

methyl sulfoxide (DMSO) (ES), 15% EG + 15% DMSO + 0.5M S (VS) using cryotop, C: 7.5% EG + 7.5% (DMSO) (ES), 15% EG + 15% DMSO + 0.65M S + 10 mg/ml Ficoll (VS) using a cryoloop, D: 1.5M 1,2 propanediol (PROH) (ES), 1.5M PROH + 0.2M S (FS) using a straw. A, B and C were performed using vitrification and D using slow freezing and rapid thawing. We performed the cryopreservation of human mature oocytes in 5 patients with informed consent for clinical use.

**RESULTS:** SR, FR, and CR were 66.7% (4/6), 50.0% (2/4) and 100% (2/2), 100% (12/12), 41.7% (5/12), and 100% (5/5), 73.3% (11/15), 45.5% (5/11), and 80.0% (4/5), 82.2% (60/73), 63.3% (38/60), and 97.4% (37/38) in groups A, B, C, and D, respectively. There were no significant differences among the 4 groups. However groups B, D showed 33.3% (1/3), 26.7% (4/15) BR respectively, whereas the other groups showed 0%. One of one patient achieved on-going pregnancy after single blastocyst transfer using group B (SR:5/5, FR:5/5, CR 4/5, BL2/5, BT: 1, re-vitrification :1) Two of four patients achieved pregnancy using the group D protocol. One delivered a female healthy baby and one miscarried.

**CONCLUSION:** The present study suggests that slow freezing (group D) is an effective method for cryopreservation of oocytes. However, with further advances, vitrification will become the main method of preserving mature human oocytes due to the many advantages of this procedure, such as: time effectiveness, simplicity and cost reduction by eliminating the need for items such as program freezers.

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#### O-338

**Efficiency of testicular sperm aspiration (TESA) to recover spermatozoa in non-obstructive azoospermia.** M. Bibancos, A. Iaconelli Jr., L. G. Maldonado, L. M. Rossi, T. C. Bonetti, E. Borges Jr. Fertility – Assisted Fertilization Center, Sao Paulo, Brazil.

**OBJECTIVE:** Recently several studies have assessed the fertilization and pregnancy rates of intracytoplasmic sperm injection (ICSI) using spermatozoa retrieved from the testis in patients with non-obstructive azoospermia. It seems that these patients have small foci of spermatogenesis in the testis,

which allows recovers sperm using surgery techniques. For couples with infertility due to non-obstructive azoospermia, testicular sperm aspiration (TESA) combined with ICSI have been enabled successful treatments for infertility. The objective of this study was to evaluate the success of sperm recovery in TESA procedures from non-obstructive azoospermic patients.

**DESIGN:** Retrospective observational study.

**MATERIALS AND METHODS:** One hundred and twenty five TESA procedures were performed for ICSI, in 86 patients with non-obstructive azoospermia. The presence of azoospermia was documented on previous semen analyses, including a centrifugation step at high speed. The 86 patients were divided as having: 47 testicular failure (testicular atrophy or FSH > 15 mU/mL) (55%), 19 cryptorchidism (22%), 11 orchitis (13%), 7 idiopathic cause (8%) and 2 abnormal karyotype (2%). Based on testicular histology, patients were diagnosed as: 31 without pathology diagnosis (36%), 21 Sertoli cell only syndrome (24%), 19 hypospermatogenesis (22%), 11 maturation arrest (13%), 4 tubular hyalinization (5%). The technique for TESA was performed under cord block anesthesia and a 21 gauge butterfly needle was used and inserted into the superior pole of testis in a perpendicular course; the needle were advanced and pulled back multiple times in the same direction. The procedure was repeated until there was a return of an opaque and yellow fluid and/or tissue in the butterfly tubing. After that, the butterfly tubing was clamped in order to produce a negative pressure, so the seminiferous tube was totally retrieved with nippers. It was followed by microscope search of the testicular tissue aspirated to confirm the presence of spermatozoa. The positive TESA was defined as the recovery of any number of motile or immotile sperm.

**RESULTS:** Successful retrieval of testicular spermatozoa using this technique was achieved in 75% of 125 procedures, 79 cases were found motile spermatozoa (63%) and 15 immotile spermatozoa (12%). Round cells or no spermatozoa were found only in 31 procedures (25%).

**CONCLUSION:** Although the literature report that average sperm recovery rate is lower than 50%, we could recover motile spermatozoa in 63% and immotile spermatozoa in 12% of TESA procedures. In general, the TESA technique used the needle insertions are followed by large exploration in testis. We are thought that better recovery rate is due to our technique, which use a punctual insertion without changes in needle direction and retrieving the seminiferous tube totally using nippers, which permit us to have a higher chance of found the foci of spermatogenesis.

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