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The complete mitochondrial genome of the Epaulette Shark, *Hemiscyllium ocellatum* (Bonnaterre, 1788)

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**ABSTRACT**

We present the complete mitochondrial genome of the Epaulette Shark *Hemiscyllium ocellatum*, sequenced with 24 primer sets. The 16,728 bp long circular genome consisted of 13 protein-coding genes, 22 tRNA genes, 2 rRNA genes, and a non-coding control region. Several protein-coding genes ended with incomplete stop codons (TA- or T-). Phylogenetic analysis indicated that *H. ocellatum* clusters within the Hemiscylliidae clade but separately from members of its sister genus *Chiloscyllium*. Utilization of the genome and primer sets presented here will be beneficial to future molecular studies involving *H. ocellatum* and other members of the Orectolobiformes.

The Epaulette Shark (*Hemiscyllium ocellatum*) is a small, benthic, oviparous shark inhabiting coral reef environments in north-eastern Australia (Allen et al. 2016). Molecular phylogeny studies involving this species are limited and often based on a single genetic marker (e.g., Naylor et al. 2012; Allen et al. 2013). As whole mitochondrial genomes can improve the resolution of elasmobranch phylogenies (Chen et al. 2013), we sequenced the complete mitochondrial genome of *H. ocellatum* to determine its phylogenetic placement within the Orectolobiformes.

DNA was extracted from a fin clip of an individual collected on Arlington Reef, Queensland (16°43′01″S; 146°01′58″E) as outlined in Geraghty et al. (2013). This sample (010 ES Skin) is currently stored at the Department of Biological Sciences, Macquarie University in 70% ethanol. Mitochondrial genome fragments were amplified using previously published and newly developed primer sets (see Supplementary Material available at https://doi.org/10.6084/m9.figshare.7300406). Forward and reverse reads of each fragment were aligned, trimmed, and checked for nucleotide assignment errors in Geneious® version 10.2.6 (http://www.geneious.com, Kearse et al. 2012). Edited fragments were mapped to the complete mitochondrial genome of *Chiloscyllium griseum* (GenBank: JQ434458) to ensure complete coverage and then de novo assembled. The resulting sequence was annotated using the MitoAnnotator tool on the MitoFish website (Iwasaki et al. 2013), and checked with tRNAscan-SE (Lowe and Chan 2016) and by comparison with annotated hemiscyllid mitochondrial genomes from GenBank.

To assess the phylogenetic position of *H. ocellatum*, a maximum likelihood (ML) tree was generated in MEGA7 (Kumar et al. 2016). Ten complete mitochondrial genomes, consisting of eight orectolobiforms and two heterodontiforms, were downloaded from GenBank and MUSCLE aligned (Edgar 2004) with *H. ocellatum*. The ML tree was constructed with the most appropriate substitution model (GTR + I + G) as indicated by the corrected Akaike Information Criterion (AICc) in jModelTest2 (Guindon and Gascuel 2003; Darriba et al. 2012), 1000 bootstrap replications, the partial deletion setting, and all codon positions included.

The mitochondrial genome of *H. ocellatum* (GenBank: MK052932) was 16,728 bp in length and consisted of 13 protein-coding genes (PCGs), 22 tRNA genes, 2 rRNA genes, and a non-coding control region (D-loop). Base composition was: A (33%), C (24%), G (12.8%), and T (30.2%). The COI gene was the only PCG that started with a GTG codon; all other PCGs ended with incomplete stop codons (TA- or T-). Phylogenetic analysis indicated that *H. ocellatum* clusters within the Hemiscylliidae clade but separately from members of its sister genus *Chiloscyllium*. Utilization of the genome and primer sets presented here will be beneficial to future molecular studies involving both *H. ocellatum* and other sharks in the order Orectolobiformes.
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Disclosure statement

The authors report no conflict of interest. The authors alone are responsible for the content and writing of the paper.

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Figure 1. Maximum likelihood tree showing the phylogenetic position of Hemiscyllium ocellatum (indicated by the star) within the order Orectolobiformes based on the complete mitochondrial genome. Members of the order Heterodontiformes served as the outgroup. Families are indicated by the vertical lines and orders by the square brackets. Scale bar indicates the number of substitutions per site and the numbers at the nodes indicate the percentage bootstrap values based on 1000 replications. GenBank Accession Numbers: Chiloscyllium plagiosum (FJ853422); C. punctatum (JQ082337); C. griseum (JQ434458); Stegostoma fasciatum (KU057952); Rhincodon typus (KF679782); Ginglymostoma cirratum (KU904394); Nebrius ferrugineus (KT852575); Orectolobus japonicus (KF111729); Heterodontus zebra (KC845548); H. francisci (NC_003137).