RAD sequencing enables unprecedented phylogenetic resolution and objective species delimitation in recalcitrant divergent taxa

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ABSTRACT

Species delimitation is problematic in many taxa due to the difficulty of evaluating predictions from species delimitation hypotheses, which chiefly relay on subjective interpretations of morphological observations and/or DNA sequence data. This problem is exacerbated in recalcitrant taxa for which genetic resources are scarce and inadequate to resolve questions regarding evolutionary relationships and uniqueness. In this case study we demonstrate the empirical utility of restriction site associated DNA sequencing (RAD-seq) by unambiguously resolving phylogenetic relationships among recalcitrant octocoral taxa with divergences greater than 80 million years. We objectively infer robust species boundaries in the genus *Paragorgia,* which contains some of the most important ecosystem engineers in the deep-sea, by testing alternative taxonomy-guided or unguided species delimitation hypotheses using the Bayes factors delimitation method (BFD*) with genome-wide single nucleotide polymorphism data. We present conclusive evidence rejecting the current morphological species delimitation model for the genus *Paragorgia* and indicating the presence of cryptic species boundaries associated with environmental variables. We argue that the suitability limits of RAD-seq for phylogenetic inferences in divergent taxa cannot be assessed in terms of absolute time, but depend on taxon-specific factors such as mutation rate, generation time and effective population size. We show that classic morphological taxonomy can greatly benefit from integrative approaches that provide objective tests to species delimitation hypothesis. Our results pave the way for addressing further questions in biogeography, species ranges, community ecology, population dynamics, conservation, and evolution in octocorals and other marine taxa.

INTRODUCTION

Species delimitation is problematic in many taxa due to the difficulty of evaluating predictions from species delimitation hypotheses derived using different species concepts. Species concepts set particular expectations of the properties used to support species delimitations (De Queiroz 2007). For example, the classic biological species concept requires intrinsic reproductive isolation between heterospecific organisms and interbreeding among homospecific organisms resulting in viable and fertile descendants (Mayr 1942; Dobzhansky 1970). In many cases, if not the majority, it is difficult to evaluate behavioral, reproductive, and ecological properties due to technical limitations of field or laboratory work, which largely determine the kind of observations and data that can be obtained. In these cases researchers conventionally rely on morphological observations and/or DNA sequence data to generate species delimitation hypotheses.

Although there have been significant attempts at developing statistical methods to objectively identify species-diagnostic morphological discontinuities (e.g., Zapata & Jimenez 2012), most species delimitations continue to be performed subjectively based on assessments made by specialized taxonomists. Molecular phylogenetic analyses of DNA sequences provide an independent way to test these species delimitation hypotheses utilizing a variety of methods, ranging from variability thresholds of barcode sequences (Hebert *et al.* 2003), to probabilistic coalescent-based model methods (Pons *et al.* 2006; Yang & Rannala 2010; Fujisawa & Barraclough 2013; Grummer *et al.* 2014). These molecular methods rely on informative DNA sequence markers, and in many cases on resolved phylogenies.

The sub-class Octocorallia (Phylum Cnidaria), which includes animals known as gorgonians, sea pens, and soft corals, is an example of a recalcitrant group where species delimitations are problematic. Octocorals are predominantly a deep-sea group (Cairns 2007; Roberts & Cairns 2014) and therefore are extremely difficult to observe and collect. Classic morphology-based species delimitation and identification in this group is arduous for non-specialists, and challenging to replicate among taxonomists (Daly *et al.* 2007; McFadden *et al.* 2010b). Variations in octocoral colony architecture and micro-skeletal structures – sclerites – are used as species diagnostic characters (Bayer 1956). However, studies over the last 15 years have shown that in many cases species delimitations and systematics based on these morphological traits keep little to no correspondence with the patterns of genetic diversity and relatedness inferred using mitochondrial and ribosomal DNA sequence markers (McFadden *et al.* 2006; Clark *et al.* 2007; France 2007; Dueñas & Sánchez 2009). A confounding factor when analyzing mitochondrial DNA markers is the fact that anthozoans, including octocorals, have slow rates of sequence evolution relative to other metazoans (Shearer *et al.* 2002; Hellberg 2006). Furthermore, octocoral mitochondrion is unique among eukaryotes by having a functional DNA mismatch repair gene — *mtMutS* — which presumably is responsible for the extremely low sequence variability observed in this group (Bilewitch & Degnan 2011). Traditional molecular markers have thus been remarkably insufficient to resolve relationships at all taxonomic levels within the octocorals (Berntson *et al.* 2001; France *et al.* 2002; Mcfadden *et al.* 2004; Smith *et al.* 2004; Thoma *et al.* 2009; Dueñas *et al.* 2014). Alternative nuclear markers, such as the ITS2 and *SRP54* have been used to examine interspecific and intraspecific relationships (Aguilar & Sánchez 2007; Concepcion *et al.* 2007; Grajales *et al.* 2007; Herrera *et al.* 2010); however, their application and impact has been limited due to issues regarding intragenomic variability (Sanchez & Dorado 2008) and low sequencing reliability (Mcfadden *et al.* 2010a). These long-standing technical problems have caused fundamental questions in octocorals regarding species differentiation, systematics, diversity, biogeography, and species ranges to remain unanswered.

Technological developments in next-generation sequencing platforms and library preparation methodologies have made genomic resources increasingly accessible and available for the study of non-model organisms, thus offering a great opportunity to overcome the difficulties inherent to the use of traditional sequencing approaches. One of these methodologies is restriction-site-associated DNA sequencing (RAD-seq), which combines enzymatic fragmentation of genomic DNA with high-throughput sequencing for the generation of large numbers of markers (Baird *et al.* 2008). RAD-seq has shown great promise to resolve difficult phylogenetic, phylogeographic, and species delimitation questions in diverse groups of eukaryotes (Emerson *et al.* 2010; Nadeau *et al.* 2012; Wagner *et al.* 2012; Eaton & Ree 2013; Jones *et al.* 2013; Cruaud *et al.* 2014; Escudero *et al.* 2014; Hipp *et al.* 2014; Leache *et al.* 2014; Herrera *et al.* 2015), including cnidarians (Reitzel *et al.* 2013) and most recently deep-sea octocorals (Pante *et al.* 2014). The number of orthologous restriction sites that can be retained across taxa, which decreases as divergence increases, limits the usefulness of RAD-seq for these kinds of studies. *In silico* studies in model organisms indicate that RAD-seq can be used to infer phylogenetic relationships in young groups of species (up to 60 million years old), such as *Drosophila* (Rubin *et al.* 2012; Cariou *et al.* 2013; Seetharam & Stuart 2013)*;* however, the real limits of this technique have not been significantly explored.

In this study we aim to empirically explore the limits of RAD-seq to solve questions in phylogenetics and species delimitation. We focus on the recalcitrant *Anthomastus-Corallium* clade of octocorals (sensu McFadden *et al.* 2006) to test the utility of RAD-seq to resolve phylogenetic relationships among divergent taxa, and to infer objective species boundaries. Corals in the *Anthomastus-Corallium* clade (hereafter referred as the AC clade) are among the most conspicuous, widely distributed, and ecologically important benthic invertebrates in deep-water ecosystems (Roberts *et al.* 2009; Wating *et al.* 2011). This clade is constituted by more than 100 species defined morphologically, divided in 10 genera, and three families (World Register of Marine Species at http://www.marinespecies.org accessed on 2014-10-10), spanning a divergence time of over 100 million years (Ardila *et al.* 2012; Herrera *et al.* 2012). However, species delimitations and phylogenetic relationships in this clade, as in other octocorals, are controversial and conflictive (Herrera *et al.* 2010; Ardila *et al.* 2012; Herrera *et al.* 2012). Many of the species in this group are considered species indicators of Vulnerable Marine Ecosystems (e.g. ICES 2013), with some of them considered endangered (CITES 2014). Accurate species identifications, as well as complete inventories and knowledge of species ranges, are therefore critical to ensure the effectiveness and appropriateness of conservation and management policies.

RESULTS

Morphological species identifications

Using current species descriptions, colony observations, and scanning electron microscopy of sclerites, we identified a total of 12 putative morphological species among the 44 examined specimens from the AC clade (Table S1, Supplementary Material images). These species correspond to the genera *Paragorgia* (*P. arborea, P. stephencairnsi, P. johnsoni, P. maunga, P. alisonae, P. kaupeka,* and *P. coralloides*) and *Sibogagorgia* (*S. cauliflora*) of the family Paragorgiidae; *Hemicorallium* (*H. laauense-imperiale*) and *Corallium* of the family Coralliidae; and *Anthomastus* and *Heteropolypus* of the family Alcyoniidae.

Octocorals are amenable to RAD sequencing

We generated a dense genome-wide set of genetic markers from the 44 AC clade specimens via RAD sequencing, using the 6-cutter restriction enzyme PstI, and used them to perform phylogenetic inferences and species delimitation analyses. We obtained roughly 3.9 ± 1.4 million reads (average ± standard deviation) per individual, of which 74.3 ± 8.1% were retained after stringent quality filtering steps (Table S2).

Optimization of RAD-loci clustering parameters

To examine the sensitivity of the phylogenetic inference to the clustering parameters used to identify loci and create nucleotide matrices in the program pyRAD (Eaton 2014), we investigated different combinations of clustering thresholds (c 0.80, 0.85 and 0.90) and minimum number of taxa per locus (m 4, 6, and 9) in a reduced **'backbone'** matrix (hereafter matrix names will be highlighted in bold) containing one individual from each of the 12 morphological species. The 9 resulting **backbone** matrices ranged in the total number of loci per matrix from approximately 9 to 60 thousand loci, increasing dramatically as the minimum number of taxa per locus was reduced (Table S3). In contrast, the different clustering thresholds did not have a significant effect on the total number of loci, but rather on the number of variable sites and, most importantly, on the number of phylogenetically informative sites (Table S3). Each resulting **backbone** matrix analyzed in RAxML (Stamatakis 2006) produced identical strongly-supported tree topologies (Fig S1). We selected c 0.80 (80% similarity among sequences) and m 9 (minimum coverage of taxa per locus of 75%) as the optimal combination of loci-clustering parameters because they minimized the proportion of missing data (0.20) in the matrix while maximizing the fraction of variable sites that were phylogenetically informative (0.24) (Table S3). The proportion of shared loci among individuals of Paragorgiidae and Coralliidae, lineages whose split has been estimated to be between 80-150 million years ago (Ardila *et al.* 2012; Herrera *et al.* 2012), was remarkably high (70-80%) (Fig 1).

We used the selected optimal loci-clustering parameters to generate the **'PHYLO'** matrix, containing the sequence data of all the 44 octocoral specimens. The use of the parameter value c 0.80 yielded approximately 71 ± 15 thousand loci – with a minimum depth of coverage of 5x and after filtering for paralogs– per specimen (mean depth of clusters used in loci construction was 23 ± 8x) (Table S4). The **PHYLO** matrix contained a total of 5,997 loci that contained data for at least 75% of the specimens (after a second paralog removal). There were 85,293 variable sites in this matrix, of which 53,150 were phylogenetically informative.

RAD-seq data support a fully resolved phylogeny

The phylogenetic analysis of the **PHYLO** concatenated RAD-seq matrix produced a completely resolved evolutionary tree of the AC specimens (Fig 2). In general, all branches were supported by high (greater than 95) bootstrap values, except for the one supporting the clade of *P. johnsoni, P. alisonae,* and *P. maunga*. Each one of the morphologically identified families, genera, and species in this dataset were monophyletic. The branching pattern of the tree is consistent with an expected transition between coalescent processes among species and genera (long deep branches), and population processes within species (short shallow branches).

The topology of the tree obtained with a traditional '**mitochondrial'** matrix (711 base pairs of the *mtMutS* gene containing 130 variable sites, of which 101 were phylogenetically informative) was incongruent with the **PHYLO** tree (Fig 2). The **mitochondrial** tree indicated a well-supported alternative divergence order for *P. coralloides* and *P. kaupeka* in the *Paragorgia* clade. In addition, the families Paragorgiidae (bubblegum corals) and Coralliidae (precious corals) were not monophyletic. The bubblegum coral genus *Sibogagorgia* appeared more closely related to the precious corals than to the other bubblegum coral genus *Paragorgia,* and the genera *Corallium* and *Hemicorallium* did not form a clade. However, these alternative relationships were not significantly supported by the bootstrap analysis,. Indeed, a substantial proportion of branches on the **mitochondrial** tree were poorly supported (bootstrap values smaller than 80%).

RAD-seq data reveal cryptic genetic diversity

Branch-length differences among individuals, as well as well-supported sub-clades, revealed intraspecific genetic diversity that was undetected by the **mitochondrial** matrix. Two sub-clades were revealed by the phylogenetic analysis of the **PHYLO** matrix in the *P. arborea* and *P. stephencairnsi* clades. The sub-clades in *P. arborea* correspond to a pattern of segregation by geographic location with specimens from the north Pacific in one sub-clade, and specimens from the south Pacific and north Atlantic in the other. Contrastingly, the sub-clades in *P. stephencairnsi* correspond to a pattern of segregation by depth with specimens collected shallower than 350m in one sub-clade, and specimens collected deeper than 1000m in the other.

Current morphological species delimitation is rejected

To evaluate the utility of RAD-seq to perform objective species delimitations in octocorals we focused on specimens the genus *Paragorgia* as it was the best-sampled taxon in our dataset, both in terms of geographic representation and number of morphological species. We used the Bayes Factor Delimitation method with genomic data (BFD*) (Leache *et al.* 2014), which allows for the comparison of conflictive species delimitation models in an explicit multispecies coalescent framework using genome-wide single nucleotide polymorphism (SNP) data. We calculated marginal likelihoods of taxonomy-guided and taxonomy-unguided species delimitation models from a matrix of unlinked SNPs including only specimens of *Paragorgia* (**'PARAGORGIA'** matrix containing 1,203 SNPs present in all individuals). We compared the marginal likelihood estimates of alternative species delimitation models to the null model '**morphid'**, which is based on current morphological species descriptions, using Bayesian factors.

The null model, **morphid,** was rejected in favor of alternative species delimitation models for *Paragorgia* (Fig 3) (**morphid** was ranked 7th among 10 evaluated models in terms of the marginal likelihood estimate). The **'PABSTE'** model, which proposes 9 species based on the 7 morphological species in the dataset plus splits corresponding to the sub-clades in *P. arborea* and in *P. stephencairnsi,* received decisive support from Bayes factors as the best species delimitation model. The taxonomy-unguided model '**geo**' – which splits the specimens based on the geographic location where they were collected – and the models proposed by the Poisson tree processes (PTP) method based on the **mitochondrial** data matrix, were the lowest ranked and most strongly rejected models overall.

