## Additional File 1: Mitochondrial genome of Sinularia cf. cruciata.

## Methods

The complete mitochondrial genome of Sinularia cf. cruciata was obtained following standard procedures, which included PCR amplification of overlapped fragments (up to 3 kb) and Sanger sequencing. Mitochondrial DNA fragments were amplified using previously published primers sets and PCR protocols [1-3]. Amplicons of different sizes (up to 3kb) were purified to remove unincorporated primers and were directly sequenced using BigDye® Terminator v3.1 chemistries on an ABI 3730 DNA Analyzer (Applied Biosystems, Foster City, CA, USA). Sequences quality was assessed by visually checking the chromatograms. Mitogenome was assembled using Sinularia peculiaris (Accession number: JX023274) as reference in Geneious 8.1.7 [4]. A preliminary screening of the coding-protein regions was carried using the "ORFs finder" tool implemented in Geneious and the resulting annotations were further compared by similarity with those of other octocoral mitogenomes. Detection of control region repeats and tRNAs was performed using Tandem Repeats Finder [5] and tRNA-scan SE [6], respectively. Nucleotide diversity  $(\pi)$  between Sinularia cf. cruciata and Sinularia peculiaris was calculated in DnaSP 5 [7] using a sliding window of 500 bp (25 bp steps). For this analysis neither nucleotide ambiguities nor sites with gaps were considered. A graphical map including comparative mitogenomic information between the two Sinularia species was inferred with the CGView Server [8].

## **Results**

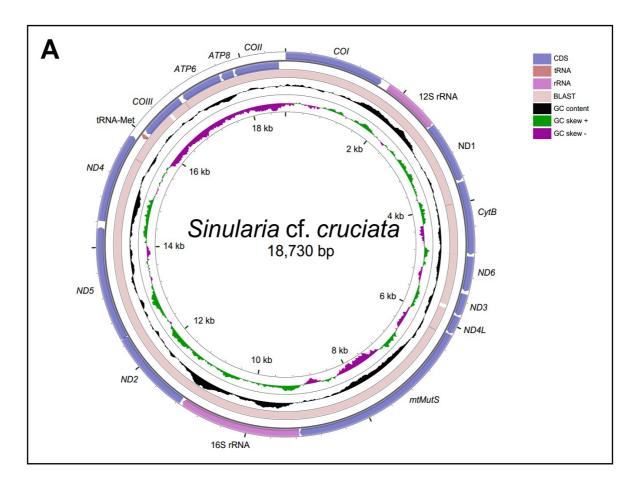
 Table 1

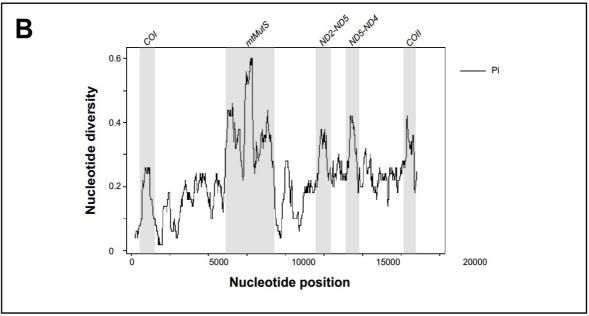
 Mitochondrial genome organisation of Sinularia ef. cruciata.

Species	Gene	Feature	Position		Codon		Strand	Lenght (bp)	Intergenic Region
_			Start	Stop	Start	Stop	<del>_</del>		_
	COI	CDS	1	1590	ATG	T	Н	1582	0
	12S	rRNA	1583	2673			Н	925	4
	ND1	CDS	2678	3649	ATG	TAG	Н	972	27
	CytB	CDS	3677	4843	ATG	TAA	Н	1167	30
	ND6	CDS	4874	5431	ATG	TAG	Н	558	44
	ND3	CDS	5476	5829	ATG	TAG	Н	354	19
	ND4L	CDS	5849	6142	ATG	TAA	Н	294	13
	mtMutS	CDS	6156	9137	ATG	TAA	Н	2982	9
Sinularia cf. cruciata	16S	rRNA	9147	11114			Н	1968	31
	ND2	CDS	11146	12519	ATG	TAG	Н	1374	-13
	ND5	CDS	12507	14324	ATG	TAA	Н	1818	97
	ND4	CDS	14422	15870	ATG	TAA	Н	1449	56
	tRNA-Met	tRNA	15927	15997			L	71	39
	COIII	CDS	16037	16822	ATG	TAG	L	786	64
	ATP6	CDS	16887	17594	ATG	TAA	L	708	24
	ATP8	CDS	17619	17834	ATG	TAA	L	216	22
	COII	CDS	17857	18618	ATG	TAA	L	762	112

**Figure 1.** Complete mitogenome of *Sinularia* cf. *cruciata* and genomic comparisons with *Sinularia peculiaris*.

- **(A)** Graphical view of the mitochondrial genome of *Sinularia* cf. *cruciata* with genome size and gene annotations. GC content is shown in black, GC skew is plotted for the entire sequence in green (GC skew +) and purple (GC skew -). The inner pink ring shows the BLAST hit detected by the *blastn* search against *Sinularia peculiaris* mitogenome.
- **(B)** Sliding window analysis of the complete mitogenomes of *Sinularia* cf. *cruciata* and *Sinularia peculiaris*. Nucleotide diversity across the genome is shown by the black line in a window of 500 bp (25 bp steps). Grey panels show the most variable regions across the two *Sinularia* species.





## References

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