

Early sex reversal during embryonic development in the Nile tilapia

by

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ABSTRACT. - The aim of this work was to study the effect of hormonal sex-reversal treatment during embryonic development of Nile tilapia. XX and XY embryos were incubated in 17α -methyltestosterone or in 17α -ethynylestradiol during embryonic development. Mean survival rates of embryos 5 days post-hatching ranged from 13.1% to 63.9%. XY embryos incubated in E2 displayed a significant proportion of females (from 50.6 to 79.0%) and XX embryos incubated in MT a significant proportion of males (from 9 to 15%). Our results proved the effectiveness of hormonal sex reversal treatment during embryonic development, as well as precocious sensitivity to hormonal treatments.

Key words. - Sex differentiation - Sex reversal - Tilapia - 17α -methyltestosterone - 17α -ethynylestradiol.

Introduction

The phenotypic sex of fish could easily be changed using exogenous sex steroids during a labile period of sex differentiation, which lasted from a few days to a few months depending on the species. In tilapia, sex reversal treatment with masculinizing or feminizing hormones incorporated into food generally induced total sex reversal. However, few data exist regarding hormonal effects on sex differentiation during embryogenesis, although early initiation of sex differentiation is suspected in the Nile tilapia and related species (Kwon *et al.*, 2001; Tsai *et al.*, 2003). The aim of this study was to test the possibility of modifying the sex differentiation process by treatment during the embryogenesis phase in Nile tilapia, *Oreochromis niloticus*.

Methods

Masculinizing treatment with 750 and 1000 mgL⁻¹ 17α -methyltestosterone (17MT) was conducted on XX embryos originating from XX female and XX male crosses. Feminizing treatment with 100 and 500 mgL⁻¹ 17α -ethynylestradiol (E2) was conducted on XY embryos originating from XX female and YY male crosses. Freshly fertilized eggs (< 12 h) were distributed into 4 batches : 1 control batch incubated in water; 1 batch incubated in 0.5% ethanol solution and 2 batches incubated in hormonal solution prepared by dilution of hormones in ethanol. The experiment covered the period of embryonic development (5 days at 27°C). Sex ratio was determined on samples of 100 3-month old fish (MBW = 3 g) per batch, using the aceto-carmin squash method.

Results and discussion

Under E2 treatment, mean survival rates of embryos 5 days post-hatching ranged from 13.1% to 63.9% (Tab. I). The MT treatment also induced a significant ($p < 0.05$) decrease in

survival rates from 53.2% to 31.5%. Incubation of XY eggs in E2 significantly skewed sex ratios in favour of females (from 50.6 to 79.0%), whereas incubation of XX eggs into MT significantly skewed sex ratios in favour of males (from 9.0 to 20.0%). Our results confirmed the possibility of hormonally controlling and modifying the sex differentiation pathway during embryonic development, before hatching. Similar results were obtained after incubation of XX embryos at 36°C during embryogenesis, resulting in a sex reversal rate of up to 20% (Rougeot *et al.*, 2008). These results support the hypothesis that sexual differentiation in fish may occur during the early embryonic stages (Kwon *et al.*, 2001, Tsai *et al.*, 2003).

Conclusion

Our results prove that sexual differentiation in fish may occur during the early embryonic stage, before the apparition of the gonads. It could be hypothesised that there is a hormonal influence on the development of primordial germ cells and/or on the future somatic cells of the presumptive gonads; or that other organs, such as the brain, play a role in the process of sex determination.

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Table I. - Survival rates of 5 days post-hatching tilapia embryos and sex ratio of the resulting progenies (90 days post-hatching) incubated in MT and E2 during the embryonic development. * significantly different from the control, $p < 0.05$.

		Mean survival \pm SE	% males	% females
Feminization E2 (μgL^{-1})	Water	63.9 \pm 10.5	100	0
	Ethanol	55.2 \pm 4.9	100	0
	100	24.6 \pm 0.7	44.5 \pm 4.3	55.5 \pm 4.3*
	500	13.1 \pm 3.7	31.8 \pm 9.2	68.2 \pm 9.6*
Masculinization MT (μgL^{-1})	Water	53.2 \pm 2.9	1.3 \pm 1.3	98.7 \pm 1.3
	Ethanol	29.2 \pm 8.2	3.5 \pm 3.0	96.5 \pm 3.0
	750	44.3 \pm 7.0	11.3 \pm 3.2	88.7 \pm 3.2*
	1000	31.5 \pm 7.1	18.1 \pm 1.7	81.9 \pm 1.7*

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