676, Nairobi, Kenya, D. K. Ondieki, Consultant Obstetrician/ Gynaecologist, PO Box 2568-00202, Nairobi.

Request for reprint to: Dr. J. Wanyoike-Gichuhi, College of Health Services, University of Nairobi, P. O. Box 19676, Nairobi, Kenya

SUCCESSFUL IVF-ICSI WITH LIVE BABY IN AN AZOOSPERMIC PATIENT WITH CRYOPRESERVED SPERMS: CASE REPORT

J. WANYOIKE-GICHUHI, S. N. K. WAWERU, B. KIHARA and D. K. ONDIEKI

SUMMARY

Male factor is one of the most frequent cause of infertility and presents a variety of endocrine, genetic and immunological etiologies, as well as sexual malfunction, varicocele and genital infections. A case of a couple with primary infertility for two years is presented. Both partners were evaluated thoroughly, with a finding of male infertility. The man was found to be azoospermic after two semen analysis were performed. The Follicle Stimulating hormone (FSH) was raised with low testosterone levels indicating testicular failure. The recommended treatment was testicular biopsy with cryopreservation of the sperm and subsequent Intra-cytoplasmic Sperm Injection (ICSI). This treatment option was undertaken with successful implantation and live baby delivery. The case study presents the diagnostic modalities and management of male infertility with azoospermia.

INTRODUCTION

Azoospermia, the most severe form of male infertility, is caused by obstructions in the genital tract or by testicular failure (1). The prevalence of azoospermia, defined as complete absence of sperms from the ejaculate, is less than 1% of all men and approximately 10-11% among infertile men (2). The management of male factor infertility has been revolutionised by the development of Intra-cytoplasmic sperm injection (ICSI) and opened a new era in the field of assisted reproductive technology (3). In azoospermic patients, testicular sperm retrieval can be achieved through epididymis (Microsurgical epididymal sperm aspiration) or seminiferous (testicular sperm extraction (TESE) with good fertilisation and pregnancy (4) Sperm maturation occurs in the epididymis and therefore sperm retrieved from testis are not mature but with IVF/ICSI fertilisation, implantation and pregnancy are achieved (5-7).

The objective of this paper is to review a couple with male infertility- azoospermia who had testicular biopsy with cryopreservation of sperm. The frozen sperm were subsequently used in IVF-ICSI with successful implantation and live delivery.

CASE REPORT

A couple consisting of 32 years male and 30 years old female had presented with primary infertility for two years.

Investigation performed: Hysterosalpingogram: The endometrial cavity was normal and bilateral fallopian tubes were patent. Hormonal profile (Female): Follicle Stimulating Hormone (FSH)- 3.5 IU/L, Luteinising Hormone (LH)-4.2 IU/L, Estradiol- 118pg/ml - all normal findings. Ultrasonography: The uterus and bilateral ovaries were normal.

Semenalysis X 2: Azoospermia. Hormonal Profile (Male): Testosterone – 12ng/dl (Normal 240-950ng/dl), FSH- 15 IU/L (Normal 2-10IU/l). A diagnosis of azoospermia with testicular failure was made. The couple were informed and counselled on the diagnosis. They were informed that the only option for fertility treatment was surgical sperm retrieval with cryopreservation and subsequent IVF/ICSI.

Surgical intervention: Open testicular biopsy was performed and testicular tissue analysed. The tissue

was teased and extracted cells concentrated using enriched culture medium. There were no spermatozoa on the left testis. On the right there was 0.1 million spermatozoa / ml with poor motility but were cryopreserved for *In Vitro* Fertilisation (IVF) / Intracytoplasmic Sperm Injection (ICSI). The female patient was subjected to controlled ovarian stimulation (COS) with gonadotrophin 225iu. Oocyte retrieval was performed and yielded 16 Metaphase II oocytes. The cryopreserved sperm were used in ICSI with 13 grade 4 embryos being obtained. Three embryos were transferred with successful implantation one embryo and subsequent live delivery.

