

## Secondary Structure and Phylogenetic Utility of the Ribosomal Internal Transcribed Spacer 2 (ITS2) in Scleractinian Corals

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**Chaolun Allen Chen, Chau-Ching Chang, Nuwei Vivian Wei, Chien-Hsun Chen, Yi-Ting Lein, Ho-E Lin, Chang-Feng Dai and Carden C. Wallace (2004)** Secondary structure and phylogenetic utility of the ribosomal internal transcribed spacer 2 (ITS2) in scleractinian corals. *Zoological Studies* 43(4): 759-771. In this study, we examined the nucleotide characteristics, the secondary structure, and phylogenetic utility of the ribosomal internal spacer 2 (ITS2) from 54 species of scleractinian corals, representing 25 genera and 11 families of both the complex and robust clades previously defined through molecular phylogenetic analyses. The lengths and nucleotide contents of the ITS2 were highly variable among corals. The ITS2 of *Acropora* is significantly shorter than those of other corals. Dinucleotide or tetranucleotide microsatellites were identified in the genera *Acropora*, *Cyphastrea*, *Favites*, *Goniastrea*, *Hydnophora*, *Montipora*, *Madracis*, and *Porites*. Three distinct types of secondary structures with the smallest free energy values were predicted using the computer software, Mfold. A standard 4 domains were observed in 17 species of corals, while 23 species has a modified 5 domains with domain I divided into 2 subdomains. These 2 types of secondary structures were observed across 11 coral families. The 3rd type, 5 domains with domain III divided into 2 subdomains, was only seen in the genus *Acropora*. Among the domains, domain II is highly conserved and is flanked by conserved sequence motifs in adjacent stems. The motif, 5'-CRCGGYC-3', and its compensatory bases were highly conserved in both the complex and robust clades of scleractinian corals. The robust-clade phylogeny constructed using ITS2 data produced a concordant tree to those based on mitochondrial and nuclear genes. The comparative analysis indicated that the extremely high ITS intragenomic divergence of *Acropora* is an exception rather than the rule for the evolutionary history of scleractinian corals. Despite the atypical and unusual pattern of molecular evolution in the genus *Acropora*, data of the ITS2 are still applicable, with adequate adjustment of secondary structures, to the primary sequence alignment of different levels of phylogenetic analyses, from populations to genera, in scleractinian corals. <http://www.sinica.edu.tw/zool/zoolstud/43.4/759.pdf>

**Key words:** Internal transcribed spacer 2, Secondary structure, Conserved domain, Scleractinian corals, Phylogenetic utility.

DNA sequences of the 2 internal transcribed spacers (ITS1 and ITS2) of the ribosomal RNA (rRNA) transcription unit have proven useful in resolving phylogenetic relationships of closely related taxa and in distinguishing species in fungal, plant, and animal taxa due to their relatively rapid evolution rates (Baldwin 1992, Schlotterer et al. 1994, Mai and Coleman 1997, Weekers et al. 2001, Oliverio et al. 2002). In addition, the tran-

script folding structure of the ITS provides some signals that guide the ribosomal coding regions when they are processed into small, 5.8S, and large ribosomal RNA (van der Sande et al. 1992, van Nues et al. 1995). The potential to predict the folding structure has enhanced the role of ITS in phylogenetic studies, since it is important to guide reliable sequence alignment based on secondary structures (Michot et al. 1999).

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