Live Births after Intracytoplasmic Sperm Injection in the Management of Oligospermia and Azoospermia in Nigeria

RA Ajayi, JH Parsons and VN Bolton

1The Bridge Clinic Limited, 1397a Tiamiyu Savage Street, P. O. Box 70294, Victoria Island, Lagos, Nigeria. E-mail: bridge@om.metrong.com 2The Assisted Conception Unit, 7th Floor, Ruskin Wing, King's College Hospital, Denmark Hill, London SE5 9RS.

Correspondence: R.A. Ajayi, The Bridge Clinic Limited, 1397a Tiamiyu Savage Street, P. O. Box 70294, Victoria Island, Lagos, Nigeria. E-mail: bridge@om.metrong.com

ABSTRACT

Intracytoplasmic sperm injection has revolutionised the management of male infertility. We report two cases that demonstrate the successful application of this technology in Nigeria in the management of both oligospermia and azoospermia. The first case relates to the treatment of a 31-year-old woman who required intracytoplasmic sperm injection of her husband's sperm for the treatment of both tubal fertility and male infertility. She had three embryos transferred on 9th June 1999 and was delivered of healthy male and female infants by caesarean section in January 2000 at 33 weeks gestation. The second case describes a 38-year-old woman who required intracytoplasmic sperm injection of the husband's surgically collected sperm for the management of azoospermia. She had two embryos transferred on 16th December 1999 and was delivered of a healthy male infant by caesarean section on 19th July 2001. (Afr J Reprod Health 2003; 7[1]: 121–124)
traitement de l'oligospermie et de l'azoospermie au Nigéria. L'injection intracytoplasmique du sperme a transformé radicalement le traitement de la stérilité masculine. Nous présentons deux cas qui illustrent le operati accompli dans l'application de cette technologie au Nigéria dans le domaine du traitement de l'oligospermie et de l'azoospermie. Dans le premier cas, il s'agit du traitement d'une femme âgée de 31 ans qui devait avoir besoin de l'injection intracytoplasmique du sperme de son mari pour le traitement de la fécondité tubaire et de la stérilité masculine. On a transféré trios embryons le 9 juin 1999. En janvier 2000, à la fin de 33 semaines de gestation, elle a accouché à l'aide d'une operation césarienne, deux enfants en bonne santé, un garçons et une fille. Le deuxième cas présente une femme âgée de 38 ans qui devait avoir besoin de l'injection intracytoplasmique du sperme de son mari collecté chirurgicalement, pour le traitement de l'azoospermie. Deux embryos ont été transféré le 16 décembre 1999 et elle avait accouché un fils en bonne santé le 19 juillet 2001 à l'aide d'une operation césarienne. (Rev Afr Santé Reprod 2003; 7[1]: 121–124)

KEY WORDS: Male infertility, azoospermia, testicular sperm, oligospermia, intra-cytoplasmic sperm injection (ICSI), ejaculated sperm, Nigeria

CASE REPORT 1

The couple presented to our clinic in Lagos in November 1998 requesting assistance in achieving a pregnancy. They had been trying to conceive since 1993. The woman, aged 30 years, and her husband, aged 37 years, had achieved two pregnancies together previously. The first was in 1993, which ended as a first trimester loss, the second in 1994, resulting in a laparotomy and right salpingectomy for a right ectopic pregnancy. The husband had fathered two pregnancies with a previous partner neither of which had resulted in live birth. The woman had had her appendix removed in 1989. A hysterosalpingogram carried out following the right salpingectomy in 1994 revealed that the left fallopian tube was not opacified, and it was concluded that the tube was blocked. There was no other significant past medical history.

The woman weighed 67kg, was 1.6 metres tall and her early follicular phase gonadotrophin levels were normal. Trans-vaginal ultrasound revealed a normal uterus and polycystic ovaries.¹ There were no other abnormal findings. The husband produced a 1.2ml semen sample; sperm concentration of 14 million per ml, 66% of which were motile and 9% of which were morphologically normal. The couple was advised that semen of this quality would almost certainly fail to achieve fertilisation with conventional in vitro fertilisation (IVF) treatment, and they agreed to undergo a cycle of intra-cytoplasmic sperm injection (ICSI).