Concatenated and coalescent species tree analyses are congruent

The topology of the species tree inferred using the SNP **PARAGORGIA** matrix was entirely congruent with the topology generated by the maximum likelihood phylogenetic analysis of the concatenated sequence matrices (Fig 4). The species tree analysis also greatly improved support for the clade of *P. johnsoni, P. alisonae,* and *P. maunga.* The posterior distribution of species trees indicated a small fraction of conflictive topologies concentrating in this region of the tree.

DISCUSSION

RAD sequencing enables unprecedented phylogenetic resolution

Our analyses of RAD-seq data provide a robust phylogenetic hypothesis for the recalcitrant octocorals in the *Anthomastus-Corallium* clade, a result never achieved before. Moreover, this study, together with the work by Pante *et al.* (2014) in the octocoral genus *Chrysogorgia*, constitute the first applications of RAD-sequencing for phylogenetics and species delimitation in cnidarians. Only a handful of previous studies, using traditional mitochondrial data and the ITS2 and 28S nuclear markers, have attempted to evaluate phylogenetic relationships in the octocoral AC clade (Herrera *et al.* 2010; Ardila *et al.* 2012; Brockman & McFadden 2012; Herrera *et al.* 2012; McFadden & van Ofwegen 2013; Uda *et al.* 2013; Figueroa & Baco 2014). These studies find support for the monophyly of the genus *Paragorgia,* the family Coralliidae, and the sister relationship between the Paragorgiidae and Coralliidae. However, those data do not provide enough phylogenetic resolution to infer the evolutionary relationships among many of the putative morphological species. Furthermore, significant incongruences between mitochondrial and nuclear ITS2 gene trees from AC taxa have been documented (Herrera *et al.* 2010). Here we reproduce similar incongruences when comparing the trees inferred from mitochondrial and RAD-seq datasets (Fig 2). Likewise, Pante *et al.* (2014) documented marked incongruence between trees inferred from mitochondrial and RAD-seq data in *Chyrsogorgia.* These observations suggest that processes that can cause gene tree heterogeneity, such as incomplete lineage sorting and horizontal gene transfer (Maddison 1997; Edwards 2009), may be more prevalent in octocorals than previously recognized.

All of our analyses based on RAD-seq matrices – varying in taxon coverage, degree of divergence among taxa, proportion of missing data, number of loci, and analysis type (concatenated or species tree) – produced completely congruent trees, which together provide extremely high confidence on the phylogenetic hypothesis inferred for the octocoral AC clade (Figs 1, 2 and 3). Consequently, we suggest that single marker gene trees in octocorals, particularly from the mitochondria, should not be considered as robust hypotheses of true species phylogenies on their own, without further validation by multiple informative and independent nuclear loci. We urge systematists to be conservative when making taxonomic rearrangements based on inferences from single-marker data alone.

RAD-seq data is suitable for phylogenetic inference in divergent taxa

Contrary to the currently accepted idea that RAD-seq data are only suitable for taxa with divergence times younger than 60 million years (MY) (Rubin *et al.* 2012), we demonstrate their suitability well beyond this age threshold. Remarkably, we were able to confidently resolve phylogenetic relationships among genera from different families diverging by at least 80 MY in the AC clade. The split between the families Paragorgiidae and Coralliidae has been dated, using coralliid fossils, to be between 80-150 MY old (Ardila *et al.* 2012; Herrera *et al.* 2012). Park *et al.* (2012) estimated the age of the most recent common ancestor of the Coralliidae at approximately 50 MY (25-100 MY 95% confidence region), using independent cnidarian fossils for molecular clock calibration. The split with the genera *Anthomastus* and *Heteropolypus* is likely older than 100 MY. It is without question that, due to evolution at restriction sites, the number of RAD loci among taxa for which orthology can be established decreases rapidly as divergence increases. However, we suggest that the suitability limits of RAD-seq for phylogenetics in divergent taxa cannot be assessed in terms absolute time, but depend on taxon-specific factors such as mutation rate, generation time and effective population size.

Bioinformatic studies addressing the issue of extent of the suitability of RAD-seq for phylogenetic inference have focused mainly on *Drosophila* as study model (Rubin *et al.* 2012; Cariou *et al.* 2013). Longer generation times and lower metabolic rates in taxa like deep-sea corals, relative to those in organisms like *Drosophila,* could cause a reduction in mutation rates (see review by Baer *et al.* (2007)), which may in turn decrease the evolutionary rates at restriction sites and allow for phylogenetic inferences using RAD-seq in situations of deeper divergence. Consistent with this hypothesis, we observe a nucleotide diversity (π) calculated across all octocoral specimens from the **PHYLO** matrix of 0.012 ± 0.002 (considered a minimum since RAD-seq can underestimate diversity (Arnold *et al.* 2013); see Table S5 and Table S6 for individual values), which is significantly lower than the nucleotide diversity in many of the *Drosophila* species included in the bioinformatic studies by Cariou *et al.* (2013) and Rubin *et al.* (2012). Nonetheless, there are other important factors known to influence genetic diversity across species – and likely the evolutionary rate as well. These factors include the effective population size, selection, habitat kind, geographic range, and mating system (Leffler *et al.* 2012). To sum up, we argue that RAD-seq can be successfully used to infer phylogenetic relationships in certain taxa with deeper divergences than previously suggested. This is particularly true when the number of RAD loci is maximized through the choice of restriction enzymes with higher cutting frequencies in the target taxon (Herrera *et al.* 2014).

RAD-seq allows the formulation of robust species delineations

Our study, the first statistical rigorous test of species hypothesis in octocorals, provides conclusive evidence rejecting the current morphological species delimitation model for the genus *Paragorgia*. We find decisive support for a nested model that combines species boundaries from morphological taxonomy with cryptic diversity linked to environmental variables of geographic location and depth (Figs 3 and 4). This nested model, proposes 9 species among the examined specimens. Five of these species correspond to the morphological species *P. coralloides, P. kaupeka, P. alisonae, P. johnsoni,* and *P. maunga*. Two splits, corresponding to sub-clades in the morphological species *P. arborea* and in *P. stephencairnsi*, indicate cases of cryptic species.

Herrera *et al.* (2012) found significant genetic differentiation of the north Pacific populations of *P. arborea* relative to the south Pacific, Atlantic and Indian ocean populations, and suggested that these populations may represent sub-species. The north Pacific populations of *P. arborea* were previously defined as a separate species, *P. pacifica,* by Verrill (1922) based on gross colony morphology, but later combined into a single species by Grasshoff (1979). Sánchez (2005) suggested potential small differences in medullar sclerite sizes and ornamentation between north Pacific specimens and specimens form elsewhere. However, we were unable to recognize these morphological differences in the few examined specimens in this study. Nonetheless, based on the decisive support for the split of *P. arborea* from analysis of genome-wide SNP makers indicates, we resurrect the species *Paragorgia pacifica* for the north Pacific populations of formerly *P. arborea*. We find no evidence of cryptic speciation between the north Atlantic and south Pacific *P. arborea* and therefore conclude it should be considered a single species as previously suggested by Herrera *et al.* (2012).

Depth is an important factor contributing to genetic differentiation and formation of species in the ocean, both shallow (Carlon & Budd 2002; Prada & Hellberg 2013) and deep (Miller *et al.* 2011; Jennings *et al.* 2013; Quattrini *et al.* 2013; Glazier & Etter 2014). The observed cryptic differentiation between specimens of *P. stephencairnsi* collected shallower than 350m and deeper than 1000m indicates that depth is also a diversifying force in octocorals from the AC clade, which had gone undetected due to the low variability of traditional sequence data (Herrera *et al.* 2012). The holotype of *P. stephencairnsi* was collected from approximately 350m in the Georgia Strait of British Columbia, overlapping in depth range and geographic region with that of most of the specimens from the shallow sub-clade examined in this study. Therefore, we propose to conserve that name *P. stephencairnsi* for that shallow sub-clade, and consider the deep sub-clade as a new species.

Other recent species delimitation studies in anthozoan corals have also revealed significant incongruences when comparing morphological and single-locus species delimitation hypotheses (particularly from mitochondrial data) with phylogenetic evidence from multi-locus datasets (Pante *et al.* 2014; Prada *et al.* 2014). In line with the findings of Pante *et al.* (2014), we find that specimens of *Paragorgia* sharing identical *mtMutS* haplotypes can belong to more than one species. Contrastingly, Herrera *et al.* (2012) present strong evidence showing significant mitochondrial haplotype diversity in the south Pacific and north Atlantic populations of *Paragorgia arborea*. Our observations, together with those from the aforementioned studies, constitute compelling evidence indicating that there is no solid basis for the widespread assumption that *mtMutS* haplotypes may be equivalent to individual octocoral species, as proposed by Thoma *et al.* (2009). The analysis with RAD-seq, or alternative genomic multi-locus methods, of a larger number of specimens from diverse geographic locations and depth horizons will likely reveal further cryptic diversity not characterized by mitochondrial haploytypes (see Fig S2, Fig S3, and Table S8), and thus further illuminates taxonomy and systematics in this an other groups.

CONCLUSIONS

In this case study we demonstrate the empirical utility of RAD-seq to resolve phylogenetic relationships among divergent and recalcitrant taxa and to objectively infer species boundaries by testing alternative delimitation hypotheses. We were able to make use of RAD-seq to overcome long-standing technical difficulties in octocoral genetics, and to resolve fundamental questions in species definitions and systematics. We show that classic morphological taxonomy can greatly benefit from integrative approaches that provide objective tests to species delimitation hypothesis. Our results pave the way for addressing further questions in biogeography, species ranges, community ecology, population dynamics and evolution of octocorals and other marine taxa. The results from this study also represent a valuable reference resource for the development of tools, such as SNP arrays, that can be used to perform accurate species identifications, and generate species inventories that will aid the design and implementation of conservation and management policies.

METHODS

To perform identifications using current morphological species descriptions we performed colony observations and scanning electron microscopy of sclerites on 44 octocoral specimens from the AC clade (Table S1).

To obtain a genome-wide set of markers that could be useful for phylogenetic inferences of deep-divergent taxa and species delimitation in the AC clade (greater than 100 million years) we performed RAD sequencing with a the 6-cutter restriction enzyme PstI, which is predicted to cut between 32,000 and 110,000 times in the genome of an octocoral (Table S7). This predicted range was obtained using the observed frequency of the PstI recognition sequence, and its probability calculated using a trinucleotide composition model, in the genomes of the cnidarians *Nematostella vectensis, Acropora digitifera, Hydra vulgaris,* and *Alatina moseri* (Herrera *et al.* 2014). Genome size range of 0.3-0.5 pg was used based on observations obtained through flow cytometry in gorgoniid octocorals by Luisa Dueñas at the Universidad de los Andes, Bogotá, Colombia (personal communication). Total genomic DNA was purified from specimens following protocols described in Herrera *et al.* (2015). Concentration-normalized genomic DNA was submitted to Floragenex Inc (Eugene, OR). for library preparation and RAD sequencing. Libraries were sequenced by 48-multiplex, using 10-base pair barcodes, on a single lane of an Illumina Hi-Seq 2000 sequencer.

To compare the inferences obtained from RAD-seq data with the inferences drawn from traditional genetic barcoding data, we performed targeted sequencing of the mitochondrial *mtMutS* gene — a genetic marker widely used for phylogenetics and species delimitation studies in octocorals. Polymerase chain reactions were carried out following the protocols by Herrera *et al.* (2015). Primer pairs used for amplifications were AnthoCorMSH (Herrera *et al.* 2010) and Mut-3458R (Sánchez *et al.* 2003). Negative controls were included in every experiment to test for contamination. Purified PCR products were submitted to Eurofins Genomics (Eurofins MWG Operon, Inc.) for sequencing.

RAD-seq data filtering

Sequence reads were de-multiplexed and quality filtered with the process_radtags program from the package Stacks v1.20 (Catchen *et al.* 2013). Barcodes and Illumina adapters were excluded from each read and length was truncated to 91bp (-t 91) Reads with ambiguous bases were discarded (-c). Reads with an average quality score below 10 (-s 10) within a sliding window of 15% of the read length (-w 0.15) were discarded (-r). The rescue barcodes and RAD-tags algorithm was enabled (-r). Additional filtering, and the clustering within and between individuals to identify loci was performed using the program pyRAD v2.01 (Eaton 2014). Reads with more than 33 bases with a quality score below 20 were discarded.

RAD-seq loci clustering and phylogenetic inference

We investigated different combinations of clustering thresholds (c 0.80, 0.85 and 0.90) and minimum number of taxa per locus (m 4, 6, and 9) in a reduced dataset that included one individual from each of the 12 putative morphological species. The minimum depth of coverage required to build a cluster and the maximum number of shared polymorphic sites in a locus were kept constant at 4 (d) and 3 (p) respectively. Loci sequences were concatenated into combined matrices. We refer to these 9 resulting matrices as the **'backbone'** matrices. Each of the resulting backbone matrices was analyzed in RAxML-HPC2 v8.0 (Stamatakis 2006) for maximum likelihood (ML) phylogenetic tree inference. For this, and all the other phylogenetic analyses in RAxML, we assumed a generalized time-reversible DNA substitution model with a gamma-distributed rate variation across sites (GTR GAMMA). Branch support was assessed by 500 bootstrap replicates.

We selected an optimal combination of loci clustering parameters as the set of parameters that minimized the number of missing data and maximized the number of phylogenetically informative sites while producing a highly supported phylogenetic tree. The optimal set of parameters chosen was a clustering threshold of 80% similarity among sequences (c 0.80) and a minimum coverage of taxa per locus of 75% (m 9). A concatenated matrix containing the sequence data of all the 44 octocoral specimens, denominated **'PHYLO',** was built using this parameter combination (c 0.80, m 33) in pyRAD and subsequently analyzed in RAxML.

