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Journal of Andrology, Vol 21, Issue 5 700–707, Copyright  $^{\odot}$  2000 by The American Society of Andrology

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JOURNAL ARTICLE

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# Cryopreservation alters the levels of the bull sperm surface protein P25b

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Fertility of frozen-thawed bull sperm is reduced by cryopreservation. Freezing-thawing procedures can result in as much as a sevenfold fertility decrease. Sperm mortality and loss of motility do not fully explain the reduced fertility of cryopreserved semen; they may be partially explained by the loss of sperm surface proteins, which are necessary for fertilization. We have previously identified P25b, a sperm surface protein, which is associated with the fertility index of

bulls used for artificial insemination. Using Western blotting techniques, we have evaluated P25b levels before and after cryopreservation of bull spermatozoa in extenders based on either egg yolk or milk. Long storage periods (28 days) in liquid nitrogen results in a threefold decrease of P25b levels associated with cryopreserved versus fresh spermatozoa. Over a short storage period (3-7 days), a stable P25b level was observed on spermatozoa cryopreserved in extender containing either egg yolk or milk. A decrease in P25b levels associated with spermatozoa was observed after 5 days of storage in egg yolk extender, whereas a significant decrease was observed after 14 days of sperm storage in milk extender (P < .05). Therefore, the loss of P25b may be responsible, at least in part, for the decrease in fertility following the freezing-thawing procedure of bull semen. Moreover, the cryopreservation extender used may have different effects on the loss of sperm surface proteins after even brief storage periods in liquid nitrogen. Considering that a sperm protein similar to P25b exists in humans (P34H), these results may have significant clinical applications in which frozen semen is used.

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