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The expressions of two myostatin paralogs in turbot Scophthalmus maximus fasted for two weeks and fed diets containing graded levels of fish protein hydrolysate

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Introduction

Myostatin (MSTN) is the negative regulator of muscle growth. There are two MSTN paralogs (MSTNa and MSTNb) in teleost fish, but their regulatory roles in muscle growth are still largely unclear. To clarify the role of each of these paralogs, mRNA levels of MSTNa and MSTNb were quantified in different tissues of turbot *Scophthalmus maximus* and in the fasting-re-feeding experiment.

Materials and Methods

Turbot (average body weight 28 g) were fed a commercial feed for one week, and then fasted for two weeks. After that, fish were re-fed three experimental diets for one week. Muscle samples were collected at 3 h, 6 h, 24 h, 3 days and 7 days after re-feeding. Three isoproteic (500 g/kg crude protein) and isolipidic (120 g/ kg crude lipid) diets were formulated to contain graded levels of fish protein hydrolysate (Tab. 2). FM, FPH8 and FPH32 diets were prepared by supplementing with 0, 80 and 320 g/kg fish protein hydrolysate.

Tab. 2 Formulation of experimental diets

Ingredient	FM	FPH8	FPH32
Fish meal	35	26.5	0
Soybean meal	16	16	16
Wheat gluten	5	5	5
Corn gluten meal	10	10	10
Fish protein hydrolysate	0	8	32
Wheat meal	24.3	24	24.1
Fish oil	2.5	3.3	5.7
Soybean oil	2.5	2.5	2.5
Soybean lecithin	1.5	1.5	1.5
Yttrium oxide	0.1	0.1	0.1
Mineral premix	0.5	0.5	0.5
Vitamin premix	1	1	1
Ca(H2PO4)2	1	1	1
Choline chloride	0.4	0.4	0.4
Vitamin C	0.2	0.2	0.2

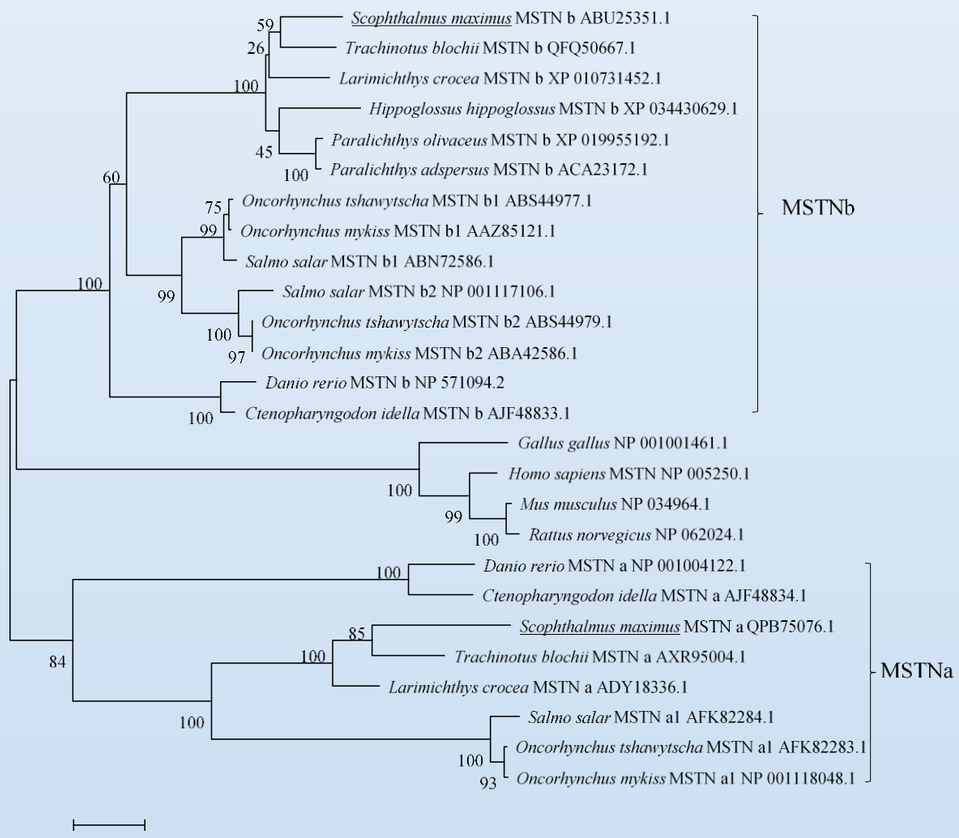

Fig. 1 Phylogenetic tree analysis based on the amino acid sequence of animal MSTN.

Tab.1 Primer sequences used for qRT-PCR.

