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Sex differences in the expression of GH/IGF axis genes underlie sexual size dimorphism in the yellow catfish (*Pelteobagrus fulvidraco*)

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Dear Editors,

Sexual dimorphism is the systematic difference in size, shape, color, physiology, and behavior, between male and female individuals of the same species (Mei and Gui, 2015). Some studies have indicated that the traits of sexual dimorphism in vertebrates are the consequences of sex-biased gene expression and are controlled by multiple critical genes during growth and development (Williams and Carroll, 2009). However, the exact molecular mechanism underlying sexual dimorphism remains unclear. Sexual dimorphism with respect to size has been observed in many farmed fish species, such as the yellow catfish, *Pelteobagrus fulvidraco* (Gui and Zhu, 2012). In teleosts, somatic growth is greatly regulated by the growth hormone (GH)/insulin-like growth factor (IGF) axis genes expressed in the hypothalamus-pituitary-gonad (HPG) axis (Dai et al., 2015). Yellow catfish males grow much faster than the females. Therefore, we cloned the GH/IGF axis genes, *GH*, *IGF-1*, and *IGF-2*, from the transcriptome of yellow catfish (Chen et al., 2015) and characterized their different expressions in males and females.

Quantitative real-time (qRT)-PCR was performed to determine the tissue distributions of *GH*, *IGF-1*, and *IGF-2* in the HPG axis and liver of adult yellow catfish. *GH*, *IGF-1*, *IGF-2* were predominantly expressed in the hypothalamus and pituitary, hypothalamus and liver, pituitary and liver, respectively. Significantly, higher expressions of *GH*, *IGF-1*, and *IGF-2* were observed in the main tissues of males than those of females (Figures 1A–C). To assess whether the GH/IGF axis genes are involved in the early stage of body growth, expression patterns of *GH*, *IGF-1*, and *IGF-2* during larval growth were investigated. In male larval fish, expressions of both *GH* and *IGF-1* gradually increased between 1 and 4 weeks post-hatching (wph). However, we did not detect obviously increasing trends in *GH* and *IGF-1* expressions in female larval fish between 1 and 3 wph (Figures 1D–E). Expression of *IGF-2* gradually increased between 1 and 3 wph and slightly decreased at 4 wph in male larval fish, whereas an inconsistent expression pattern was observed in female larval fish (Figure 1F). Interestingly, expressions of *GH*, *IGF-1*, and *IGF-2* were significantly higher in male fish than in female fish during larval growth (Figures 1D–F).

17-Alpha-methyltestosterone (MT) treatment has been shown to activate the GH/IGF axis genes and enhance somatic growth in tilapia (Riley et al., 2002). The direct effect of MT on the mRNA expressions of *GH*, *IGF-1*, and *IGF-2* was investigated in yellow catfish. Larval fish at 4 days post-hatching were treated with MT, genotyped using

