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## **The Relative Viability of Human Spermatozoa from the Vas Deferens, Epididymis and Testis Before and After Cryopreservation**

N. E. Bachtell, J. Conaghan, P. J. Turek

Departments of Urology and Obstetrics, Gynecology and Reproductive Sciences,  
University of California San Francisco School of Medicine, San Francisco, California,

### **Abstract**

Testicular and epididymal sperm are routinely used with in vitro fertilization (IVF) and intracytoplasmic sperm injection (ICSI) to achieve pregnancies. In addition, excess cryopreserved sperm can be thawed and used for ICSI. However, information on the recoverability of epididymal and testicular sperm after freeze-thaw is lacking. This is important to determine the feasibility of using previously cryopreserved aspirated sperm for ICSI. We prospectively compared the viability of fresh and frozen-thawed sperm from the vas deferens, epididymis and testicle by several measures. Testis sperm was obtained from men with nonobstructive azoospermia (n=5), epididymal sperm from obstructed men (n=8), and vasal sperm by vasal irrigation at vasectomy in fertile men (n=5). The viability of fresh sperm was assessed by motility, 2 vital stains (carboxyfluorescein, 0.08mg/mL and propidium iodide, 20mg/mL) and the hypoosmotic swelling assay (HOS; 100mM citrate and fructose). After cryopreservation, sperm were thawed and all viability measures repeated. Although fresh vasal sperm is the most motile, testicular sperm exhibits similar, high viability (91% and 86%, respectively)

by vital stain. Sperm from testis, epididymis and vas deferens survive cryopreservation equally well by vital stain, but not by motility. As a selection measure, the HOS assay identifies significantly more viable epididymal and testicular sperm than does motility in both fresh and frozen-thawed populations. It appears feasible to use frozen-thawed extracted sperm for ICSI when motility and a selection measure such as the HOS assay are used. With fresh testis sperm, selection methods may not be necessary prior to ICSI, as cell viability is very high.