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Abstract

Determination of exact sperm concentrations in straws is necessary for standardizing protocols of fish cryopreservation. The aim of this study was to test the effect of final sperm concentration in straws on the post-thaw quality of rainbow trout semen in conditions of constant cryoprotectant concentration (0.15 M glucose and 7.5% methanol). In addition, the utility of 0.5-ml (at two different thawing times of 10 s and 13 s) and 0.25-ml straws for the cryopreservation of rainbow trout semen was tested. A shorter thawing time (10 s) of rainbow trout semen cryopreserved in 0.5-ml straws significantly improved the motility of sperm in thawed semen (62.7 ± 5.0%) compared to a 13-s thawing time (57.4 ± 9.4%). Dilution of semen to sperm concentrations of 0.5–1.0 × 10^9 spermatozoa ml^{-1} and cryopreservation in 0.5-ml straws resulted in higher sperm motility (60, 62 and 66% for 0.5; 0.75 and 1.0 × 10^9 sperm ml^{-1}, respectively) in comparison to 0.25-ml straws containing the same sperm concentrations (43, 48 and 46%, respectively). However, sperm motility was reduced to similar values in semen thawed from 0.25-ml and 0.5-ml straws containing higher final sperm concentrations of 2.0–3.0 × 10^9 spermatozoa ml^{-1}. Our results clearly demonstrate that cryopreserved sperm quality is related to final sperm concentration and straw volume. This study establishes a detailed and practical procedure for rainbow trout sperm cryopreservation using 0.5-ml straws and adjusting sperm concentrations in the straw to 0.5–1.0 × 10^9 spermatozoa ml^{-1}, while maintaining constant concentrations of cryoprotectant.

Keywords: fish, cryopreservation, sperm concentration, sperm motility