



Structure and immune expression analysis of hemoglobin genes from the blood clam *Tegillarca granosa*

Y.B. Bao¹, Q. Wang¹, X.M. Guo² and Z.H. Lin¹

¹College of Biological and Environmental Sciences,
Zhejiang Wanli University, Ningbo, Zhejiang, China

²Haskin Shellfish Research Laboratory,
Institute of Marine and Coastal Sciences, Rutgers University,
Port Norris, NJ, USA

Corresponding authors: Z.H. Lin / Y.B. Bao
E-mail: zhihua9988@126.com / bobbao2001@gmail.com

Genet. Mol. Res. 12 (3): 3110-3123 (2013)
Received May 26, 2012
Accepted November 10, 2012
Published February 28, 2013
DOI <http://dx.doi.org/10.4238/2013.February.28.5>

ABSTRACT. Hemoglobin (Hb) is the major protein component of erythrocytes in animals with red blood, although it can serve additional functions beyond the transport of oxygen. The blood clam (*Tegillarca granosa*) is one of the few mollusks that has Hb, although the structure and function of molluscan Hbs remain unclear. We characterized two unique and highly compartmentalized blood clam hemoglobin genes, *Tg-HbIIA* and *Tg-HbIIB*, at the molecular level. The full-length cDNA of *Tg-HbIIA* was 731 bp with a 450-bp open reading frame encoding 150 amino acids; that of *Tg-HbIIB* was 698 bp, with a 456-bp open reading frame encoding 152 amino acids. Their intronic regions were amplified by PCR. The two genes showed the typical 2 intron/3 exon organization found in *T. granosa*. The 3-D structures of the three blood clam Tg-Hbs were predicted using the SWISS-MODEL Protein Modeling Server, and a phylogenetic analysis was conducted to investigate its evolution. As quantified by qRT-PCR, the expression levels of *Tg-HbIIA* and *Tg-HbIIB* were significantly upregulated upon challenge by *Vibrio*

parahaemolyticus, lipopolysaccharides, and peptidoglycans. Three Hb isoforms, *Tg-HbI*, *Tg-HbIIA*, and *Tg-HbIIB*, were found. Specific structures and evolutionary features were found in these molluscan Hb genes. Challenge experiments indicated that Tg-Hbs are involved in immune defense responses against bacterial infection and bacterial pathogenic factors. As this is the first functional research on Hb genes in the blood clam, our findings provide new insight into the innate immune defense mechanisms of *T. granosa*.

Key words: Hemoglobins; *Tegillarca granosa*; Gene structure; Immune response; qRT-PCR