Phylogenetic inference with traditional genetic barcoding data

To compare the tree topology obtained from the phylogenetic inferences of the **PHYLO** RAD-seq dataset with traditional genetic barcoding data we analyzed the '**mitochondrial'** dataset (containing the *mtMutS* sequences) using RAxML. These two datasets – **PHYLO** and **mitochondrial** – contain data from the same individuals. To place the specimens from this study in a broader phylogenetic context we also analyzed the **mitochondrial** dataset in RAxML with the addition of *mtMutS* data from 233 additional specimens belonging to the AC clade, as well as outgroups (see Table S8, Fig S2, and Fig S3).

Testing species delimitation models for Paragorgia

We constructed 5 taxonomy-guided species delimitation models for *Paragorgia*: **i**) '**morphid**' model: 7 species based on current morphological species descriptions (Sánchez 2005); **ii**) '**PAB**' model: 8 species based on the 7 morphological species plus a split of *P. arborea* based on previous evidence of genetic differentiation of north Pacific populations (Herrera *et al.* 2012); **iii**) '**STE**' model: 8 species based on the 7 morphological species plus a split of *P. stephencairnsi* based on depth differences (specimens collected <350m vs. >1000m), as depth is known to be an important structuring variable in marine taxa (Jennings *et al.* 2013; Prada & Hellberg 2013; Quattrini *et al.* 2013); **iv**) '**PABSTE**' model: 9 species based on the 7 morphological species plus the splits of the **PAB** and **STE** models; **v**) '**splitPAB**' model: 10 species based on the 7 morphological species plus the split of the **STE** model and an additional split in the **PAB** model where *P. arborea* is split in 3 species corresponding to the ocean basin where the specimens were collected (north Pacific, south Pacific and north Atlantic).

We also generated taxonomy-unguided species delimitation models for *Paragorgia* through Bayesian and ML implementations of the Poisson tree processes model (PTP) (available at http://species.h-its.org/ptp/). PTP estimates the number of speciation events in a rooted phylogenetic tree in terms of nucleotide substitutions (Zhang *et al.* 2013). We used PTP to analyze the trees obtained from phylogenetic inferences in RAxML of reduced *mtMutS* and RAD-seq datasets that include only members of the family Paragorgiidae (genera *Paragorgia* and *Sibogagorgia*). The '**PARAGORGIIDAE**' RAD-seq concatenated matrix was generated in pyRAD using a clustering threshold of 80% similarity among sequences (c 0.80) and a minimum coverage of taxa per locus of 100% (m 33). The resulting phylogenetic trees of *Paragorgia* were rooted with the specimens of *Sibogagorgia* and analyzed by the PTP method using a Markov Chain Monte Carlo (MCMC) chain length of 500,000 generations (100 thinning, 25% burnin). We assessed convergence by examining the likelihood trace. The combinations of the ML or Bayesian PTP implementations (mlPTP and bPTP, respectively) with the *mtMutS* or RAD-seq trees of *Paragorgia* resulted in four species delimitation models: **i) 'mlPTP*mt*'** model; **ii) 'bPTP*mt*'** model; **iii) 'mlPTPrad'** model; and **iv) 'bPTPrad'** model. Lastly, because deep-sea corals are known to show genetic differentiation at ocean basin/regional scales (Miller *et al.* 2011; Morrison *et al.* 2011; Herrera *et al.* 2012), we constructed an additional taxonomy-unguided species delimitation model – the '**geo**' model – based on the geographic location where the specimens were collected (north Pacific, south Pacific or north Atlantic ocean basins).

To estimate the marginal likelihood of each species delimitation model we generated a matrix including only specimens of *Paragorgia*, denominated **'PARAGORGIA'** using a clustering threshold of 80% similarity among sequences (c 0.80) and a minimum coverage of taxa per locus of 100% (m 31) in pyRAD. In contrast to the **backbone**, **PHYLO**, and **PARAGORGIIDAE** RAD-seq matrices, this matrix contained the data of one SNP per locus and not the entire locus sequence. We analyzed these data using the implementation of BFD* in the SNAPP (Bryant *et al.* 2012) plug-in for the program BEAST v2.1.3 (Bouckaert *et al.* 2014). We performed a path-sampling of 48 steps, with a MCMC chain length of 100,000 (10,000 pre-burnin), following the guidelines from Leache *et al.* (2014). Bayesian factors were calculated from the marginal likelihood estimates for each model and compared using the framework proposed by Kass and Raftery (1995)

Species tree inference

To test the tree topology in the genus *Paragorgia* obtained by the phylogenetic analysis of the **PHYLO** and **PARAGORGIIDAE** concatenated matrices we performed a species tree inference from the SNP data in the **PARAGORGIA** matrix using the program SNAPP. This program allows the inference of species trees from unlinked SNP data (only one SNP per locus retained) bypassing the inference of individual gene trees (Bryant *et al.* 2012). We performed 3 independent runs using a MCMC chain length of 10,000,000 (sampling every 1,000 generations; pre-burnin of 1,000) with default prior distributions for coalescence rate, mutation rate and ancestral population size parameters. We assessed convergence to stationary distributions and effective sample sizes >200 after 10% burnin in the program TRACER

(Rambaut & Drummond 2007). Species trees in the posterior distribution were summarized with the program DENSITREE v2.01 (Bouckaert 2010).

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FIGURES & TABLES

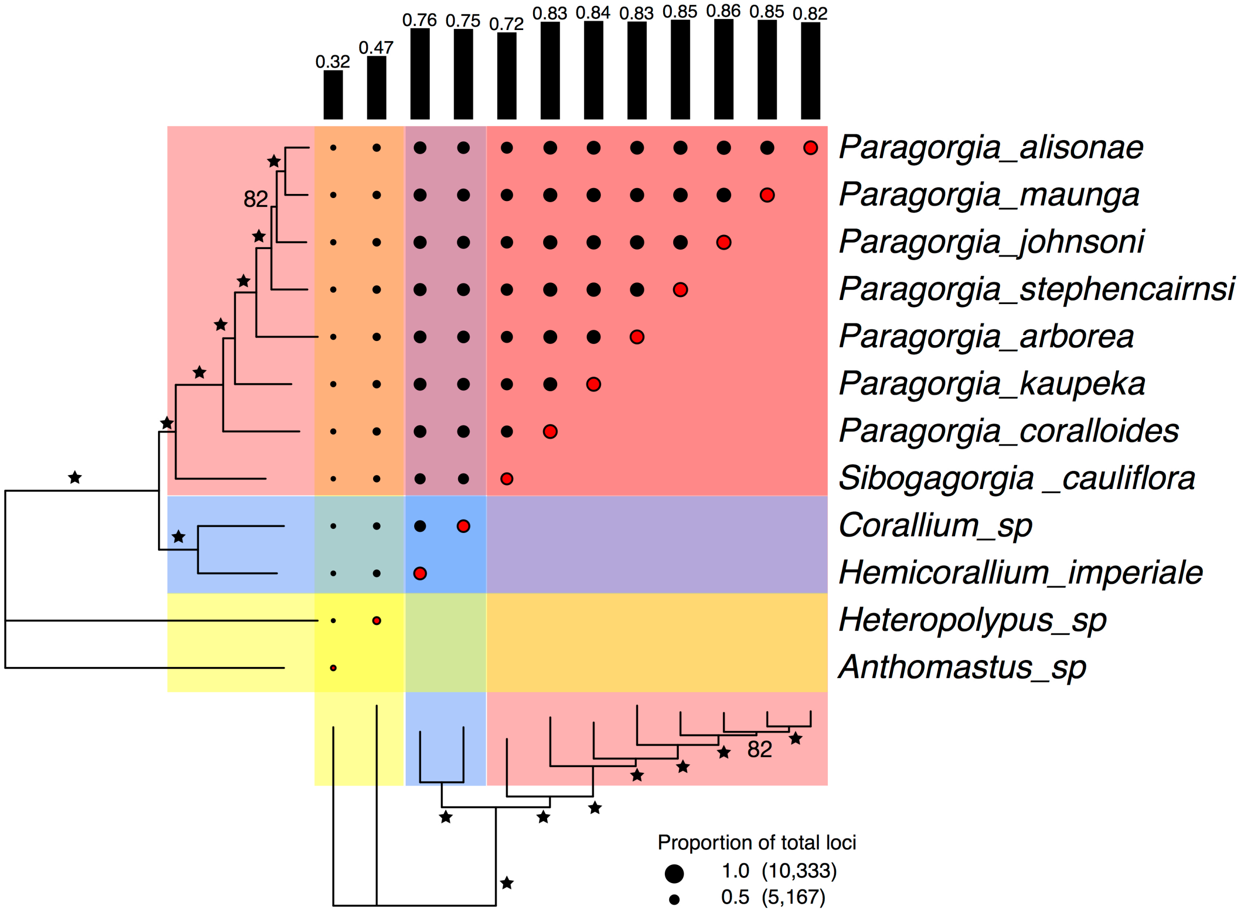


Figure 1. Proportion of loci shared among individuals of the AC clade in the optimal backbone matrix (c 0.80, m 9). Each family is indicated with a different color: red for Paragorgiidae; blue for Coralliidae; and yellow for Alcyoniidae. Black-filled circles represent the proportion of the total number of loci shared among individuals. Red-filled circles represent the proportion of the total number of loci present in each individual. Circle scale shows the number of loci represented by 1.0 and 0.5 circle sizes. Black vertical bars represent the average proportion of loci shared by each individual. Phylogenetic tree was inferred with RAxML. Stars on the tree represent branch bootstrap support of 100. Smaller bootstrap support values are indicated with numbers. This figure was generated with the package RADami (Hipp *et al.* 2014).

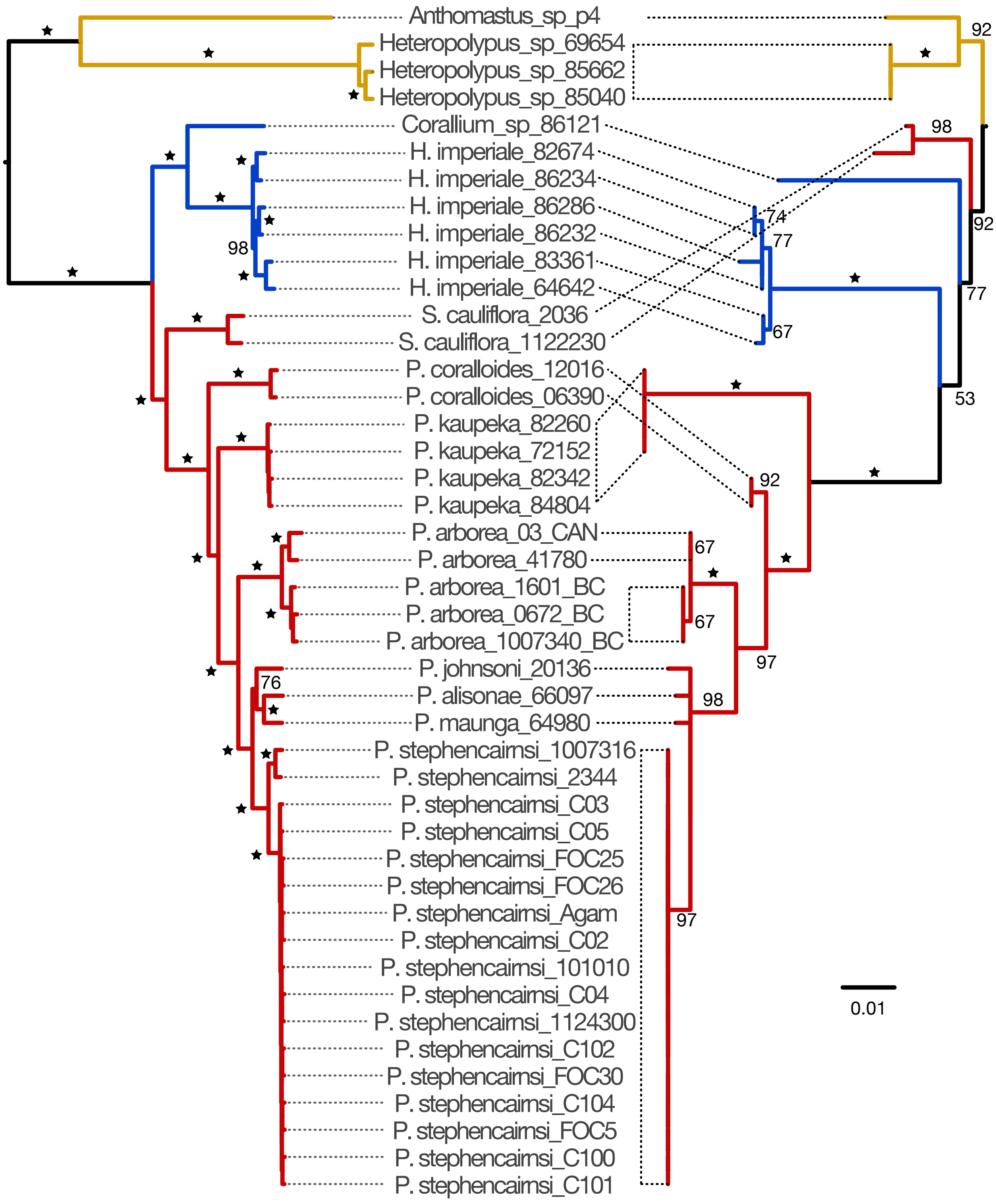
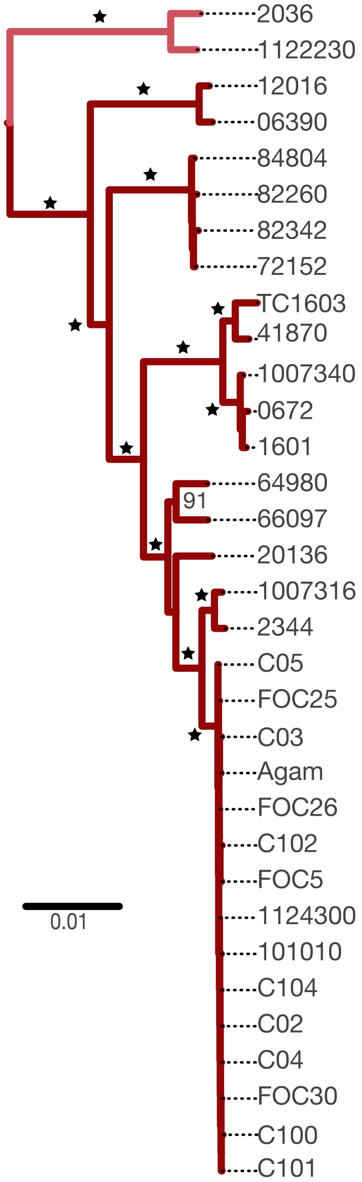


Figure 2. Phylogenetic trees of the AC clade based on RAD-seq and mitochondrial data. Left tree based on the RAD-seq concatenated PHYLO matrix. Right tree based on the mtMutS mitochondrial matrix. Each family is indicated with a different branch color: blue red for Paragorgiidaea; blue for Coralliidae; and yellow for Alcyoniidae. Phylogenetic trees were inferred with RAxML. Stars on the trees represent bootstrap support of 100. Smaller bootstrap values are indicated in numbers. Scale bar indicates substitutions per site.



