Activation of sperm motility in the euryhaline tilapia *Sarotherodon melanotheron heudelotii* (Dumeril, 1859) acclimatized to fresh, sea and hypersaline waters

by

Marc LEGENDRE (1), Jacky COSSON (2), S.M. Hadi ALAVI (3) & Otomar LINHART (3)

ABSTRACT. - The effects of osmolality and ions were examined on motility of sperm from males of *Sarotherodon melanotheron heudelotii* acclimatized in tanks at salinities set at 0, 35 and 70 g L^{-1}. The range of osmolality that enabled sperm activation, shifted and broadened as the maintenance salinity of broodfish increased. The requirement of extracellular Ca^{2+} for activation of sperm motility increased when the maintenance salinity of broodfish was higher.

Key words. - Sperm - Motility - Tilapia - Euryhalinity - Adaptation.

Introduction

Sarotherodon melanotheron is an estuarine tilapia naturally distributed in West Africa from Senegal to Congo. In Senegal, the sub-species *S. m. heudelotii* was found to reproduce successfully at salinities ranging from *circa* 0 (Guiers Lake) up to 120 g L^{-1} (Saloum estuary). In order to better understand adaptive mechanisms enabling reproduction in such different environments, the effects of osmolality and ions were examined on motility of spermatozoa from males acclimatized to fresh (FW), sea (SW) or hypersaline (HW) waters.

Methods

Fish used descended from a Senegalese population reared in fresh water in the "GAMET" facilities since 10 years. Ten months before observations, three groups of 10 males (243-402 g) were stocked together with females in recycling water systems at salinities set at *circa* 0 (FW), 35 (SW) and 70 g L^{-1} (HW). Intratesticular sperm was collected by squeezing testes and stored in tubes on ice. Motility parameters were assessed *vs*. time after activation, using video images recorded through a microscope (dark field) equipped with a stroboscopic illuminator and a camera. Velocity and percentage of motile spermatozoa were extracted from successive video frames with an image analyzer. Motility was initiated by 1:100 sperm dilution in a drop of swimming medium on a microscope slide. The effects of osmolality and ions were tested using synthetic sea salt or sucrose solutions of different osmolalities as swimming media. Media were added with EGTA or $CaCl_2$ (1 mM) for evaluating the Ca^{2+} requirements for activation of spermatozoa.

Results and discussion

The osmolality of seminal fluid tended to be higher in HW-reared than in FW- and SW-reared males (340-360 *vs*. 305-330 mOsm kg^{-1}).

After activation in sea salt solution, the highest sperm velocities were observed at osmolalities around 300 mOsm kg^{-1} in FW males and around 700-900 mOsm kg^{-1} in both SW and HW males. Sperm velocity reached higher scores in sperm collected from FW males (60-90 μm s^{-1}) than in sperm from SW or HW males (40-45 μm s^{-1}).

The percentage of motile spermatozoa in synthetic sea salt solutions was highest (70-100%) at 1-350, 300-700 and 450-1200 mOsm kg^{-1}, for fish reared in FW, SW and HW, respectively. Sperm from FW-reared males showed a similar response in sucrose solutions. By contrast, sperm motility in sucrose was strongly depressed for SW-reared males, and almost fully depressed for HW-reared males, at all osmolalities tested. The addition of $CaCl_2$ to sucrose caused all responses to rise close to the levels observed in synthetic sea salt solutions, while full removal of Ca^{2+} with EGTA almost blocked motility. Concerning FW and SW sperm motility comparison, our results are in agreement with those previously reported for *Oreochromis mossambicus* (Linhart *et al*., 1999; Morita *et al*., 2004).

Conclusion

To our knowledge, this is the first study describing the osmolality and ions dependence of fish sperm motility for fish acclimatized to FW, SW and HW waters. The range of osmolality that enabled sperm activation, shifted and broadened as the maintenance salinity of broodfish increased. Hence, sperm motility was sufficient (even though not maximal) to ensure egg fertilization at the maintenance salinity. The requirement of extracellular Ca^{2+} for activation of sperm motility increased when the maintenance salinity of broodfish was higher.

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(1) IRD, UR 175, GAMET, BP 5095, 34196 Montpellier CEDEX 05, France. [Marc.Legendre@ird.fr]
(2) UMR7009 CNRS/UPMC, Quai de la Darse, 06234 Villefranche-sur-Mer CEDEX, France
(3) Research Institute of Fish Culture and Hydrobiology, University of South Bohemia, 389 25 Vodnany, Czech Republic.

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