Fertilization and Embryo Development Outcomes of Fresh Versus Frozen Oocytes with Surgically Retrieved Sperm

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Background:
Surgical sperm retrieval is the standard of care for patients wishing to use autologous gametes with azoospermia [1]. Prior studies have assessed the use of fresh versus frozen surgical sperm sources, with outcomes comparable in most circumstances, when sperm survive freeze-thaw [1-3]. With the increasing use of oocyte cryopreservation, the impact of using fresh versus frozen oocytes with surgically retrieved sperm has not been adequately investigated.

Objective:
To determine if fertilization and embryo development outcomes differ between fresh and frozen oocytes when using fresh surgically retrieved sperm, with a focus on sperm from men with non-obstructive azoospermia.

Materials and Methods:
This is a retrospective study performed at a single university-affiliated academic center. Records of frozen oocyte thaw cycles between May 2013 and December 2023 were assessed to identify individuals undergoing simultaneous intracytoplasmic sperm injection (ICSI) of both fresh and cryopreserved oocytes using fresh, surgically retrieved sperm via either testicular sperm extraction (TESE) or epididymal sperm aspiration. Analysis was restricted to oocytes vitrified at our center.

The primary outcome was the proportion of good quality day 3 embryos, defined as 6 or more cells, per oocyte inseminated. Secondary outcomes were the proportion of normal (2 pronuclei) fertilization per oocyte inseminated and the proportion of good quality day 3 embryos per normally fertilized oocyte. The Wilcoxon signed-rank test was used to compare outcomes between fresh and frozen oocytes from the same patients.

Results:
During this period, 28 individuals met inclusion criteria. Upon paired analysis of the population as a whole (Table 1), there was no statistically significant differences observed in the proportion of normal fertilization between groups. Similarly, the proportion of good quality day 3 embryos per inseminated oocyte ($p=0.33$) and the proportion of good quality day 3 embryos per normally fertilized oocyte ($p>0.99$) were comparable.

When restricting the analysis to the 22 patients undergoing TESE for non-obstructive azoospermia (NOA), similar outcomes were observed between the use of fresh or frozen oocytes when assessing fertilization rate ($p=0.20$), rate of good quality day 3 embryos per

oocyte (p=0.31), and rate of good quality day 3 embryos per normally fertilized oocyte (p=0.69) (Table 2).

Table 1: ICSI Outcomes of Fresh versus Frozen Oocytes

n= 28 individuals	Fresh Oocytes	Frozen Oocytes	p-value
Inseminated Oocytes (n)	255	210	
Percent normal fertilizations (median, IQR)	50 (21, 68)	40 (24, 60)	0.34
Percent of good quality day 3 embryos per oocyte (median, IQR)	25 (6, 48)	24 (8,40)	0.33
Percent of good quality day 3 embryos per 2pn embryo (median, IQR)	60 (35, 79)	50 (33, 100)	>0.99

Table 2: ICSI Outcomes of Fresh versus Frozen Oocytes with TESE-NOA Sperm Source

n=22 individuals	Fresh Oocytes	Frozen Oocytes	p-value
Inseminated Oocytes (n)	207	180	
Percent of 2pn fertilizations (median, IQR)	48 (20, 57)	33 (16, 58)	0.20
Percent of good quality day 3 embryos per oocyte (median, IQR)	25 (3, 46)	21 (0,36)	0.31
Percent of good quality day 3 embryos per 2pn embryo (median, IQR)	61 (42, 85)	50 (33, 100)	0.69

Conclusions:
Patients undergoing ICSI with fresh surgically retrieved sperm have similar fresh and frozen oocyte fertilization and day 3 embryo development outcomes when assessed on a per-individual basis. While there was a trend towards better fertilization and embryo development with fresh oocytes, this did not reach statistical significance. Emphasis when managing these clinical situations should be placed on maximizing the number of oocytes available for fertilization using either fresh or frozen oocytes.

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References:
[1] Schlegel PN, Sigman M, Collura B, et al. Diagnosis and treatment of infertility in men: AUA/ASRM guideline part II. Fertil Steril. 2021;115(1):62-69.
[2] Yu Z, Wei Z, Yang J, et al. Comparison of intracytoplasmic sperm injection outcome with fresh versus frozen-thawed testicular sperm in men with nonobstructive azoospermia: a systematic review and meta-analysis. J Assist Reprod Genet. 2018;35(7):1247-1257.
[3] Liu H, Xie Y, Gao L, et al. Impact on using cryopreservation of testicular or epididymal sperm upon intracytoplasmic sperm injection outcome in men with obstructive azoospermia: a systematic review and meta-analysis. J Assist Reprod Genet. 2020;37(11):2643-2651.