	Species models									
	Taxonomy-informed					Taxonomy-independent				
	morphid	PAB	STE	PAB STE	split PAB	mlPTP mt	bPTP mt	mlPTP rad	bPTP rad	geo
2036	*S. cauliflora*	-	-	-	-	-	-	-	-	-
1122230	*S.cauliflora*	-	-	-	-	-	-	-	-	-
12016	*P. coralloides*	1	1	1	1	8	1	11	8	1
06390	*P. coralloides*	1	1	1	1	7	1	12	9	1
84804	*P. kaupeka*	2	2	2	2	1	17	1	1	2
82260	*P. kaupeka*	2	2	2	2	1	16	1	1	2
82342	*P. kaupeka*	2	2	2	2	1	19	1	1	2
72152	*P. kaupeka*	2	2	2	2	1	20	1	1	2
TC1603	*P. arborea*	3	3	3	3	2	10	7	6	1
41870	*P. arborea*	3	3	3	4	2	10	8	7	2
1007340	*P. arborea*	4	3	4	5	2	13	6	5	3
0672	*P. arborea*	4	3	4	5	2	14	6	5	3
1601	*P. arborea*	4	3	4	5	2	14	6	5	3
64980	*P. maunga*	5	4	5	6	4	3	3	3	2
66097	*P. alisonae*	6	5	6	7	5	4	4	4	2
20136	*P. johnsoni*	7	6	7	8	3	2	2	2	1
1007316	*P. stephencairnsi*	8	7	8	9	6	23	9	12	3
2344	*P. stephencairnsi*	8	7	8	9	6	9	10	13	3
C05	*P. stephencairnsi*	8	8	9	10	6	24	5	10	3
FOC25	*P. stephencairnsi*	8	8	9	10	6	11	5	11	3
C03	*P. stephencairnsi*	8	8	9	10	6	15	5	11	3
Agam	*P. stephencairnsi*	8	8	9	10	6	22	5	11	3
FOC26	*P. stephencairnsi*	8	8	9	10	6	5	5	11	3
C102	*P. stephencairnsi*	8	8	9	10	6	8	5	11	3
FOC5	*P. stephencairnsi*	8	8	9	10	6	24	5	11	3
1124300	*P. stephencairnsi*	8	8	9	10	6	7	5	11	3
101010	*P. stephencairnsi*	8	8	9	10	6	7	5	11	3
C104	*P. stephencairnsi*	8	8	9	10	6	12	5	11	3
C02	*P. stephencairnsi*	8	8	9	10	6	18	5	11	3
C04	*P. stephencairnsi*	8	8	9	10	6	23	5	11	3
FOC30	*P. stephencairnsi*	8	8	9	10	6	21	5	11	3
C100	*P. stephencairnsi*	8	8	9	10	6	6	5	11	3
C101	*P. stephencairnsi*	8	8	9	10	6	11	5	11	3
Number of species	7	8	8	9	10	8	24	12	13	3
Marginal Likelihood	-6240	-6221	-6039	-6014	-6028	-6251	-6243	-6050	-6045	-10296
Rank	7	6	3	1	2	9	8	5	4	10
Bayes Factor	-	-38	-401	-451	-424	23	7	-379	-388	8113

Figure 3. Species delineation hypotheses for *Paragorgia.* Table shows the different species delimitation models for *Paragorgia* evaluated with the BFD* method and their results. *Sibogagorgia* was included as outgroup to root the inferences for *Paragorgia*. Each row indicates a different specimen. Each column indicates a different species delimitation model. The first column, model morphid, indicates the species identifications based on morphology.

For all other models, numbers indicate the species assignments. Bottom rows show the total number of species proposed, the marginal likelihood estimate, and rank for each model. The Bayes factor comparisons were calculated with respect to the null morphid model. Phylogenetic tree on the left, shown only for visual reference, was inferred with the RAD-seq concatenated PARAGORGIID matrix in RAxML. Each genus is indicated with a different branch color: pink for *Sibogagorgia*; and dark red for *Paragorgia*. Stars on the trees represent bootstrap support of 100. Smaller bootstrap values are indicated in numbers. Scale bar indicates substitutions per site.

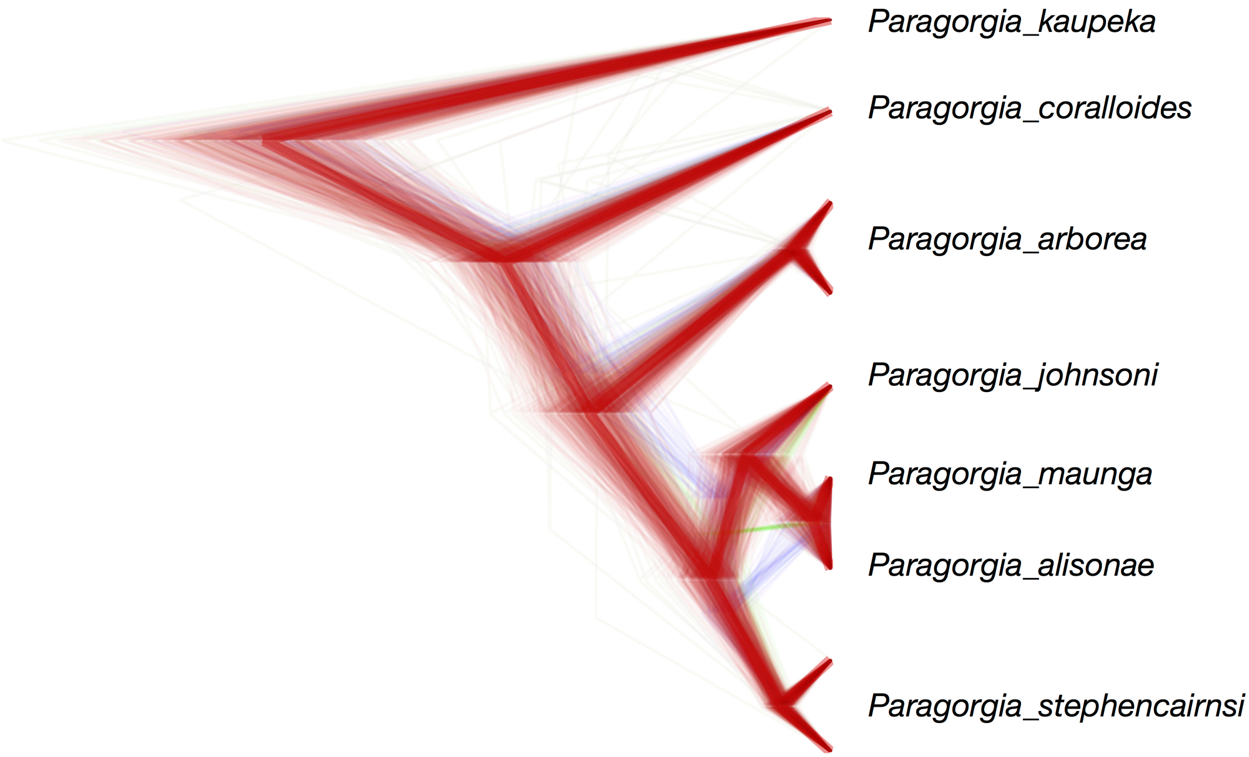


Figure 4. Species tree of *Paragorgia.* This claudogram illustrates the posterior distribution of species trees inferred with SNAPP based on the best species delimitation model PABSTE. High color density is indicative of areas in the species trees with high topology agreement. Different colors represent different topologies. The maximum clade credibility species tree is shown with thicker branches. Trees with the same topology as the maximum clade credibility species tree are colored in red. Trees with different topologies are colored green or blue. With the exception of the branch leading to the clade of *P. johnsoni, P. maunga*, and *P. alisonae,* which has a posterior probability of 0.87*,* all interior branches have posterior probabilities of 1.0.

SUPPLEMENTARY MATERIALS

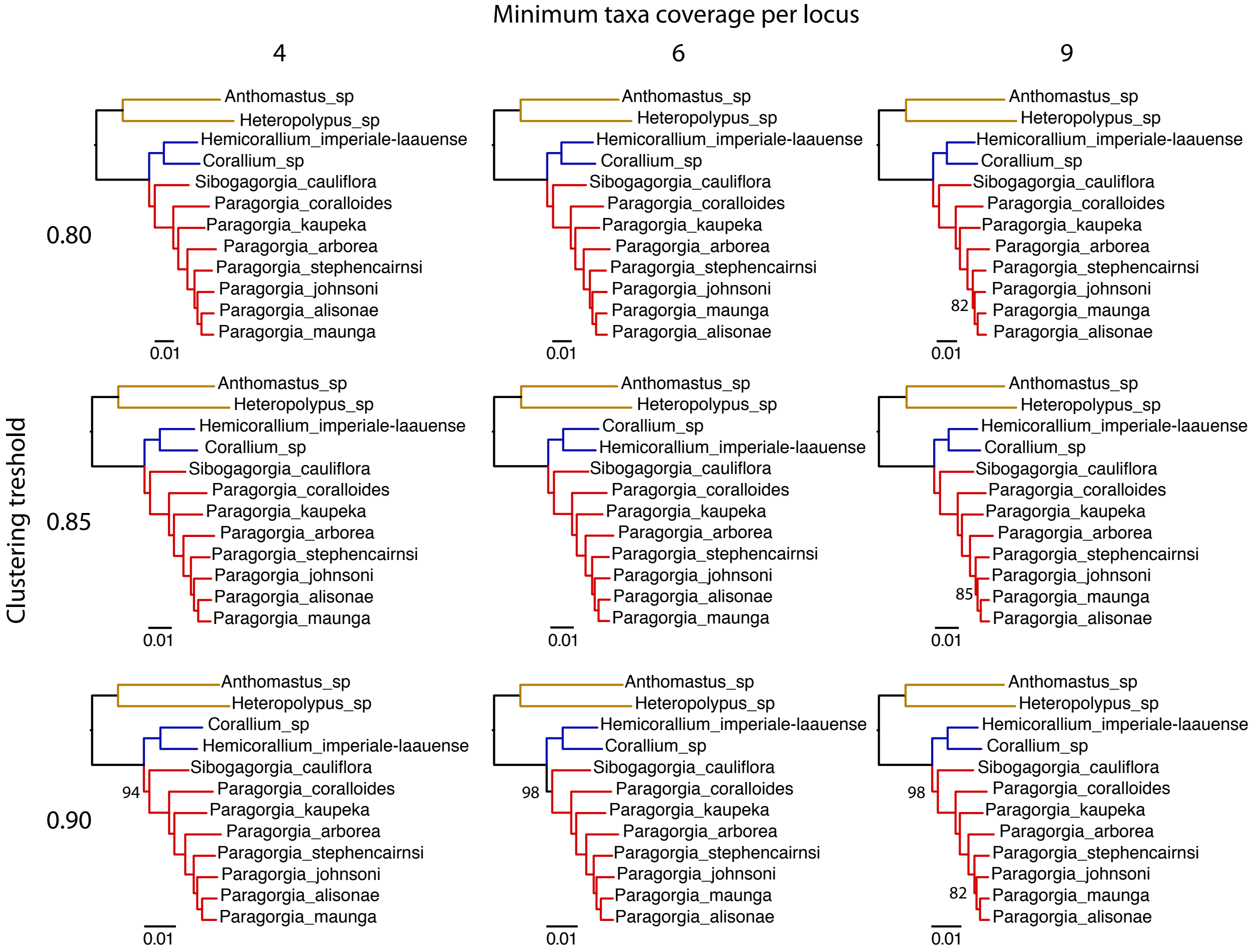
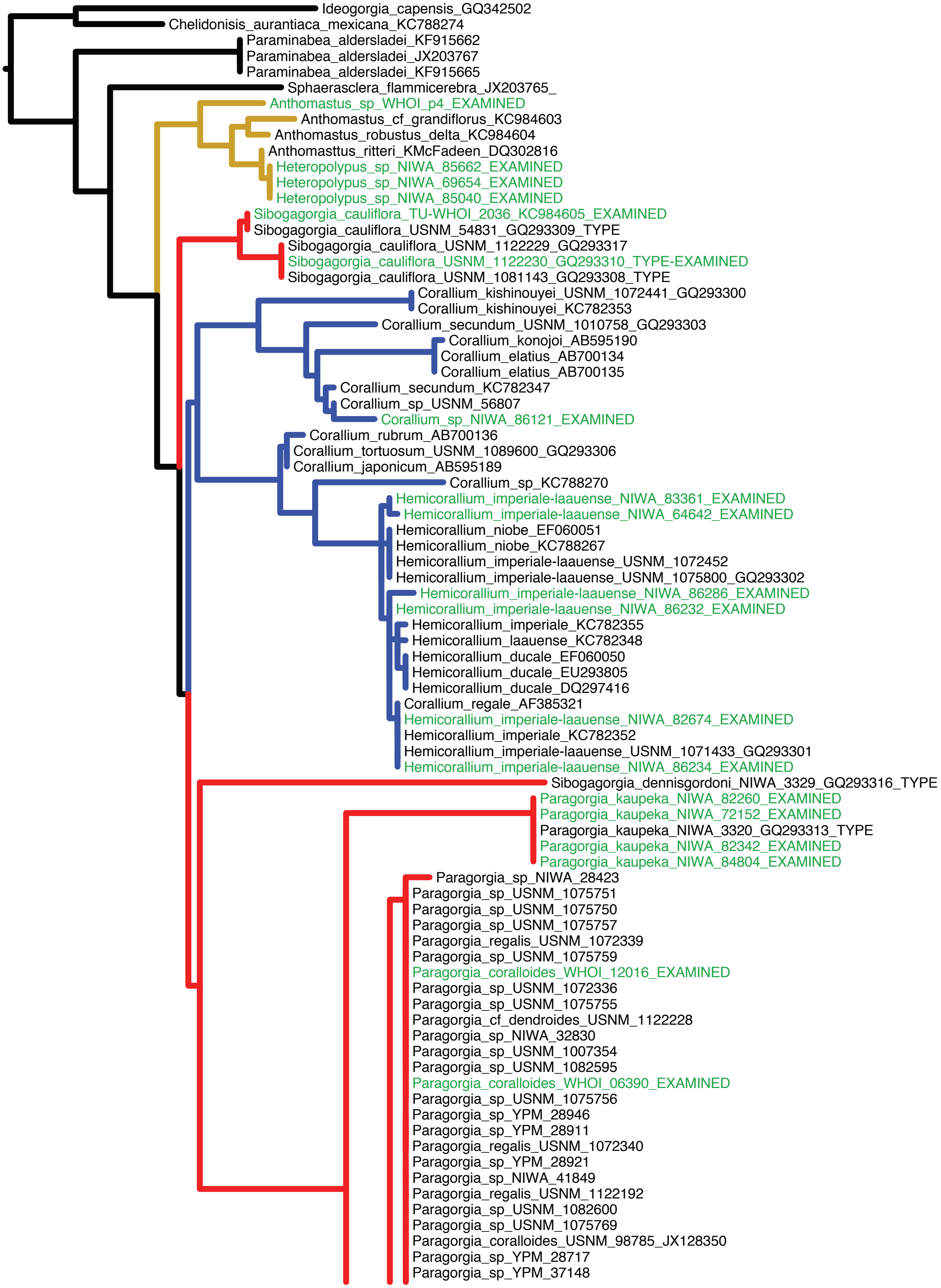