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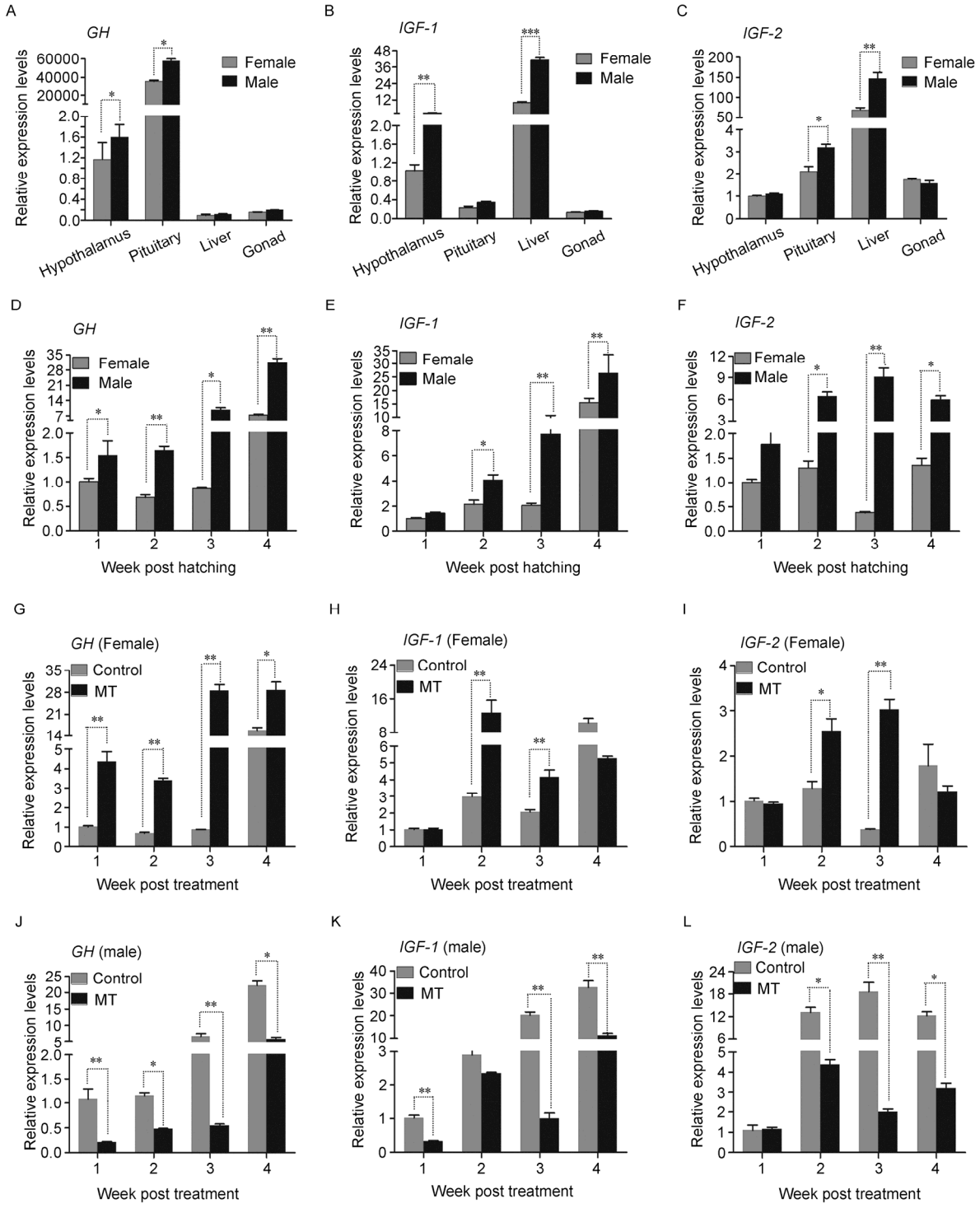

Figure 1 Sexual dimorphic expression of GH/IGF axis genes and their responses to sex hormone treatment. A–C, Expressions of *GH*, *IGF-1*, and *IGF-2* genes in the tissues of adult yellow catfish. D–F, Expressions of *GH*, *IGF-1*, and *IGF-2* genes during larval growth in male and female yellow catfish. G–I and J–L represent the effect of MT on *GH*, *IGF-1*, and *IGF-2* expressions in females and males, respectively. β-Actin was used as the internal control. Significant differences between the two groups are indicated by *(P<0.05) and **(P<0.01).

sex-linked markers, and then collected for RT-PCR analysis. In female larval fish, MT treatment activated the expression levels of GH/IGF axis genes (Figures 1G–I). *GH* was activated at 1 week post-treatment (wpt) and increased at 3 wpt (Figure 1G). *IGF-1* was activated at 2 wpt, decreased a little at 3 wpt, and then remained at a uniform

level at 4 wpt (Figure 1H). *IGF-2* was activated at 2 wpt, consistently increased at 3 wpt, and then decreased at 4 wpt (Figure 1I). Moreover, somatic growth (body weight and body length) in female larvae was increased after MT treatment (data not shown). In male larval fish, both *GH* and *IGF-1* gradually increased in the control groups during larval development (Figures 1J–K). Expression of *GH* was much lower in the MT-treated group than in the control group at all stages, although it gradually increased in the MT-treated group (Figure 1J). Expression of *IGF-1* was reduced by MT treatment, whereas an inconsistent expression pattern was observed in the MT-treated group (Figure 1K). Expression of *IGF-2* was not reduced by MT at 1 wpt, while it was significantly reduced between 2 and 4 wpt (Figure 1L). Our results suggest that the GH/IGF axis genes are activated by MT in female larval fish, but reduced by MT in male larval fish.

In a previous study, 17-alpha-ethinyl estradiol (EE2) treatment significantly reduced the growth of yellow catfish, whereas MT had no obvious effect on average body weight and body length (Jiang, 2007). We observed that MT promoted somatic growth in female larval fish and reduced somatic growth in male larval fish. Overdose of androgens in male larval fish may change the expressions of some transcriptional factors that inhibit the expressions of GH/IGF axis genes. The sexual dimorphic response of GH/IGF axis gene expressions could explain why MT has no obvious effect on the body growth of yellow catfish.

In conclusion, expressions of *GH*, *IGF-1*, and *IGF-2* were significantly higher in males than in females in both larval and adult yellow catfish. MT treatment activated the expressions of GH/IGF axis genes in female larval fish and reduced their expressions in male larval fish. These results provide evidence that key endocrine genes of GH/IGF axis are involved in the sexual size dimorphism of yellow catfish.

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