DISCUSSION

Thus, azoospermia defines the condition of absence of spermatozoa in the semen and is due to either obstruction in the genital tract or testicular failure. In obstructive azoospermia the process of spermatogenesis is not impaired. Men with obstructive azoospermia may father children either through surgical correction of the obstruction with natural conception or through sperm retrieval from epididymis or testis, followed by ICSI (2). Non-obstructive azoospermia is defined as a condition in which there is an impairment of various degrees in the process of spermatogenesis (1). The combination of azoospermia with normal levels of testosterone, FSH, and LH indicates a mechanical obstruction to the passage of sperm. An elevated serum concentration of FSH with low serum testosterone indicates germinal cell insufficiency or primary testicular failure. Low concentration of testosterone and FSH indicates hypothalamic or pituitary insufficiency. The patient presented azoospermia with high level of FSH and low levels of testosterone and hence a diagnosis of testicular failure was made.

For patients with testicular failure (with the exception of hypogonadotropic hypogonadism), no corrective approach is available, as medical treatment is not effective. The condition of hypogonadotropic hypogonadism is caused by pituitary or hypothalamic deficiency and treatment includes hormonal supplementation for the duration of time required for resumption of spermatogenesis. Thus, the only approach for treatment for non-hypogonadotropic, non-obstructive azoospermic patients is a surgical intratesticular attempt for the retrieval of spermatozoa (1). Sperm retrieval is generally performed under general anaesthesia.

Common methods for sperm retrieval are listed above. The choice of the method depends on the surgeon and embryologist involved in the patient care. The technique of sperm retrieval and the source of the sperm have no significant effect on the pregnancy. As long as viable sperms can be achieved, neither the duration of obstruction nor the motility of sperm

affects the outcomes achieved with IVF/ICSI (2). The patient presented underwent testicular sperm extraction (TESE) under general anaesthesia.

The first azoospermic patients to be treated by assisted reproduction techniques were those with obstructive azoospermia. Surgical sperm retrieval (MESA, PESA, TESE, TESA) combined with IVF has led to the achievement of viable pregnancies but was found to offer limited success primarily due to low fertilisation and implantation rates (8,9,10). The introduction of intra-cytoplasmic sperm injection (ICSI) has opened new horizons for the treatment of the patients with azoospermia (11). The efficacy of combining surgical sperm retrieval with ICSI for the treatment of infertility was analysed, demonstrating high fertilisation (58%) and pregnancy (35.7%) rates even with the use of grossly impaired testicular or epididymal spermatozoa (12).

Results showed high fertilisation rates and the development of normal embryos in 82% of sperm retrieval/ICSI cycles as compared to 19% with conventional IVF. Overall fertilisation and pregnancy rates were significantly higher using ICSI, being 45% and 47% as compared to 6.9% and 4.5% for conventional IVF, respectively. It was thus suggested that ICSI may be mandated for all patients who undergo surgical sperm retrieval.

The combination of surgical sperm retrieval/ICSI also enabled the freezing of testicular or epididymal spermatozoa to be used successfully in additional ICSI cycles avoiding repeated scrotal surgery (13). Frozen-thawed testicular and epididymal spermatozoa have been shown to have equivalent fertilisation and pregnancy rates as compared with fresh testicular and epididymal spermatozoa. There is no substantial evidence to indicate fresh testicular and epididymal sperms yield superior results (14,17). The duration of cryopreservation to IVF/ICSI does not appear to affect fertilisation rates or pregnancy rates (15,16). Based on these results, a new approach has been suggested in which sperm and oocyte harvesting need not be performed simultaneously.

This scheme may ease the burden of partner scheduling and assure the availability of spermatozoa prior to ovulation induction, thus avoiding unnecessary ovarian stimulation and oocyte retrieval. The patient presented had TESE with cryopreservation of the sperm with the utilisation of the sperm in later ICSI cycle.

The outcome of ICSI in the couple with azoospermia requiring testicular sperm retrieval, stipulates that the origin of the spermatozoa had no consequence on the prognosis (6). In patients with azoospermia, cryopreserved sperm after surgical sperm retrieval, showed acceptable fertilisation, embryonic development, and pregnancy rates (6). The promulgation of surgical sperm retrieval and ICSI has created a window where many men with

azoospermia can have a family. The patient presented had successful fertilisation with cryopreserved sperm, implantation and live birth.

In conclusion, cryo-preservation of the viable sperms should be performed while testicular biopsy is being performed. Open testicular biopsy with cryo-preservation of the sperm is good option for patients with azoospermia. Usually adequate sample of testicular tissue with high possibilities of sperm recovery is achieved. In the last few years, ICSI has provided a remarkably effective solution for severe male factor infertility. It is now well established that ICSI using testicular as well as epididymal spermatozoa in men with various causes of azoospermia provides high fertilisation and pregnancy rates with normal deliveries. Thus, routine cryo-preservation of testicular sperm at the time of diagnostic testicular biopsy or reconstructive surgery is a convenient, cost-effective procedure for patients requiring TESE for IVF / ICSI.

