

The use of epididymal and testicular sperm for ICSI

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Intracytoplasmic sperm injection (ICSI) has made the most severe cases of oligoasthenoteratozoospermia treatable. The presence of only a few weakly motile sperm in a centrifuged ejaculate are sufficient to produce results equivalent to that of IVF in couples with completely normal semen (Van Steirteghem et al., 1993a; Van Steirteghem et al., 1993b). ICSI with epididymal, and even testicular biopsy extracted sperm, gives pregnancy rates equivalent to that of normal ejaculated sperm from a fertile male (Devroey et al., 1994; Silber et al., 1994; Silber et al., 1995). Extensively applying these techniques, we have now found that even in men with apparently absent spermatogenesis, who have complete azoospermia, with no sperm even in the centrifuged semen, there is nonetheless usually some small amount of sperm production to be found somewhere in the testicles.

The rationale and results using testicular and epididymal sperm with ICSI for severe cases of male infertility began with the treatment of hopeless cases of obstructive azoospermia.

One hundred four (104) consecutive MESA cases were performed for congenital absence of vas (CAV) and for irreparable obstructive azoospermia, using direct intracytoplasmic injection (ICSI) of an individual sperm into metaphase II oocytes of the wife (see Tables 1, 2, and 3). Fertilization and normal embryos were obtained for transfer in 93% of cases. There was an overall fertilization rate of 50%, and a normal cleavage rate of 70%. The pregnancy rate per transfer and delivery rate per transfer were 53% and 37%, respectively. The delivery rate per cycle was 35%.

Table 1
OBSTRUCTIVE AZOOSPERMIA

Fertilization and cleavage rate after ICSI with epididymal, and with testicular biopsy sperm

Source of Sperm	Number of Cycles	Number of Eggs M-II Injected	2PN Oocytes (%)	Normal Cleaved Embryos
Fresh Epididymal (MESA)	50	670	356 (53%)	256 (72%)
Frozen Epididymal	8	128	48 (38%)	36 (75%)
Testicular Biopsy (TESE)	46	582	287 (49%)	194 (68%)
TOTALS	104	1380	691 (50%)	486 (70%)

In many cases, there was no epididymal sperm available, and testicular biopsy i.e., testicular sperm extraction (TESE) was resorted to for sperm retrieval. This approach had only a minor negative effect on results. The transfer rate was lower with TESE (89% versus 96%), and the sperm could not be frozen and saved for future cycles. But there was no dramatic difference in ongoing and delivered pregnancy rates with epididymal versus testicular tissue sperm (38% versus 30%).

Table 2
OBSTRUCTIVE AZOOSPERMIA

Fertilization and delivery rate after ICSI with epididymal and with testicular biopsy sperm

Source of Sperm	Number of Cycles	Number of Transfers	Clinical Pregnancies Per Transfer	Ongoing and Delivered Per Transfer	Ongoing and Delivered Per Cycle
Fresh Epididymal Sperm	50	48 (96%)	28 (58%)	19 (40%)	38%
Frozen Epididymal	8	8 (100%)	5 (63%)	3 (38%)	38%
Testicular Biopsy (TESE)	46	41 (89%)	18 (44%)	14 (34%)	30%
TOTALS	104	97 (93%)	51 (53%)	36 (37%)	35%

Table 3
OBSTRUCTIVE AZOOSPERMIA

Fertilization and delivery rate after ICSI with epididymal and testicular biopsy sperm

Age of Wife	# of Cycles	# of Eggs M-II	2PN (%)	Normal Cleaved Embryos	Clinical Pregs Per Cycle	Ongoing and Delivered Per Cycle
<30	27	374	186 (50%)	134 (73%)	20 (74%)	16 (59%)
30-38	55	771	403 (52%)	384 (70%)	27 (48%)	19 (34%)
>38	22	235	102 (43%)	68 (67%)	4 (18%)	1 (5%)
Totals	104	1380	691 (50%)	486 (70%)	51 (48%)	36 (35%)

Ongoing and delivery rates for wives under 39 = 35/82 - 43%

Results were not affected by whether the obstruction was caused by congenital absence of the vas (CAV) or failed vasoepididymostomy (V-E). Frozen epididymal sperm gave results similar to fresh sperm. The only significant factor which affected results, in fact, had nothing to do with any aspect of the male partner or the retrieved sperm, but was simply the age of the wife. Women under 30 had a 59% ongoing or delivered pregnancy rate. Women in the 30 to 38 age group had a 34% ongoing or delivered pregnancy rate. Women over 38 had only a 5% ongoing or delivered pregnancy rate. Fertilization and cleavage rates were not at all affected by the wife's age. This means that with ICSI, it doesn't matter how severe the obstructive azoospermia, or where we have to search to retrieve the sperm, from the epididymis or the testis. The only issue that could ever interfere with success rate is the wife's age.

Complex mechanisms involving epididymal transport may be beneficial for conventional fertilization of human oocytes (whether in vivo or in vitro), but none of these mechanisms are required for fertilization after direct microinjection into the egg (Silber, 1989). Thus, because of the consistently good results using epididymal sperm with ICSI as opposed to conventional IVF, and the similarly good results with testicular tissue sperm, ICSI is mandated for all future patients with obstructive azoospermia. All CAV patients and their wives require genetic screening for cystic fibrosis, and pre-implantation embryo diagnosis must, therefore, also be available in any full service MESA program.

The most exciting evolution of this work is the ability to obtain normal pregnancy rates using sperm extracted from testicular tissue not only in cases of obstruction, but surprisingly also in non-obstructive azoospermia. In fact, it is now clear that even with non-obstructive azoospermia, e.g., "Sertoli cell only," or maturation arrest, there are often some small foci of spermatogenesis which allow TESE with ICSI to be performed (Amelar et al., 1977; Colgan et al., 1979; Del Castillo et al., 1947; Micic et al., 1983; Nagpal et al., 1993; and Silber and Rodriguez-Rigau, 1993). In most men with apparently absent spermatogenesis causing non-obstructive azoospermia, a direct attack on the testicle will yield very small numbers of sperm or at least spermatids stuck to Sertoli cells. These can be used for ICSI and achieve normal pregnancy rates.

Thus, a future scenario that would be applicable for IVF with andrological patients is as follows: If there are any sperm in the ejaculate, no matter how few, use ICSI. If there are no sperm in the ejaculate, do direct testicular sperm extraction with ICSI. No further evaluation may be necessary.

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