At that time, all the assisted conception procedures at the Lagos clinic were carried out with the direct assistance of clinicians and embryologists from the assisted conception unit
of the King's College Hospital, London. Therefore, the couple's treatment was scheduled for June 1999 to coincide with the visit of the team from United Kingdom.

Pituitary down regulation using buserelin acetate (Suprefact®, Hoecht Marion Roussel, UK), ovarian stimulation using human menopausal gonadotrophin (Pergonal®, Serono, UK), human chorionic gonadotrophin (Profasi®, Serono, UK) and transvaginal ultrasound-directed follicle aspiration were carried out as described previously. Oocytes and embryos were cultured in universal IVF medium (Medi-Cult, Copenhagen, Denmark) and ICSI was carried out as described previously.

Of the 25 oocytes retrieved, 16 were metaphase II stage (MII). The repeat semen sample contained 4 million spermatozoa per ml, of which 66% were motile and 9% were morphologically normal.

Each of the 16 MII oocytes were injected with a single spermatozoon as described previously, and 12 fertilised normally, as indicated by the development of two pronuclei by 16–18 hours post-injection. All 12 zygotes underwent cleavage and three four-cell embryos, all of which showed good morphology, were transferred to the woman's uterus on 9th June 2001, two days after oocyte retrieval. The remaining embryos were discarded according to the couple's wishes. The luteal phase was supported with 400mg progesterone daily (Cyclogest®, Shire Pharmaceuticals Ltd, UK). A positive pregnancy test was performed 14 days after embryo transfer, on 23rd June 2001, and a viable triplet pregnancy was confirmed with the identification of three fetal hearts using ultrasound two weeks later.

The antenatal course was complicated by the intrauterine death of one of the fetuses at 19 weeks gestation, but the remaining fetuses were not compromised and the pregnancy continued without complications until 33 weeks when the woman went into premature labour. A live male infant weighing 1.8kg and a live female infant weighing 1.6kg were delivered by caesarean section on 12th January 2000. The children are well and have achieved all developmental milestones.

**CASE REPORT 2**

The couple presented for treatment in June 1999, having been trying to achieve a pregnancy since 1992. The woman, aged 38 years, and her husband, aged 46 years, had never achieved a pregnancy together. The man was confirmed azoospermic in 1993. Since then he had had various treatments including bilateral varicocelectomy in 1995.

The woman weighed 89kg and was 1.7m tall. Her early follicular phase gonadotrophin levels were normal. Trans-vaginal ultrasound revealed the uterus to be normal sized and
anteverted, containing two intramural fibroids of 1.5cm and 1.3cm respectively. The fibroids were not near the endometrial echo, which was clear and not distorted. Both ovaries were normally placed and had normal appearances. The man's semen was found to contain no spermatozoa even after centrifugation at 1800g on two separate occasions. His serum FSH was 4 IU/L and he had normal genitalia.

Elective testicular sperm extraction (TESE)\(^7\) was carried out under local anaesthesia on 11th June 1999. Percutaneous epididymal sperm aspiration (PESA)\(^8\) and testicular sperm aspiration (TESA)\(^9\) had both failed to yield sperm. A sample containing six non-motile spermatozoa per slide was obtained from the right testis, and one containing five spermatozoa per slide, of which only one was motile, was obtained from the left testis. The samples were divided into two, one of which was cryopreserved for subsequent ICSI. The other was sent for histological assessment, which showed both testes to have “severe hypospermatogenesis”.

The implications of these findings were explained to the couple and they elected to undertake a cycle of ICSI treatment. At that time, all the assisted conception procedures at the Lagos clinic were carried out with the direct assistance of clinicians and embryologists from the assisted conception unit of King's College Hospital, London.\(^2\) Therefore, the couple's treatment was scheduled for December 1999, to coincide with the visit of the team from United Kingdom.

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Of the five oocytes retrieved, four were MII stage. On thawing, the cryopreserved testicular sample was found to contain no motile spermatozoa. A second TESE procedure was carried out and samples containing approximately 10 non-motile spermatozoa per slide were obtained from each testis. A single twitching spermatozoon was seen on a slide prepared from one of the samples after incubation for four hours. This sample was used for ICSI.

