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Cloning and sequence analysis of the coding sequence of *β-actin* cDNA from the Chinese alligator and suitable internal reference primers from the *β-actin* gene

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ABSTRACT. *β-Actin* is an essential component of the cytoskeleton and is stably expressed in various tissues of animals, thus, it is commonly used as an internal reference for gene expression studies. In this study, a 1731-bp fragment of *β-actin* cDNA from *Alligator sinensis* was obtained using the homology cloning technique. Sequence analysis showed that this fragment contained the complete coding sequence of the *β-actin* gene (1128 bp), encoding 375 amino acids. The amino acid sequence of *β-actin* is highly conserved and its nucleotide sequence is slightly variable. Multiple alignment analyses showed that the nucleotide sequence of the *β-actin* gene from *A. sinensis* is very similar to sequences from birds, with 94-95% identity. Ten pairs of primers with different product sizes and different annealing temperatures were screened by PCR amplification, agarose gel electrophoresis, and DNA sequencing, and could be used as internal reference primers in gene expression studies. This study expands our knowledge of *β-actin* gene

phylogenetic evolution and provides a basis for quantitative gene expression studies in *A. sinensis*.

Key words: *Alligator sinensis*; β-actin; cDNA cloning; Sequence analysis; Internal reference

