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JOURNAL ARTICLE

Post-thaw bovine spermatozoal quality estimated from fresh samples

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Comparisons were made among flow cytometrically quantified populations of fresh and cryopreserved bovine spermatozoa, tri-stained with fluorophores rhodamine 123 (R123), 5- (and 6-) carboxy-4',5'-dimethylfluorescein diacetate (CMFDA), and propidium iodide (PI), and analyzed by dual parameter flow cytometry. The purpose was to find parameters in fresh semen samples that were potentially predictive of frozen sample parameters so that bulls with marginal cryopreservation capacity could be identified. Fresh and cryopreserved aliquots of

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semen from two sets of six bulls were semen processed in either milk (bulls 1-6) or egg yolk citrate (bulls 11-16). Membrane-damaged red (PI) and intact green (CMFDA + R123) populations were evaluated as percentages of 10,000-cell samples or numbers per straw. In milk, gated central subsets of membrane-damaged sperm cells in fresh samples and moribund cells in post-thaw samples were significantly correlated for sample percentages (r = 0.90, P = 0.014) and cell numbers per straw (r = 0.94, P = 0.006). In egg yolk citrate, fresh and frozen membrane-damaged populations were correlated (percentages: r = 0.81, P = 0.048; numbers: r = 0.88, P = 0.019). Additionally, post-thaw motility estimated by a photographic method was correlated with the number of sperm cells per straw in the intact central green subset (r = 0.98, P = 0.0006). These findings suggest that partitioning red and gree populations into smaller, central subset populations reveals significant relationships between fresh and cryopreserved bull ejaculates. The proportion of membrane-damaged spermatozoa in fresh semen seems to be predictive of the proportion of post-thaw membrane-damaged or moribund spermatozoa. The data consistently showed ejaculates of bulls 1 and 12 as having the greatest cryopreservation potentials, and bulls 4 and 16 the least. Thus, flow cytometric evaluation of fresh semen may be useful for identifying young sires with relatively poor fertilizing potential.

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