

— Cryopreservation of sea bass (



A sperm cryopreservation protocol adapted from turbot, was tested on sea bass using either 250- μ L straws or 1.5-mL cryovials. A dilution to 1/3 in Mounib's extender and a cooling rate of $-65\text{ }^{\circ}\text{C}\cdot\text{min}^{-1}$ allowed frozen sperm to recover an initial motility similar to that of fresh sperm at thawing; however, significant differences in motility ($P < 0.001$, $n = 10$ fish semen) were observed at further post-activation times, the motility decrease being faster in thawed sperm. At the experimental scale, triplicate inseminations of 2-mL aliquots (approximately $2\cdot 10^3$ eggs) showed a significant fertility decay of thawed sperm compared to that of fresh sperm ($P < 0.01$, $n = 12$ fish semen) when a discriminating $35\cdot 10^3$ spermatozoa to egg ratio was applied. When $70\cdot 10^3$ and $200\cdot 10^3$ spermatozoa per egg were provided in the same experimental conditions, no significant difference appeared between the fertilisation rates of fresh and thawed sperm. In order to validate the procedure for production or cryobank purpose, a scaled-up protocol was established. Two and 50 mL batches of eggs (approximately $2\cdot 10^3$ and $50\cdot 10^3$ eggs, respectively) were inseminated in triplicate using either

fresh or thawed individual sperms of 5 males with $200\cdot 10^3$ spermatozoa per egg. The mean fertility decreased by 23.5 % due to cryopreservation. This decline was explained by the loss of fertility of only one sperm, and only in large-volume conditions, probably due to the delay of use after thawing.

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