# ROSI from TESE the First Case in Thailand : A Case Report

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**Abstract**

This is a report of a success in the treatment of male infertility in a case of azoospermic male patient. Testicular sperm extraction technique (TESE) was used and round cell spermatids were retrieved. *In vitro* fertilization was carried out using round cell spermatid injection technique (ROSI). The zygotes were laparoscopically transferred into right fallopian tube and intrauterine pregnancy was achieved. Cesarean delivery was performed at thirty eighth week of gestation yielding a healthy male infant weighing 2820 g. The child's physical and mental developments were normal after 2 years of follow-up.

**Key word** : ROSI, TESE, Case Report

Round cell spermatid injection (ROSI) is one of the new techniques in assisted reproduction. This technique was developed and applied for the correction of male infertility especially azoospermia. In normal semen, the round cell spermatid can be seen only if we examine the sample carefully. The round cell spermatid is the young form of the sperm and can not fertilize the egg naturally or even in actual IVF. However, it is able to do so when in conjunction with ICSI technique. This report presents a special case of a couple having a successful pregnancy after ROSI from testicular sperm extraction.

## CASE REPORT

An infertile couple first came to Siriraj Hospital in 1991. The woman was 26 years old and her husband was 32 years old. After the basic work up, the problem found in this couple was severe oligospermia. The first semen analysis showed a sperm concentration of about 50,000 sperms/ml and

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the sperm motility was only 15 per cent. The husband's serum FSH and testosterone level was tested and the result was 1.25 IU/L and 21.74 nmol/L respectively. After discussion on the aspects of infertility and the possibility of pregnancy, donor sperm insemination was suggested. The couple refused and requested medical treatment even though there was little chance of the woman becoming pregnant. The husband was given a low dosage of testosterone orally everyday. After six months of treatment, repeated semen analysis showed a sperm concentration of 75,000 sperms/ml and the motility of 30 per cent. There was not much improvement and the couple was lost follow-up.

In early 1996, the couple came back again and asked for the new assisted reproductive technology. Intracytoplasmic sperm injection (ICSI) was introduced and the couple accepted it. Unfortunately, prior to the program, the semen analysis was done again and showed azoospermia in two consecutive tests. Testicular biopsy was performed and resulted in a small area of active spermatogenesis. The elongated type of spermatid could hardly be seen but lots of round cell sperms were seen. So the testicular sperm extraction (TESE) and ICSI program was applied. The couple had signed the consent form and they also accepted the trial of round cell spermatid injection (ROSI) if the elongated form could not be found.

The long suppression protocol, using GnRH agonist 600 microgram per day was employed to the wife at the mid luteal phase. The super ovulation started on day four after the following menstruation by daily subcuticular injection of 150 units of hMG (human menopausal gonadotropin). The folliculogenesis was monitored by transvaginal ultrasound. When the follicular size reached 17 mm in diameter, intramuscular injection of 10,000 units of hCG (human chorionic gonadotropin) was given to the patient. Thirty seven hours later, transvaginal oocyte retrieval was done and yielded eight metaphase II oocytes and two of the metaphase I.

The eggs were cultured at 37°C in the human tubal fluid medium (HTF) and equilibrated with 5 per cent $CO_2$ in the incubator. After culture for awhile the surrounding granulosa cells were removed as the actual ICSI technique[1]. Then the husband's testicular biopsy was performed under local anesthesia and four small pieces of testicular tissue were excised. All the testicular tissues were dissected into smaller pieces, cultured at 37°C in the HTF medium and examined under an inverted light microscope. This testicular sample showed no elongated type of spermatid and few round cell type. ROSI was done six hours after the egg retrieval. The technique of injection (ICSI) was just as described before[1,2]. Fertilization was checked twelve hours after the ROSI and found that there were four normal two-pronucleated zygotes, one was degenerated and three were not fertilized. Three of the two-pronucleated zygotes were transferred back to the right fallopian tube under the laparoscopic technique. The patient was then given 200 mg of progesterone daily for luteal support.

Two weeks later serum ß-hCG was done and found to be greater than 250 mIU/ml. Serial ultrasound was performed at five and seven weeks of gestation and demonstrated that there was a single intrauterine pregnancy with an active fetal heart beat. The pregnancy was uneventful. When the pregnancy exceeded 18 weeks of gestation, ultrasonography of the fetus showed no gross anomaly and amniocentesis was suggested but the patient refused. On January 24, 1997 when the pregnancy was thirty eight weeks of gestation the mother developed pregnancy induced hypertension with premature rupture of membranes. Cesarean section was performed and yielded a healthy male infant weighing 2,820 g. The child is now nearly two years of age and development both physical and mental is normal.