Figure S1. Phylogenetic trees based on backbone matrices. Trees inferred from the 9 backbone RAD-seq matrices built with different parameters of clustering threshold (c 0.80, 0.85 and 0.90; indicated by vertical labels) and minimum number of taxa per locus (m 4, 6, and 9; indicated by horizontal labels). Each family is indicated with a different branch color: red for Paragorgiidae; blue for Coralliidae; and yellow for Alcyoniidae. Trees were inferred with RAxML. All interior branches have bootstrap support values of 100, except for those shown. Scale bars indicate substitutions per site.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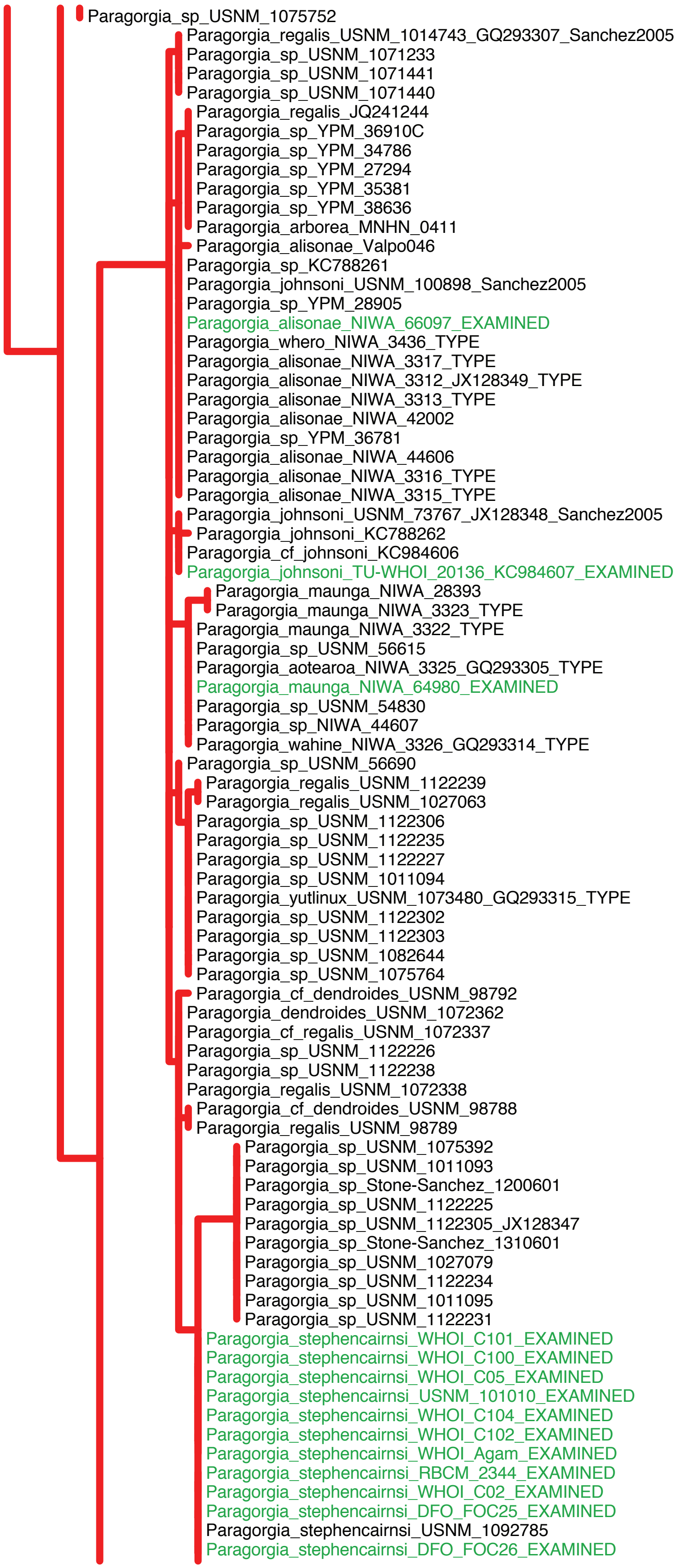
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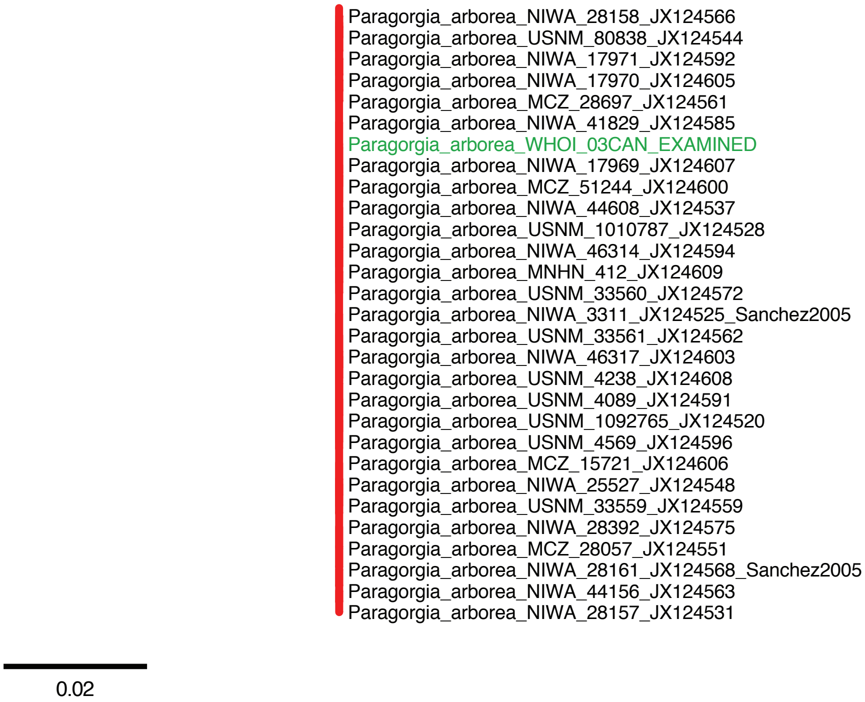
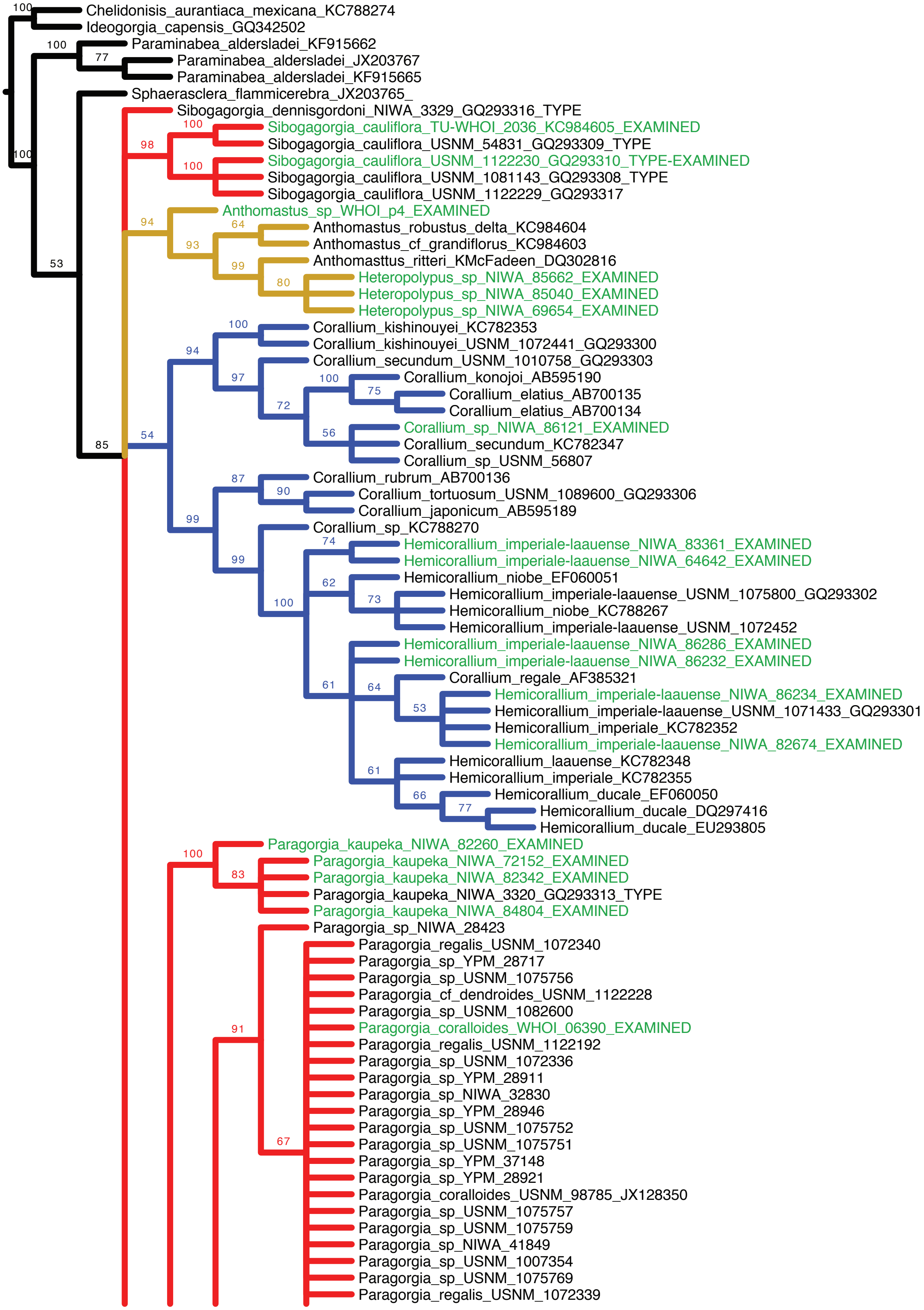