Each of the four MII oocytes was injected with a single spermatozoon that had shown twitching movements prior to injection and two fertilised normally, as indicated by the presence of two pronuclei 16–18 hours post-injection. Both zygotes cleaved to generate good quality four-cell and five-cell stage embryos\(^6\) which were transferred to the woman's uterus on 16th December 1999, two days after oocyte retrieval. The luteal phase was supported with 400mg progesterone daily (Cyclogest\(^\text{®},\) Shire Pharmaceuticals Ltd, UK).
A positive pregnancy test was performed 14 days after embryo transfer, on 30th December 1999, and a viable singleton pregnancy was confirmed with the identification of a beating fetal heart using ultrasound three weeks later. The antenatal course was complicated by the onset of pre-eclampsia and she was delivered of a live male infant at 33 weeks gestation, on 19 July 2001. He is currently very well and has achieved all developmental milestones.

**DISCUSSION**

These reports describe the first conception and live birth following ICSI in Nigeria and, to our knowledge, in any other West African country. In developed countries, assisted conception techniques such as IVF and ICSI\(^\text{10}\) for the treatment of infertility have become routinely available, but couples in the developing world have more limited access, usually having to travel overseas at considerable expense. We have been able to achieve success with this highly specialised treatment in Nigeria through collaboration between a newly established clinic in Lagos, Nigeria, and a long established assisted conception unit at King's College Hospital, London, UK. Its success demonstrates how such collaboration can lead to the transfer of technology, making this specialised form of infertility treatment more accessible to those in developing countries who may need it.

The success of ICSI using spermatozoa recovered from the epididymis\(^\text{7}\) or the testes\(^\text{8,9}\) is well established in the treatment of infertility due to azoospermia, and in developed countries, the limiting factor for success in such cases is whether or not it is possible to retrieve sperm.

The presumed diagnosis in the management of the second couple was obstructive azoospermia and, therefore, sperm retrieval was attempted first using percutaneous epididymal sperm aspiration.\(^\text{8}\) When this was unsuccessful, testicular sperm aspiration\(^\text{9}\), and then the more invasive testicular sperm extraction\(^\text{7}\) were attempted. Even with TESE, few motile spermatozoa were retrieved, suggesting that the patient's azoospermia was, in fact, non-obstructive.

The initial TESE procedure was carried out in advance of the proposed date for treatment of the woman in order to confirm successful sperm retrieval before she embarked on expensive ovarian stimulation. Satisfactory fertilisation and pregnancy rates have been achieved with cryopreserved testicular sperm, and the facilities and expertise for cryopreservation were available in the Lagos clinic through collaboration with the centre in the UK. Despite this, the spermatozoa did not survive cryopreservation and thawing and it was necessary to perform a further TESA procedure on the day of the woman's oocyte retrieval.

An alternative approach would have been to perform the sperm extraction procedure after
oocyte retrieval, having prepared the couple in advance with appropriate counselling and informed consent for the use of donor sperm in the event of failure to retrieve spermatozoa.

ICSI has always been considered a risky procedure from its inception. The injection process is invasive to the oocyte and has the potential risk of causing biochemical or mechanical damage. More importantly, ICSI bypasses natural selection in that the sperm being injected into the oocyte is usually incapable of fertilising the oocyte without assistance and may carry structural defects or genetic abnormalities. Although these abnormalities may not impair the fertilisation process, they may manifest at birth or later in life. Initial studies confirmed these suspicions and demonstrated an increased risk of sex chromosone aneuploides in ICSI children.11

A Swedish follow-up study of 1,139 ICSI children also showed that when compared to children conceived normally, the ICSI children had a slightly higher number of cases of congenital malformations, but these malformations were mainly the result of the high rate of multiple births in ICSI children. The same study showed that the incidence of hypospadias was higher in ICSI children and this was probably related to paternal sub fertility.12 The largest study to date compared 2,995 IVF children and 2,899 ICSI children. It showed that there is no higher risk of neonatal complications or major malformations with ICSI and that the malformation rate in ICSI is not related to sperm origin or sperm quality.

REFERENCES