## DISCUSSION

In the past, azoospermia was one of the most difficult problems and almost impossible to solve. Donor insemination was only one way to correct such a problem. But nowadays the assisted reproduction technique has been improved so much that the male problem can now be corrected. Micromanipulation is the technique that can be applied for husbands who suffer from oligospermia, asthenospermia or even teratospermia. There are so many micromanipulation techniques. Intracytoplasmic sperm injection (ICSI) is the most commonly used among those techniques especially in oligospermic patients or cases with failed fertilization[3]. Pregnancy after ICSI is as possible as the conventional in vitro fertilization (IVF). Recently, in cases of azoospermia, if there were some sperms in the testis it was possible to perform ICSI and finally result in pregnancy[4,5]. The testicular sperm morphology

varies depending on the stage of spermatogenesis. The round cell spermatid is the one in the early stage of sperm maturation and then develops to the more mature, elongated spermatid. The round cell spermatid was unable to move and its morphology was difficult to separate from the white blood cell. Testicular sperm extraction (TESE) is quite easy to perform and pregnancy after injection of elongated spermatid (ELSI) has been reported from many centers[6,7]. The round cell spermatid injection (ROSI) when compared to ELSI is an uncommon procedure and has yielded a lower pregnancy rate. Pregnancy after ROSI has been reported from either ejaculated semen or TESE[8,9]. In Thailand, ROSI is not usually performed because of its difficulty and the low fertilization rate. In the reported case, the husband showed a degradation of spermatogenesis from oligospermia to azoospermia. It was so poor that even elongated spermatids were not found. After TESE the ROSI technique, embryo transfer was done, resulting in pregnancy. The woman delivered a healthy male infant. The development of the child has been followed-up and is normal.

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## REFERENCES

1. Terasik J, Mendoza C. Spermatid injection into human oocytes. I. Laboratory techniques and special features of zygotes development. Hum Reprod 1996;11:772-9.
2. Devroey P, Liu J, Nagy Z, Tournaye H, Silber SJ, Van Steirteghem AC. Normal fertilization and implantation rate after intracytoplasmic sperm injection. Fertil Steril 1994;62:639-41.
3. Van Steirtghem AC, Liu J, Joris H, et al. Higher success rate by intracytoplasmic sperm injection than by subzonal insemination. Report of a second series of 300 consecutive treatment cycles. Hum Reprod 1993;8:1055-60.
4. Devroey P, Liu J, Nagy Z, et al. Pregnancies after testicular sperm extraction and intracytoplasmic sperm injection in non-obstructive azoospermia. Hum Reprod 1995;10:1457-60.
5. Craft I, Tsirigotis M, Bennett V, Nicholson N. Fertilizing ability of testicular spermatozoa. Lancet 1993;342:864-5.
6. Fishel S, Green S, Bishop M, et al. Pregnancy after intracytoplasmic injection of spermatid. Lancet 1995;345:1641-2.
7. ilber SJ, Van Steirteghem AC, Liu J, et al. High fertilization and pregnancy rate after intracytoplasmic sperm injection with spermatozoa obtained from testicle biopsy. Hum Reprod 1995:10:148-52.
8. Terasik J, Rolet F, Brami C, et al. Spermatid injection into human oocytes. II. Clinical application in the treatment of infertility due to non-obstructive azoospermia. Hum Reprod 1996;11:780-3.
9. Antinori S, Versaci C, Dani G, Antinori M, Selman HA. Successful fertilization and pregnancy after injection of frozen-thrawed round spermatids into human oocytes. Hum Reprod 1997; 12: 554-6.

# ความสำเร็จของการตั้งครรภ์ครั้งแรกในประเทศไทยที่ได้จากวิธี ROSI จาก TESE

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รายงานความสำเร็จในการรักษาผู้มีบุตรยากเนื่องจากสาเหตุทางฝ่ายชายที่มีปัญหาเนื่องจากไม่มีตัวอสุจิในน้ำอสุจิที่หลั่งออกมาและพบเพียงเซลล์อสุจิชนิด round cell ในเนื้อเยื่อลูกอัณฑะ ได้นำอสุจิชนิด round cell ดังกล่าวมาผ่านขบวนการเด็กหลอดแก้วร่วมกับ intracytoplasmic sperm injection ภายหลังการย้ายตัวอ่อนกลับคืนสู่มารดาให้ผลการตั้งครรภ์และคลอดเด็กปรกติ เพศชาย น้ำหนัก 2,820 กรัม ภายหลังการติดตามเป็นเวลาสองปีพบว่าเด็กมีการพัฒนาเป็นไปตามปรกติ

**คำสำคัญ** : อาร์โอเอสไอ, ทีอีเอสอี, รายงานความสำเร็จ

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