Figure S2. Mitochondrial *mtMutS* gene tree of all available sequences for the clade AC. Tree inferred from *mtMutS* sequence data from specimens examined in this study, GenBank, and additional specimens. Each family is indicated with a different branch color: red for Paragorgiidae; blue for Coralliidae; and yellow for Alcyoniidae. Outgroups are indicated with black branches. Specimens examined in detail in this study are indicated with green labels. Type specimens are labeled TYPE. Specimens examined in Sanchez (2005) are labeled “Sanchez2005”. Tree was inferred with RAxML. Scale bars indicate substitutions per site.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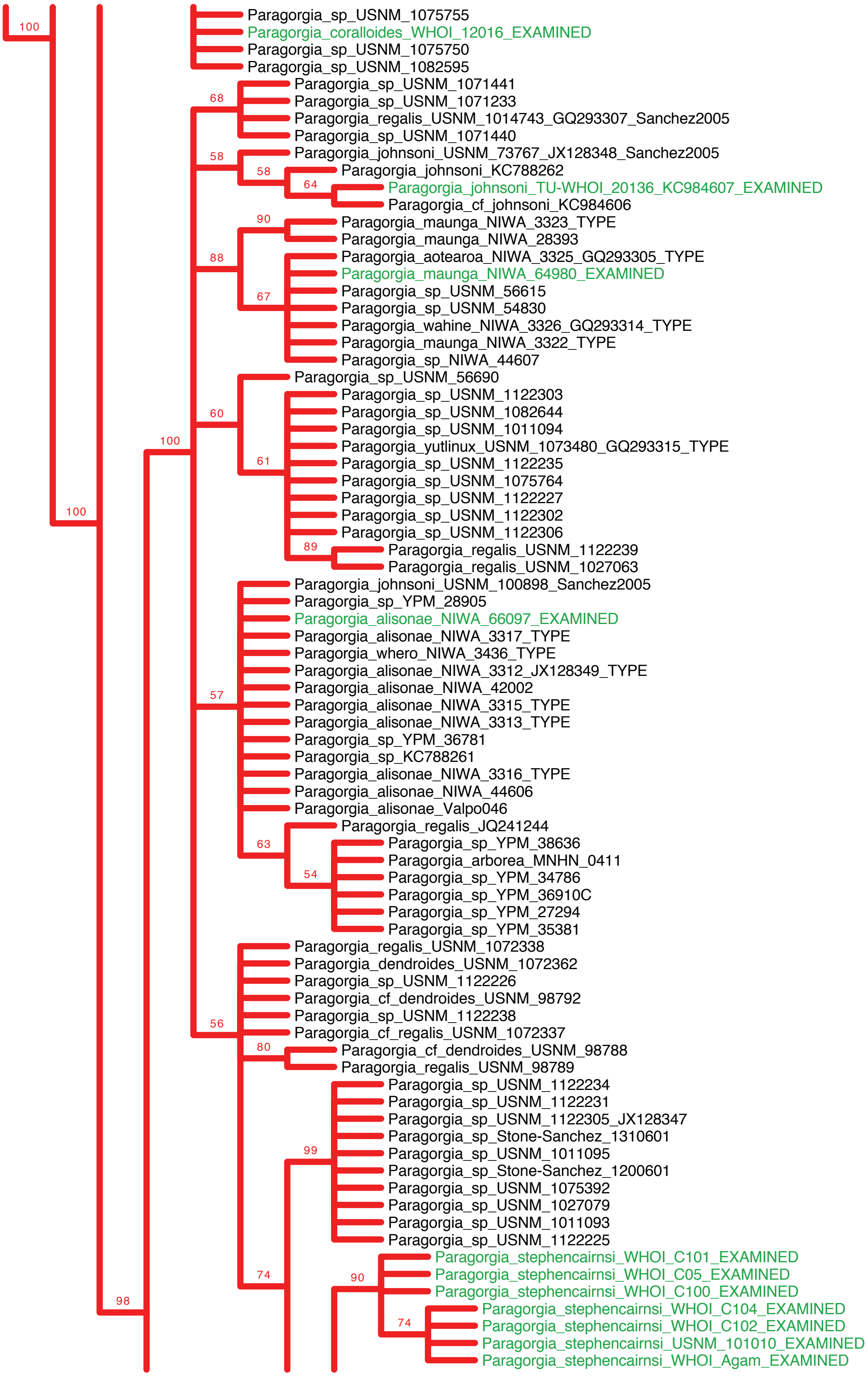
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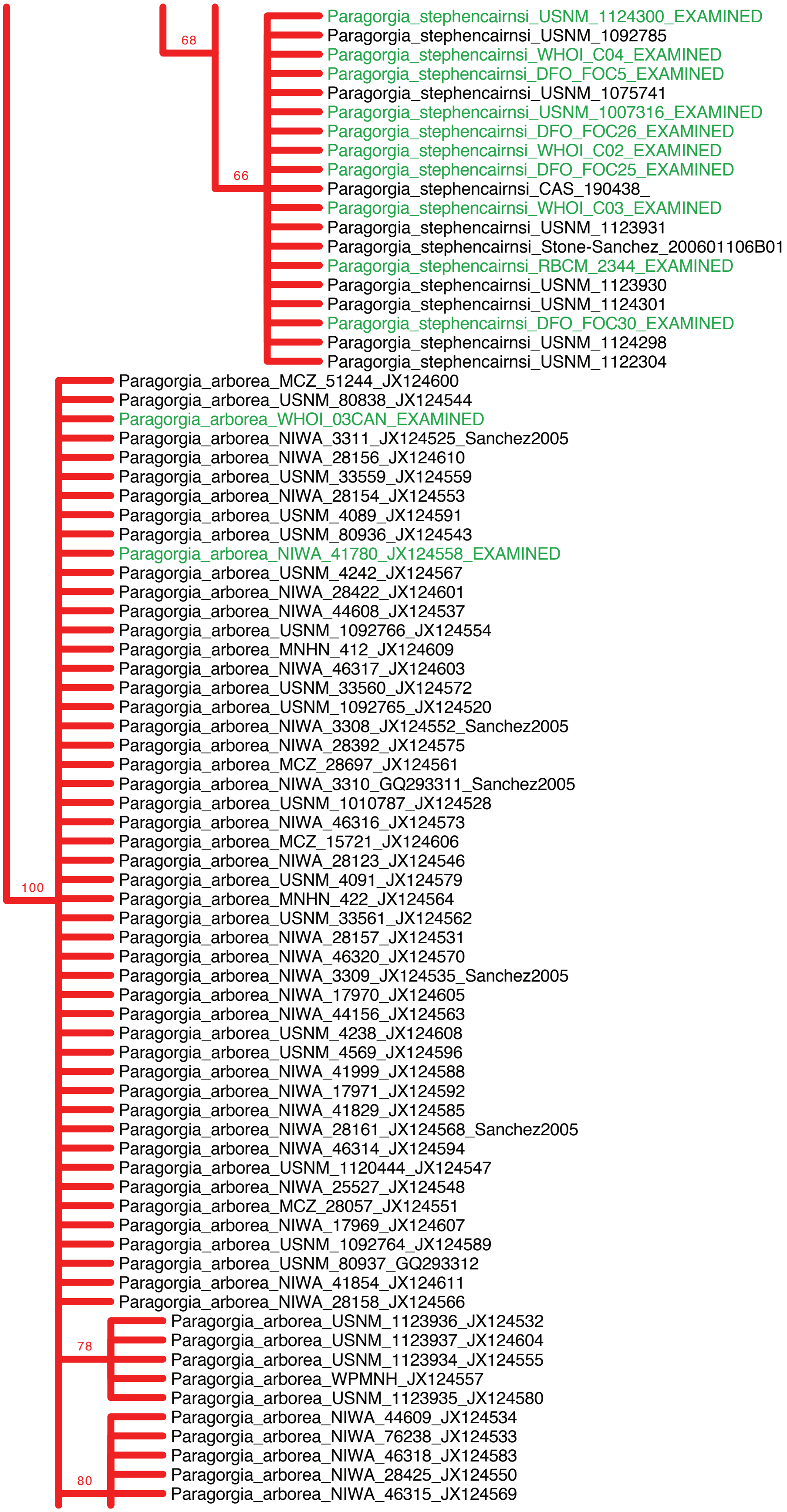
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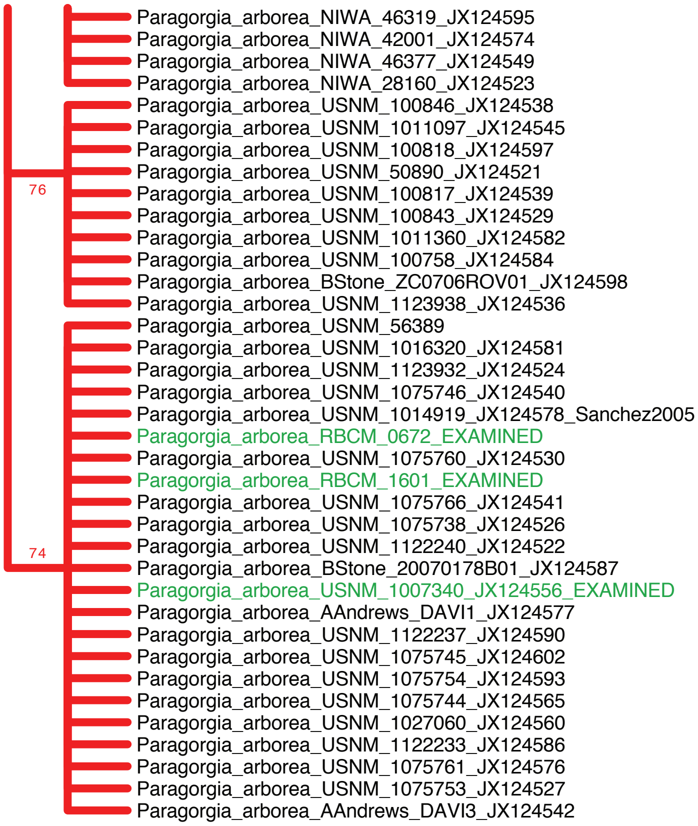

Figure S3. Mitochondrial *mtMutS* bootstrap support consensus tree of all available sequences for the clade AC. Tree inferred from *mtMutS* sequence data from specimens examined in this study, GenBank, and additional specimens. Each family is indicated with a different branch color: red for Paragorgiidae; blue for Coralliidae; and yellow for Alcyoniidae. Outgroups are indicated with black branches. Specimens examined in detail in this study are indicated with green labels. Type specimens are labeled "TYPE". Specimens examined in Sanchez (2005) are labeled "Sanchez2005". Tree was created with RAxML using a 50% majority consensus from 500 bootstrap replicates.

Table S1. Collection and sequence information for the specimens used in this study.

ID	Morphological Species	Delimitation model PABSTE	Collection	Date Collected	Locality	Depth (m)	Lat.	Lon.
p4	*Anthomastus sp*		WHOI	2011	Patagonian shelf			
86121	*Corallium sp*		NIWA	2012	Kermadec Ridge; Colville ridge volcano; TAN1213/18	380	-30.19	179.72
64642	*Hemicorallium imperiale-laauense*		NIWA	2010	Kermadec Ridge; Rumble II East seamount cone; TAN1007/97	1050	-35.42	178.65
83361	*Hemicorallium imperiale-laauense*		NIWA	2012	Bay of Plenty; Site SM1b, Matatara Knoll; TAN1206/168	948	-37.19	176.98
82674	*Hemicorallium imperiale-laauense*		NIWA	2012	Kermadec Ridge; Site SM2aa, summit of Whakatane Seamount; TAN1206/77	878	-36.81	177.47
86234	*Hemicorallium imperiale-laauense*		NIWA	2012	Kermadec Ridge; Northeast pimple volcano; TAN1213/22	483	-30.08	179.82
86286	*Hemicorallium imperiale-laauense*		NIWA	2012	Kermadec Ridge; Havre volcano; TAN1213/30	860	-31.13	-179.05
86232	*Hemicorallium imperiale-laauense*		NIWA	2012	Kermadec Ridge; Northeast pimple volcano; TAN1213/22	483	-30.08	179.82
69654	*Heteropolypus sp*		NIWA	2011	Chatam Rise; TRIP3306/78	495	-44.33	-177.22
85040	*Heteropolypus sp*		NIWA	2012	Chatam Rise; TAN1208/22	2098	-42.59	179.42
85662	*Heteropolypus sp*		NIWA	2012	Chatam Rise; TAN1208/61	1931	-42.59	179.59
66097	*Paragorgia alisonae*	*P. alisonae*	NIWA	2008	Cambell Plateau; TRIP2718/50	875	-50.02	175.00
41780	*Paragorgia arborea*	*P. arborea*	NIWA	2008	Chatam Rise; TRIP2617/120	600	-44.52	175.78
TC16_03	*Paragorgia arborea*	*P. arborea*	WHOI	2013	NE US Canyons; Munson Canyon; TowCam 16	540	40.54	-67.01
0672	*Paragorgia arborea*	*P. pacifica*	RBCM	2004	British Columbia; VE14280		53.33	-135.66
1007340	*Paragorgia arborea*	*P. pacifica*	USNM	2001	British Columbia; Vancouver Island	1168	48.44	-126.38
1601	*Paragorgia arborea*	*P. pacifica*	RBCM	2009	British Columbia; VE14444	695	53.31	-135.58
063902	*Paragorgia coralloides*	*P. coralloides*	WHOI	2005	New England Seamounts; Rehobot Seamount; H13	1821	37.46	-59.95
12016	*Paragorgia coralloides*	*P. coralloides*	WHOI	2003	New England Seamounts; Manning Seamount, station 4; AD3890	2000	38.23	-60.46
20136	*Paragorgia johnsoni*	*P. johnsoni*	TU/WHOI	2009	Gulf of Mexico; MC751; J2-464	438	28.19	-89.80
72152	*Paragorgia kaupeka*	*P. kaupeka*	NIWA	2011	Kermadec Ridge; Clark Seamount, chimney field, north cone; TAN1104/13	877	-36.45	177.84
82260	*Paragorgia kaupeka*	*P. kaupeka*	NIWA	2012	Kermadec Ridge; Site SM3a, summit of Clark Seamount; TAN1206/34	850	-36.45	177.84
82342	*Paragorgia kaupeka*	*P. kaupeka*	NIWA	2012	Kermadec Ridge; Site SM3a, Clark Seamount; TAN1206/40	1100	-36.45	177.84
84804	*Paragorgia kaupeka*	*P. kaupeka*	NIWA	2005	Kermadec Ridge; Clark Seamount; KOK0506/12	870	-36.45	177.84
64980	*Paragorgia maunga*	*P. maunga*	NIWA	2010	Kermadec Ridge; Silent II seamount; TAN1007/120	772	-35.17	178.89
1007316	*Paragorgia stephencairnsi*	*P. sp. nov*	USNM	2001	British Columbia; Vancouver Island	1168	48.44	-126.38
2344	*Paragorgia stephencairnsi*	*P. sp. nov*	RBCM	2004	British Columbia; VE13978	1194	53.37	-133.31

101010	*Paragorgia stephencairnsi*	*P. stephencairnsi*	USNM	2008	California; Piggy Bank, southern California; DW-026-02	283	33.92	-119.47
1124300	*Paragorgia stephencairnsi*	*P. stephencairnsi*	USNM	2006	British Columbia; Vancouver Island, Ohiat Island; OC 06/952	188	48.83	-125.13
Agam	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2012	British Columbia; Agamemnon Channel	32	49.72	-124.05
C02	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2013	British Columbia; Agamemnon Channel; Dive02	41	49.74	-124.03
C03	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2013	British Columbia; Agamemnon Channel; Dive02	41	49.74	-124.03
C04	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2013	British Columbia; Agamemnon Channel; Dive02	41	49.74	-124.03
C05	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2013	British Columbia; Agamemnon Channel; Dive02	41	49.74	-124.03
C100	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2013	British Columbia; Vancouver Island, Tahsis Inlet; Dive07	40	49.86	-126.67
C101	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2013	British Columbia; Vancouver Island, Tahsis Inlet; Dive07	40	49.86	-126.67
C102	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2013	British Columbia; Vancouver Island, Tahsis Inlet; Dive07	40	49.86	-126.67
C104	*Paragorgia stephencairnsi*	*P. stephencairnsi*	WHOI	2013	British Columbia; Vancouver Island, Tahsis Inlet; Dive07	40	49.86	-126.67
FOC25	*Paragorgia stephencairnsi*	*P. stephencairnsi*	DFO	2012	British Columbia; W of Graham Island; 2012-65	204	53.31	-133.03
FOC26	*Paragorgia stephencairnsi*	*P. stephencairnsi*	DFO	2012	British Columbia; W of Graham Island; 2012-65	221	53.30	-133.04
FOC30	*Paragorgia stephencairnsi*	*P. stephencairnsi*	DFO	2012	British Columbia; W of Graham Island; 2012-65	318	53.48	-133.07
FOC5	*Paragorgia stephencairnsi*	*P. stephencairnsi*	DFO	2009	British Columbia; E of Graham Island; 2009-47	201	52.13	-128.90
1122230	*Sibogagorgia cauliflora*		USNM	2006	California; Davidson seamount; dive 945	2502	35.83	-122.61
2036	*Sibogagorgia cauliflora*		TU/WHOI	2009	Gulf of Mexico; DC583; J2-454	2440	28.39	-87.39

Table S2. RAD sequencing results and filtering statistics.