Gene	Primer sequence (5′to 3′)	Amplicon size (bp)	Accession number
MSTNa	F-TGACCGCTAAGCTGTGG R-GAGAGCTGCAGGAAGACA	232	EF683115.1
MSTNb	F-ACTGCGAATGAAAGAAGC R- TCCTCCATAACTACATCCCT	135	MT925725.1
MSTN2	F-AAACTGCGAATGAAAGAA R-ACATCCCTGTTGTCATCTC	125	EF683115.1
β-actin	F- CCAAAGCCAACAGGGAGAA R- AGAGGCATACAGGGACAGCACA	101	AY008305.1

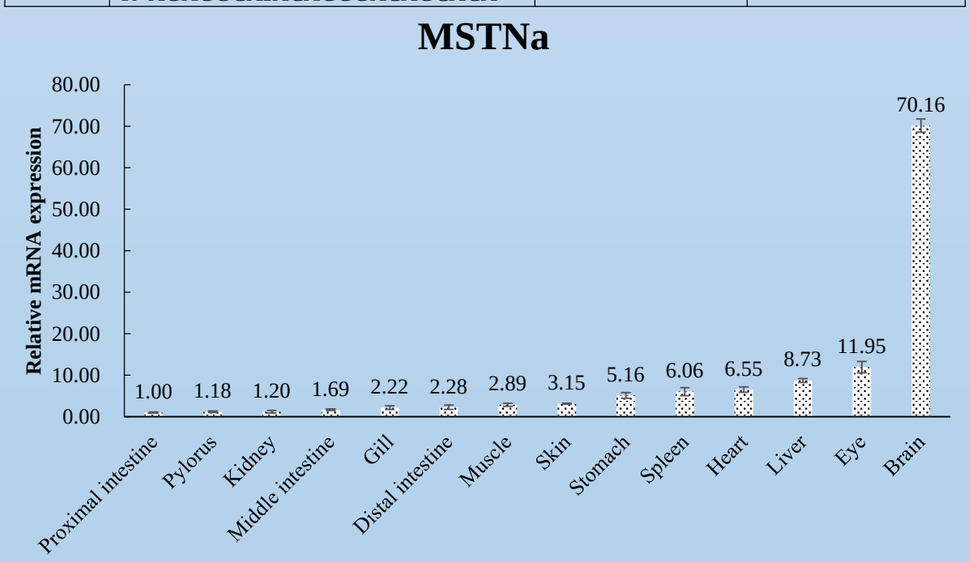

Fig. 2 Tissue distribution of MSTNa expression in turbot (n=12).

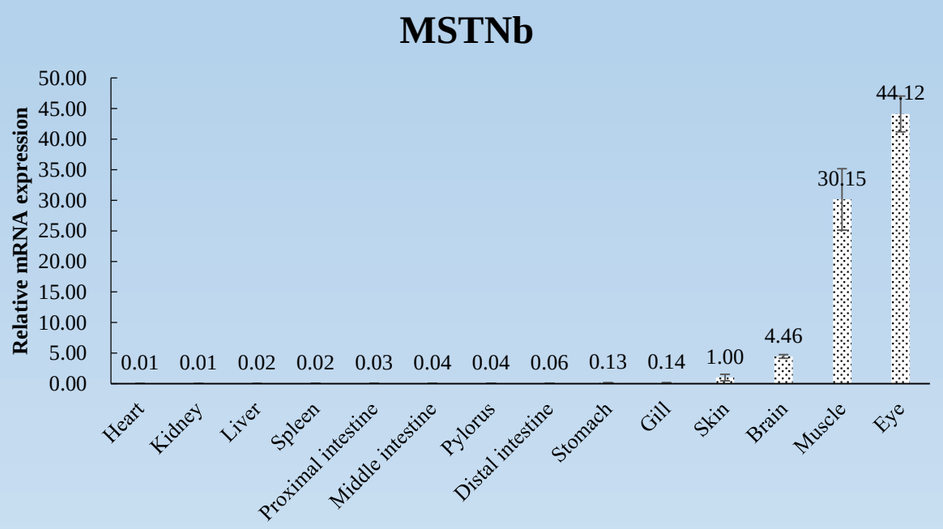

Fig. 3 Tissue distribution of MSTNb expression in turbot (n=12).

