COMPARISON OF CREATINE KINASE ACTIVITY BETWEEN FRESH AND CRYOPRESERVED SPERM FROM NORMOZOOSPERMIC AND SUBFERTILE MEN

J. Hanak, R.K. Sharma, A.J. Thomas Jr., and A. Agarwal; Andrology Research & Clinical Laboratories, Department of Urology, Cleveland Clinic Foundation, Cleveland, OH.

Pregnancy rates are poor with cryopreserved spermatozoa compared with fresh sperm, regardless of male fertility status. Sublethal damage to the plasma membrane occurs during the freeze-thaw process, resulting in poor sperm motion and functional characteristics. In fresh spermatozoa, elevated creatine kinase (CK) activity is related to an abnormal cytoplasm level and morphological defects in sperm. This study determined whether sperm cell damage and changes in functional characteristics that occur during the freeze-thaw process are related to an inhibition or reduction in sperm cell metabolism as indicated by CK level. Semen specimens were obtained by masturbation from normal healthy volunteers (n = 10) and subfertile men (n = 19) after 2 to 3 days of sexual abstinence. Sperm concentration and motility were determined by the World Health Organization method after liquefaction. Each ejaculate was then divided into two aliquots and one aliquot of fresh semen was used to measure CK activity using a kit (Sigma Diagnostics). Results are expressed as median and interquartile range. The second aliquot was cryopreserved with TEST-yolk buffer. After 24 hours, samples were thawed for 20 minutes at 37°C in an incubator. After the cryoprotective medium was removed, percentage motility and CK were assessed in thawed specimens and compared with prefreeze values. CK levels did not significantly differ between fresh and cryopreserved specimens in donors: 0.01 units/10^8 sperm (0.01 to 0.05) vs. 0.01 units/10^8 sperm (0.01 to 0.04) (P = 0.49). In subfertile men, CK levels decreased significantly in cryopreserved specimens compared to fresh specimens: 0.06 units/10^8 sperm (0.04 to 0.24) vs. 0.06 units/10^8 sperm (0.03 to 0.15) (P = 0.01). Spermatozoa from subfertile men may be more susceptible to cryopreservation-induced damage as indicated by a reduced CK level. This may be due to the loss of cytoplasm during cryoprotectant removal before processing the specimen for CK activity.