				STACKS filtering		**pyRAD filtering**			
Morphological Species	**Delimitation model PABSTE**	**RAD-seq data file ID**	**Total sequenced reads**	**Reads discarded due to low quality**	**Retained reads**	**Reads that passed quality filtering**	**Trimmed reads due to detection of adapters**	**Total number of retained reads**	**% of retained reads after filtering steps**
Anthomastus sp		PoC_p4_ARG	2,207,834	151,391	1,312,903	1,076,607	38,570	1,115,177	50.51
Corallium sp		COR_86121_NZ	3,826,317	305,019	3,013,324	2,555,644	22,650	2,578,294	67.38
H. imperiale-laauense		COR_64642_NZ	4,344,702	239,625	4,105,077	3,697,256	21,823	3,719,079	85.60
H. imperiale-laauense		COR_83361_NZ	3,351,944	291,963	2,588,492	2,103,039	37,016	2,140,055	63.85
H. imperiale-laauense		HEM_82674_NZ	4,455,288	364,162	3,777,461	3,216,577	40,911	3,257,488	73.12
H. imperiale-laauense		HEM_86234_NZ	2,408,325	186,372	2,142,057	1,876,187	16,195	1,892,382	78.58
H. imperiale-laauense		HEM_86286_NZ	5,324,532	446,268	4,266,674	3,577,980	42,661	3,620,641	68.00
H. imperiale-laauense		PAR_86232_NZ	6,092,276	503,438	5,206,376	4,454,042	35,518	4,489,560	73.69
Heteropolypus sp		ANT_69654_NZ	4,300,289	349,226	3,645,814	3,139,309	32,395	3,171,704	73.76
Heteropolypus sp		ANT_85040_NZ	4,498,336	393,748	3,882,304	3,333,172	26,687	3,359,859	74.69
Heteropolypus sp		ANT_85662_NZ	3,487,138	283,856	2,812,750	2,382,184	40,135	2,422,319	69.46
P. alisonae	*P. alisonae*	PAR_66097_NZ	3,007,362	220,209	2,511,717	2,219,265	21,207	2,240,472	74.50
P. arborea	*P. arborea*	PAR_41780_NZ	6,668,080	367,144	6,300,936	5,655,328	33,758	5,689,086	85.32
P. arborea	*P. arborea*	PAR_TC16_03_CAN	2,259,880	167,106	1,799,244	1,556,363	17,448	1,573,811	69.64
P. arborea	*P. pacifica*	PAR_0672_BC	4,348,226	293,526	4,054,700	3,543,642	29,284	3,572,926	82.17
P. arborea	*P. pacifica*	PAR_1007340_BCD	2,808,882	251,102	2,431,829	2,063,940	25,831	2,089,771	74.40
P. arborea	*P. pacifica*	PAR_1601_BC	4,202,185	221,386	3,980,799	3,606,334	25,434	3,631,768	86.43
P. coralloides	*P. coralloides*	PAR_063902_NES	3,244,860	185,122	3,059,738	2,746,228	20,408	2,766,636	85.26
P. coralloides	*P. coralloides*	PAR_12016_NES	3,838,375	226,253	3,612,122	3,255,440	15,945	3,271,385	85.23
P. johnsoni	*P. johnsoni*	PAR_20136_GOM	7,751,624	422,203	7,329,421	6,623,446	38,692	6,662,138	85.95
P. kaupeka	*P. kaupeka*	PAR_72152_NZ	3,909,139	268,871	2,926,960	2,531,796	26,984	2,558,780	65.46
P. kaupeka	*P. kaupeka*	PAR_82260_NZ	5,511,873	475,292	4,558,291	3,824,683	42,782	3,867,465	70.17
P. kaupeka	*P. kaupeka*	PAR_82342_NZ	4,056,319	307,425	3,339,653	2,866,786	33,020	2,899,806	71.49
P. kaupeka	*P. kaupeka*	PAR_84804_NZ	2,828,879	230,493	2,313,997	1,968,495	19,602	1,988,097	70.28
P. maunga	*P. maunga*	PAR_11369_NZ	2,536,311	252,554	2,008,028	1,619,915	34,477	1,654,392	65.23
P. stephencairnsi	*P. sp. nov*	PAR_1007316_BCD	2,257,183	151,729	1,606,180	1,371,398	36,692	1,408,090	62.38
P. stephencairnsi	*P. sp. nov*	PAR_2344_BC	2,858,311	159,073	2,699,238	2,414,787	26,272	2,441,059	85.40
P. stephencairnsi	*P. stephencairnsi*	PAR_101010_CA	6,642,673	398,317	6,244,356	5,595,342	39,240	5,634,582	84.82
P. stephencairnsi	*P. stephencairnsi*	PAR_1124300_WA	3,843,250	215,859	3,627,391	3,260,864	27,921	3,288,785	85.57
P. stephencairnsi	*P. stephencairnsi*	PAR_Agam_BC	4,322,564	260,095	4,062,469	3,619,988	26,669	3,646,657	84.36

P. stephencairnsi	*P. stephencairnsi*	PAR_C02_BCS	2,149,938	153,978	1,594,818	1,377,002	16,157	1,393,159	64.80
P. stephencairnsi	*P. stephencairnsi*	PAR_C03_BCS	2,815,327	245,051	2,264,951	1,916,452	19,800	1,936,252	68.78
P. stephencairnsi	*P. stephencairnsi*	PAR_C04_BCS	3,240,713	267,042	2,754,373	2,379,284	22,261	2,401,545	74.11
P. stephencairnsi	*P. stephencairnsi*	PAR_C05_BCS	3,465,396	294,547	2,972,548	2,538,577	25,005	2,563,582	73.98
P. stephencairnsi	*P. stephencairnsi*	PAR_C100_BCS	5,998,914	480,465	4,984,766	4,330,930	38,445	4,369,375	72.84
P. stephencairnsi	*P. stephencairnsi*	PAR_C101_BCS	5,332,619	446,452	4,652,535	4,030,919	30,684	4,061,603	76.17
P. stephencairnsi	*P. stephencairnsi*	PAR_C102_BCS	4,347,757	348,662	3,710,381	3,221,934	26,559	3,248,493	74.72
P. stephencairnsi	*P. stephencairnsi*	PAR_C104_BCS	2,720,994	210,479	2,203,568	1,893,649	19,213	1,912,862	70.30
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC25_BCD	5,479,109	453,625	4,693,804	4,073,503	34,583	4,108,086	74.98
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC26_BCD	5,242,949	477,361	4,528,629	3,778,655	37,137	3,815,792	72.78
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC30_BCD	4,417,520	337,501	3,781,973	3,312,791	32,621	3,345,412	75.73
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC5_BCD	2,834,377	239,051	2,455,041	2,109,251	23,444	2,132,695	75.24
S. cauliflora		SIB_1122230_DAV	1,701,751	123,755	1,220,077	1,016,203	26,104	1,042,307	61.25
S. cauliflora		SIB_2036_GOM	2,638,533	191,392	2,373,232	2,077,092	20,727	2,097,819	79.51
		AVERAGE	3,944,974	292,232	3,395,704	2,950,279	28,840	2,979,119	74.27
		STD DEV.	1,392,573	105,332	1,352,276	1,219,146	7,968	1,222,799	8.09

Table S3. RAD-seq **backbone** clustering and matrix statistics.

				Number of loci recovered in final data set for each taxon															
Matrix	**Min. # of taxa per locus (m)**	**Cluster thres-hold (c)**	**Total # loci in matrix**	**ANT_85040_NZ**	**COR_86121_NZ**	**HEM_82674_NZ**	**PAR_0672_BC**	**PAR_11369_NZ**	**PAR_12016_NES**	**PAR_20136_GOM**	**PAR_66097_NZ**	**PAR_82260_NZ**	**PAR_PR27_BCD**	**PoC_p4_ARG**	**SIB_2036_GOM**	**Total # of variable sites (var)**	**Total # of phylogenetically informative sites (pis)**	**pis/var**	**% of missing data**
c80d5m4p3	4	80	62,726	10,588	24,303	25,369	41,190	44,891	38,333	46,858	42,472	41,810	45,377	7,154	26,243	601,763	109,290	0.18	49.6
c80d5m6p3	6	80	35,340	8,518	18,512	19,033	28,087	29,555	27,273	30,193	28,098	28,763	29,647	5,665	19,485	388,349	81,574	0.21	38.3
c80d5m9p3	9	80	10,333	5,176	8,607	8,701	9,558	9,778	9,528	9,879	9,477	9,635	9,794	3,458	8,183	132,803	32,017	0.24	20.7
c85d5m4p3	4	85	62,318	8,692	22,647	23,757	40,930	45,124	37,243	46,984	42,725	41,154	45,540	5,918	24,943	519,766	89,806	0.17	50.1
c85d5m6p3	6	85	33,785	7,020	17,146	17,608	27,047	28,504	26,012	29,192	27,197	27,640	28,608	4,715	18,225	328,464	66,419	0.20	38.3
c85d5m9p3	9	85	9,411	4,374	7,947	8,024	8,752	8,922	8,729	9,041	8,666	8,852	8,953	2,960	7,447	109,189	26,048	0.24	20.1
c90d5m4p3	4	90	58,765	5,796	18,775	19,255	37,994	43,101	33,306	44,722	40,899	37,911	43,075	4,073	21,371	402,245	63,938	0.16	51.4
c90d5m6p3	6	90	28,923	4,674	13,826	13,932	23,212	24,715	22,141	25,263	23,694	23,804	24,705	3,226	14,961	236,051	45,285	0.19	38.5
c90d5m9p3	9	90	7,312	2,943	6,277	6,318	6,855	6,955	6,836	7,048	6,779	6,905	6,984	2,029	5,815	72,149	16,695	0.23	19.4

Table S4. RAD-seq **PHYLO** clustering and matrix statistics.

Morphological Species	**Delimitation model PABSTE**	**RAD-seq data file ID**	**Total number of clusters**	**Mean cluster depth**	**Std. dev. of cluster depth**	**Number of clusters with depth greater than 5**	**Mean depth of clusters with depth greater than 5**	**Std. dev. of cluster depth for clusters with depth greater than 5**	**Number of loci**	**Number of loci with >5 depth coverage**	**Number of loci with >5 depth and passed paralog filter**	**Number of sites across loci**	**Number of polymorphic sites**	**Frequency of polymorphic sites**
Anthomastus sp		PoC_p4_ARG	142,551	5.7	11.5	54,060	11.9	16.9	142,551	54,050	46,191	3,917,645	12,866	0.0032841
Corallium sp		COR_86121_NZ	176,085	9.6	34.3	85,869	17.8	47.8	176,085	85,816	74,563	6,332,203	29,627	0.0046788
H. imperiale-laauense		COR_64642_NZ	177,897	4.3	26.1	29,162	16.7	62.8	177,897	29,125	14,235	1,205,608	6,515	0.0054039
H. imperiale-laauense		COR_83361_NZ	190,447	7.2	63.8	74,091	15.7	101.7	190,447	74,069	63,941	5,426,154	20,057	0.0036964
H. imperiale-laauense		HEM_82674_NZ	219,724	8.9	53.4	92,866	18.8	81.1	219,724	92,810	80,468	6,834,388	29,969	0.0043850
H. imperiale-laauense		HEM_86234_NZ	155,795	7.8	35.4	70,304	14.7	51.8	155,795	70,274	57,724	4,900,969	25,071	0.0051155
H. imperiale-laauense		HEM_86286_NZ	252,395	9.0	76.6	96,846	20.8	122.8	252,395	96,772	81,349	6,912,750	30,928	0.0044741
H. imperiale-laauense		PAR_86232_NZ	219,190	12.6	95.8	106,265	24.0	136.7	219,190	106,145	85,667	7,282,625	32,347	0.0044417
Heteropolypus sp		ANT_69654_NZ	151,154	15.1	52.2	88,266	24.7	66.7	151,154	88,178	80,757	6,864,699	18,791	0.0027373
Heteropolypus sp		ANT_85040_NZ	159,357	15.1	42.4	92,201	24.8	53.7	159,357	92,099	83,695	7,115,901	27,473	0.0038608
Heteropolypus sp		ANT_85662_NZ	165,100	10.5	120.2	82,840	19.2	169.2	165,100	82,793	74,272	6,309,974	23,863	0.0037818
P. alisonae	*P. alisonae*	PAR_66097_NZ	134,879	11.1	54.8	79,570	17.5	70.7	134,879	79,531	70,095	5,954,159	23,831	0.0040024
P. arborea	*P. arborea*	PAR_41780_NZ	107,360	17.7	78.9	76,515	24.0	92.7	107,360	76,367	68,928	5,855,658	20,899	0.0035690
P. arborea	*P. arborea*	PAR_TC16_03_CAN	138,933	7.7	46.4	62,427	14.9	68.6	138,933	62,399	54,495	4,625,417	13,801	0.0029837
P. arborea	*P. pacifica*	PAR_0672_BC	116,545	21.4	71.8	85,334	28.5	82.8	116,545	85,165	77,189	6,559,823	25,068	0.0038214
P. arborea	*P. pacifica*	PAR_1007340_BCD	149,606	9.9	30.4	76,311	17.6	41.0	149,606	76,259	67,539	5,736,203	22,134	0.0038587
P. arborea	*P. pacifica*	PAR_1601_BC	118,243	21.7	65.9	85,263	29.3	76.2	118,243	85,074	77,035	6,547,638	24,879	0.0037997
P. coralloides	*P. coralloides*	PAR_063902_NES	132,778	20.1	61.7	87,919	29.4	74.1	132,778	87,756	79,692	6,774,134	26,579	0.0039236
P. coralloides	*P. coralloides*	PAR_12016_NES	129,675	36.8	207.3	90,736	51.8	246.3	129,675	90,474	82,903	7,049,554	21,990	0.0031193
P. johnsoni	*P. johnsoni*	PAR_20136_GOM	122,432	29.8	487.2	89,665	40.0	568.9	122,432	89,630	81,301	6,911,399	23,857	0.0034518
P. kaupeka	*P. kaupeka*	PAR_72152_NZ	132,520	12.7	197.2	77,976	20.2	256.8	132,520	77,929	69,780	5,926,828	16,005	0.0027004
P. kaupeka	*P. kaupeka*	PAR_82260_NZ	186,768	13.1	77.9	89,892	25.4	110.9	186,768	89,766	80,556	6,845,659	17,710	0.0025870
P. kaupeka	*P. kaupeka*	PAR_82342_NZ	148,202	12.6	43.0	83,517	21.0	55.8	148,202	83,429	74,747	6,349,820	16,935	0.0026670
P. kaupeka	*P. kaupeka*	PAR_84804_NZ	124,756	10.5	119.9	69,174	17.3	160.6	124,756	69,119	61,022	5,182,009	15,300	0.0029525
P. maunga	*P. maunga*	PAR_11369_NZ	113,838	19.3	161.8	84,649	25.2	187.3	113,838	84,510	76,038	6,461,976	26,079	0.0040358
P. stephencairnsi	*P. sp nov*	PAR_1007316_BCD	166,722	5.9	17.7	60,649	12.9	27.9	166,722	60,636	51,368	4,357,427	19,584	0.0044944