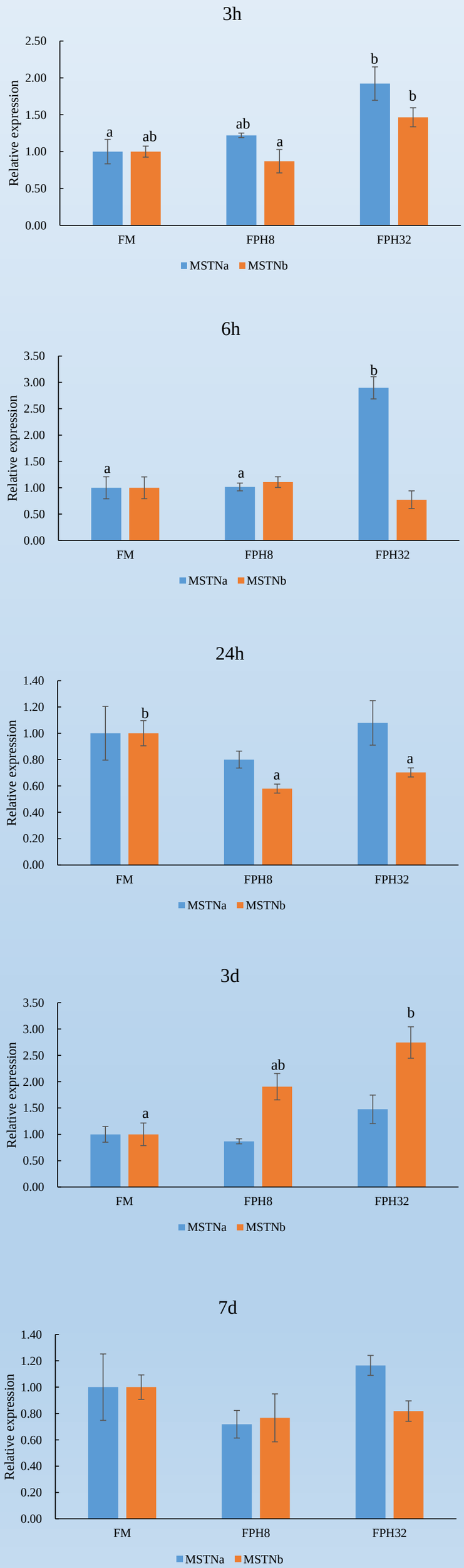

Fig. 4 Relative expression of MSTNa and MSTNb in the muscle at different times postprandially after re-feeding experimental diets.

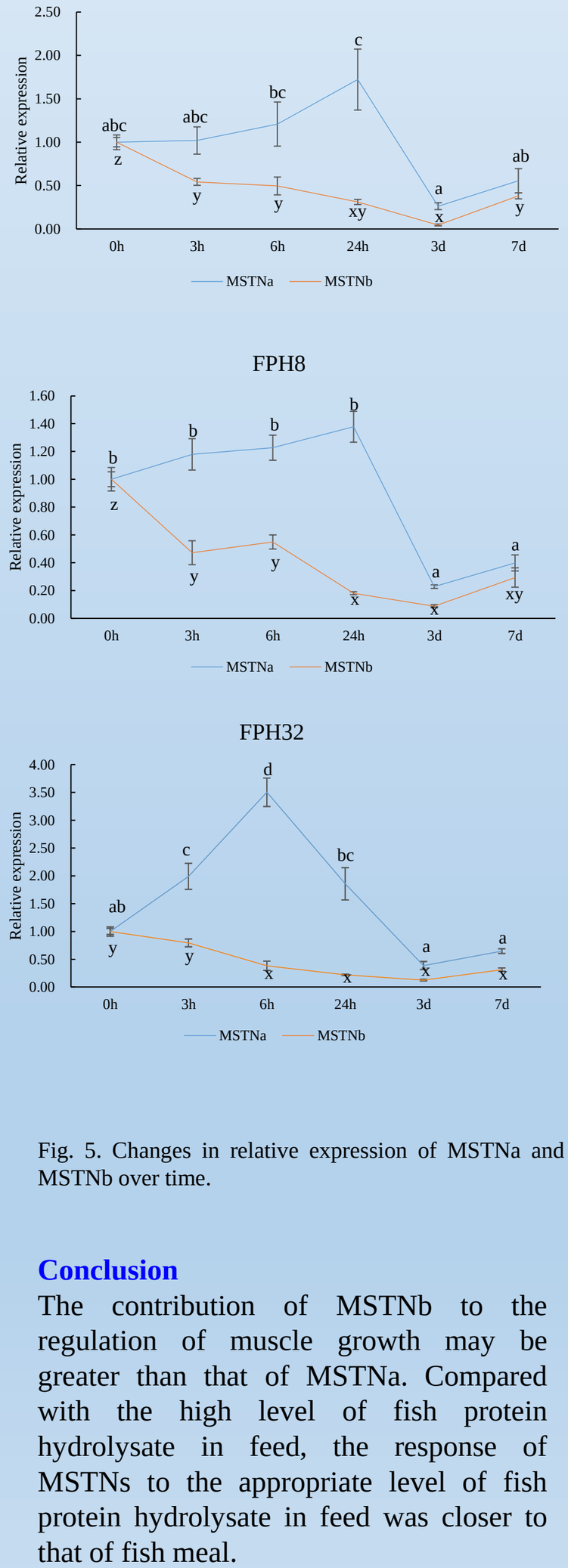

Fig. 5. Changes in relative expression of MSTNa and MSTNb over time.

Conclusion

The contribution of MSTNb to the regulation of muscle growth may be greater than that of MSTNa. Compared with the high level of fish protein hydrolysate in feed, the response of MSTNs to the appropriate level of fish protein hydrolysate in feed was closer to that of fish meal.

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