REFERENCES

- Ishikawa, T., Shiotani, M., Izumi, Y., *et al.* Fertilisation and pregnancy using cryopreserved testicular sperm for intracytoplasmic sperm injection with azoospermia. *Fert Ster.* 2009; **92**: 174-179.
- Hung, A. J., King, P. and Schlegel, P. N. Uniform testicular maturation arrest: A unique subset of men with nonobstructive azoospermia. *J Urol.* 2007; **178**: 608-612.
- Silber, S. J., Nagy, Z., Liu, J., Tournaye, H., *et al.* The use of epididymal and testicular spermatozoa for testicular spermatozoa for intracytoplasmic sperm injection: the genetic implications for male infertility. *Hum. Reprod.* 1995; **10**: 2031-2043.
- Nagy, Z., Liu, J. and Cecile, J. Using ejaculated, fresh and frozen-thawed epididymal and testicular spermatozoa gives rise to comparable results after intracytoplasmic sperm injection. *Fert. Steril.* 1995; **63**: 808-815.
- Silber, S. J., Van Streiteghem, A. C., Nagy, Z., Liu, J., *et al.* Normal pregnancies resulting for testicular sperm extraction and intracytoplasmic sperm injection for azoospermia due to maturation arrest. *Fertil. Steril.* 1996; **66**: 110-117.
- Sofran, N., Reubinoff, B. E., Porat-Katz, A., *et al.* Assisted reproduction for the treatment of azoospermia. *Hum Reprod.* 1998; **13**: 4 TES
- The management of infertility due to obstructive azoospermia. Practice committee of the American society for reproductive medicine evaluation. *Fertil. Steril.* 2008; **90**: 74-77.
- Temple Smith, P. D., Southwick, G. J. and Yates, C. A. Human pregnancy by in-vitro fertilisation (IVF) using sperm aspirated from the epididymis. *J. In Vitro Fertil. Embryo Transfer.* 1985; 112-122.
- Silber, S. J., Ord, T., Balmaceda, J., *et al.* Congenital absence of the vas deferens: the fertilizing capacity of human epididymal sperm. *N. Eng. J. Med.* 1990; **323**: 1788-1792.
- Hirsh, A., Mills, C. and Bekir, J. Factors influencing the outcome of in-vitro fertilisation with epididymal spermatozoa in irreversible obstructive azoospermia. 1994; **9**: 1710-1716.
- Palermo, G., Joris, H., Devroey, P., *et al.* Pregnancies after intracytoplasmic injection of a single spermatozoa into an oocyte. 1992; **340**: 17-18.
- Tournaye, H., Devroey, P. and Liu, J. Microsurgical epididymal sperm aspiration and intracytoplasmic sperm injection; a new effective approach to infertility as a result of congenital absence of the vas deferens the epididymis. *Fertil. Steril.* 1994; **61**: 1045-1051.
- Devroey, P., Silber, S., Nagy, Z., *et al.* Ongoing pregnancies and birth after intracytoplasmic sperm injection with frozen-thawed epididymal spermatozoa. *Hum. Reprod.* 1995; **10**: 903-906.
- Oates, R. D., Lobel, S. M. and Harris, D. H. Efficacy of intracytoplasmic sperm injection using intentionally cryopreserved epididymal spermatozoa. *Hum. Reprod.* 1996; 133-138.
- Huang, F. J., Lan, K. C., Lin, Y. C., *et al.* Impact of cryopreservation of spermatozoa obtained through testicular sperm extraction on Intracytoplasmic sperm Injection. *Fertil. Steril.* 2004; **81**: 1405-1407.
- Woods, S., Sephton, V., Scarlet, T., *et al.* Effect on clinical outcome of the interval between collection of epididymal and testicular spermatozoa and intracytoplasmic sperm injection in obstructive azoospermia. *J Androl.* 2003; **24**: 67-72.
- Park, Y. S., Lee, S. H., Song, S. T., *et al.* Influence on motility on the outcome of in vitro fertilisation / Intracytoplasmic sperm injection with fresh Vs frozen testicular sperm from men with obstructed azoospermia. *Fertil. Steril.* 2003; **80**: 526-350.