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Artificial insemination of African catfish (*Clarias gariepinus*) using cryopreserved sperm

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Revised highlighted

2	Artificial insemination of African catfish (Clarias gariepinus) using cryopreserved sperm
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16	
17	Abstract
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19	In this study, we aimed to develop a practical protocol for using cryopreserved sperm for
20	induced/wild/tank spawning of fish species with external fertilization. Experiments were
21	carried out on African catfish (Clarias gariepinus) as a model species. Sperm was collected
22	for cryopreservation and diluted with the cryomedium (266 mM fructose, 20% methanol,) at a
23	ratio of 1:1 with a final methanol concentration of 2.47 M pH7.73. Diluted sperm was loaded
24	into 0.5-ml straws and cryopreserved by conventional protocol. Samples were prepared for
25	insemination 24 hours later, by thawing for 13 s in a 40 °C water bath, and centrifuged at 500

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