P. stephencairnsi	*P. sp nov*	PAR_2344_BC	125,366	13.4	51.7	80,271	19.9	63.7	125,366	80,218	72,194	6,132,435	25,912	0.0042254
P. stephencairnsi	*P. stephencairnsi*	PAR_101010_CA	115,888	33.2	134.3	92,526	41.1	149.3	115,888	92,225	84,367	7,173,308	24,279	0.0033846
P. stephencairnsi	*P. stephencairnsi*	PAR_1124300_WA	109,466	20.9	103.4	83,749	26.8	117.6	109,466	83,645	76,157	6,471,962	21,532	0.0033270
P. stephencairnsi	*P. stephencairnsi*	PAR_Agam_BC	131,554	19.3	95.4	87,778	28.1	115.8	131,554	87,648	79,612	6,766,163	23,143	0.0034204
P. stephencairnsi	*P. stephencairnsi*	PAR_C02_BCS	111,985	9.0	22.7	63,618	14.2	29.0	111,985	63,597	56,380	4,786,614	16,361	0.0034181
P. stephencairnsi	*P. stephencairnsi*	PAR_C03_BCS	116,493	11.7	41.1	75,396	17.0	50.3	116,493	75,356	67,485	5,732,283	20,104	0.0035072
P. stephencairnsi	*P. stephencairnsi*	PAR_C04_BCS	128,452	13.0	47.9	81,845	19.4	59.1	128,452	81,792	73,789	6,269,830	21,010	0.0033510
P. stephencairnsi	*P. stephencairnsi*	PAR_C05_BCS	128,134	13.9	64.4	83,590	20.3	79.0	128,134	83,533	75,306	6,399,562	21,288	0.0033265
P. stephencairnsi	*P. stephencairnsi*	PAR_C100_BCS	152,749	19.9	55.9	92,858	31.6	69.2	152,749	92,693	84,309	7,168,326	22,907	0.0031956
P. stephencairnsi	*P. stephencairnsi*	PAR_C101_BCS	152,447	18.4	82.8	92,630	29.2	104.8	152,447	92,487	83,853	7,129,265	23,371	0.0032782
P. stephencairnsi	*P. stephencairnsi*	PAR_C102_BCS	131,680	17.3	46.6	88,133	24.9	55.3	131,680	88,038	79,983	6,798,340	21,816	0.0032090
P. stephencairnsi	*P. stephencairnsi*	PAR_C104_BCS	119,764	11.4	29.6	74,773	17.0	36.3	119,764	74,738	67,146	5,703,606	18,976	0.0033270
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC25_BCD	154,768	18.3	56.3	93,419	29.2	70.4	154,768	93,284	84,570	7,190,050	24,801	0.0034494
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC26_BCD	440,488	6.3	26.0	107,701	20.5	49.9	440,488	107,621	94,298	8,008,313	28,868	0.0036048
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC30_BCD	314,181	7.6	44.3	95,360	21.1	78.8	314,181	95,276	84,774	7,202,768	24,900	0.0034570
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC5_BCD	197,822	7.5	60.1	75,192	16.9	96.8	197,822	75,145	66,755	5,669,791	19,056	0.0033610
S. cauliflora		SIB_1122230_DAV	123,947	5.5	29.3	40,167	12.8	50.7	123,947	40,157	33,058	2,804,036	11,864	0.0042310
S. cauliflora		SIB_2036_GOM	127,391	11.3	36.4	71,374	18.6	47.3	127,391	71,315	63,094	5,358,510	17,312	0.0032307
		AVERAGE	157,171	13.9	76.4	80,744	22.6	99.0	157,171	80,658	71,425	6,068,313	21,810	0.0036393
		STANDARD DEV	59,920	7.2	77.0	15,153	8.0	89.6	59,920	15,119	14,833	1,262,657	5,318	0.0006234

Table S5. Nucleotide diversity and error rate estimates per specimen based on the **PHYLO** matrix

Morphological Species	**PABSTE**	**RAD-seq data file ID**	**Nucleotide diversity (Pi)**	**Error Rate**
Anthomastus sp		PoC_p4_ARG	0.01295412	0.0029032
Corallium sp		COR_86121_NZ	0.01467291	0.00258843
*H. imperiale-laauense**		COR_64642_NZ	0.04406216	0.01630744
H. imperiale-laauense		COR_83361_NZ	0.01399387	0.00269528
H. imperiale-laauense		HEM_82674_NZ	0.01440962	0.00268885
H. imperiale-laauense		HEM_86234_NZ	0.01676557	0.00254558
H. imperiale-laauense		HEM_86286_NZ	0.01526906	0.00234296
H. imperiale-laauense		PAR_86232_NZ	0.01633107	0.00217237
Heteropolypus sp		ANT_69654_NZ	0.00936378	0.00151989
Heteropolypus sp		ANT_85040_NZ	0.01122064	0.00146665
Heteropolypus sp		ANT_85662_NZ	0.01161827	0.00189506
P. alisonae	*P. alisonae*	PAR_66097_NZ	0.0127996	0.00205016
P. arborea	*P. arborea*	PAR_41780_NZ	0.01209999	0.00186767
P. arborea	*P. arborea*	PAR_TC16_03_CAN	0.01293451	0.00241858
P. arborea	*P. pacifica*	PAR_0672_BC	0.01189447	0.00170062
P. arborea	*P. pacifica*	PAR_1007340_BCD	0.01285038	0.00223148
P. arborea	*P. pacifica*	PAR_1601_BC	0.01205019	0.00164477
P. coralloides	*P. coralloides*	PAR_063902_NES	0.01182997	0.00146894
P. coralloides	*P. coralloides*	PAR_12016_NES	0.01035726	0.00116583
P. johnsoni	*P. johnsoni*	PAR_20136_GOM	0.01159218	0.00137491
P. kaupeka	*P. kaupeka*	PAR_72152_NZ	0.01085919	0.00209157
P. kaupeka	*P. kaupeka*	PAR_82260_NZ	0.01077562	0.00211998
P. kaupeka	*P. kaupeka*	PAR_82342_NZ	0.01082052	0.00225821
P. kaupeka	*P. kaupeka*	PAR_84804_NZ	0.01169178	0.00221668
P. maunga	*P. maunga*	PAR_11369_NZ	0.01225252	0.00176518
P. stephencairnsi	*P. sp. nov*	PAR_1007316_BCD	0.01488811	0.00295065
P. stephencairnsi	*P. sp. nov*	PAR_2344_BC	0.01260347	0.00188425
P. stephencairnsi	*P. stephencairnsi*	PAR_101010_CA	0.01063818	0.00134692
P. stephencairnsi	*P. stephencairnsi*	PAR_1124300_WA	0.01092481	0.00163497
P. stephencairnsi	*P. stephencairnsi*	PAR_Agam_BC	0.01117934	0.00151902
P. stephencairnsi	*P. stephencairnsi*	PAR_C02_BCS	0.01180863	0.00194975
P. stephencairnsi	*P. stephencairnsi*	PAR_C03_BCS	0.01129071	0.00175198
P. stephencairnsi	*P. stephencairnsi*	PAR_C04_BCS	0.01098867	0.00171446
P. stephencairnsi	*P. stephencairnsi*	PAR_C05_BCS	0.01117442	0.00167389
P. stephencairnsi	*P. stephencairnsi*	PAR_C100_BCS	0.01063313	0.00155401
P. stephencairnsi	*P. stephencairnsi*	PAR_C101_BCS	0.0109634	0.00160305
P. stephencairnsi	*P. stephencairnsi*	PAR_C102_BCS	0.01060609	0.00159815
P. stephencairnsi	*P. stephencairnsi*	PAR_C104_BCS	0.01124495	0.00175065
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC25_BCD	0.01104859	0.00154584
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC26_BCD	0.01263196	0.00180015
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC30_BCD	0.01196811	0.00182162
P. stephencairnsi	*P. stephencairnsi*	PAR_FOC5_BCD	0.0119129	0.00186229
S. cauliflora		SIB_1122230_DAV	0.01525699	0.00328691
S. cauliflora		SIB_2036_GOM	0.01214983	0.00221517
		AVERAGE	0.01291599	0.00228532
		SD	0.00513429	0.00223806

*Excluded from calculations due to low number of loci

Table S6. Nucleotide diversity and error rate estimates per species based on the **PHYLO** matrix

		Nucleeotide diversity (Pi)			**Per-site sequence error rate (Epsilon)**		
Morphological Species	**Delimitation model PABSTE**	**mean**	**SD**	**SE**	**mean**	**SD**	**SE**
Anthomastus sp		0.01295412			0.00290320		
Corallium sp		0.01467291			0.00258843		
H. imperiale-laauense		0.01935775	0.01094005	0.00489254	0.00447727	0.00522005	0.00233448
Heteropolypus sp		0.01073423	0.00120338	0.00069477	0.00162720	0.00023350	0.00013481
P. alisonae	*P. alisonae*	0.01279960			0.00205016		
P. arborea	*P. arborea*	0.01245029	0.00056582	0.00040010	0.00193813	0.00041487	0.00029336
P. arborea	*P. pacifica*	0.01073423	0.00120338	0.00069477	0.00162720	0.00023350	0.00013481
P. coralloides	*P. coralloides*	0.01245029	0.00056582	0.00040010	0.00193813	0.00041487	0.00029336
P. johnsoni	*P. johnsoni*	0.01159218			0.00137491		
P. kaupeka	*P. kaupeka*	0.01241323	0.00175196	0.00087598	0.00229768	0.00048923	0.00024462
P. maunga	*P. maunga*	0.01225252			0.00176518		
P. stephencairnsi	*P. sp. nov*	0.01290646	0.00280248	0.00198165	0.00229281	0.00093033	0.00065784
P. stephencairnsi	*P. stephencairnsi*	0.01137950	0.00066172	0.00016543	0.00169174	0.00016364	0.00004091
S. cauliflora		0.01370341	0.00219709	0.00155358	0.00275104	0.00075783	0.00053587

Table S7. Predictions of # of RAD-tags in octocorals using PstI. Data for *Nematostella vectensis* obtained from the U.S. Joint Genome Institute (JGI-DOE) database. Data for *Acropora digitifera, Hydra vulgaris*, and *Alantina moseri* obtained from the U.S. National Center for Biotechnology Information (NCBI) WGS database. Observed frequency of recognition sequences and calculated probability based on a trinucleotide genome composition model were generated following the methodology described by Herrera et al. (2014). Octocoral genome size ranges were obtained by Luisa Dueñas from gorgoniid octocorals through flow cytomery at the Universidad de los Andes, Bogota, Colombia. Abbreviation: restriction sites (RS).

Cnidarians with sequenced genomes

Species	**Common name**	**C-value**	**Genome size (Mbp) Unambiguous**	**Observed frequency of PstI RS per bp**	**Prob. of PstI recognition site per bp based on trinucleotide model**
Nematostella vectensis	Starlet Anemone		297.39	0.00016661	0.00019608
Acropora digitifera	Staghorn Coral		364.97	0.00021313	0.00022777
Hydra vulgaris	Hydra		1189.96	0.00010830	0.00010871
Alatina moseri	Sea Wasp		1544.15	0.00020617	0.00021637

Predictions of # of PstI RS in known octocoral genome size range

Octocoral C-value	**Octocoral Genome size (Mbp)**	**Predicted # of PstI RS based on *N. vectensis* observed frequency**	**Predicted # of PstI RS based on *A. digitifera* observed frequency**	**Predicted # of PstI RS based on *H. vulgaris* observed frequency**	**Predicted # of PstI RS based on *A. moseri* observed frequency**
0.3	293.40	48,882.95	62,533.09	31,774.37	60,489.13
0.5	489.00	81,471.59	104,221.81	52,957.29	100,815.22

Octocoral C-value	**Octocoral Genome size (Mbp)**	**Predicted # of PstI RS based on *N. vectensis* trinucleotide genome composition probability**	**Predicted # of PstI RS based on *A. digitifera* trinucleotide genome composition probability**	**Predicted # of PstI RS based on *H. vulgaris* trinucleotide genome composition probability**	**Predicted # of PstI RS based on *A. moseri* trinucleotide genome composition probability**
0.3	293.40	57,529.94	66,828.40	31,895.01	63,482.46
0.5	489.00	95,883.23	111,380.67	53,158.36	105,804.09

Predictions of # of PstI RAD-tags in known octocoral genome size range

Octocoral C-value	**Octocoral Genome size (Mbp)**	**Predicted # of PstI RS based on *N. vectensis* observed frequency**	**Predicted # of PstI RS based on *A. digitifera* observed frequency**	**Predicted # of PstI RS based on *H. vulgaris* observed frequency**	**Predicted # of PstI RS based on *A. moseri* observed frequency**
0.3	293.40	97,765.91	125,066.17	63,548.75	120,978.26
0.5	489.00	162,943.18	208,443.62	105,914.58	201,630.43

Octocoral C-value	**Octocoral Genome size (Mbp)**	**Predicted # of PstI RS based on *N. vectensis* trinucleotide genome composition probability**	**Predicted # of PstI RS based on *A. digitifera* trinucleotide genome composition probability**	**Predicted # of PstI RS based on *H. vulgaris* trinucleotide genome composition probability**	**Predicted # of PstI RS based on *A. moseri* trinucleotide genome composition probability**
0.3	293.40	115,059.87	133,656.80	63,790.03	126,964.91
0.5	489.00	191,766.45	222,761.34	106,316.71	211,608.19

Table S8. Collection information for all specimens in the clade AC with available mtMutS sequences

Species	ID	Genbank	Collection	Catalog Number	Date	Locality	Depth (m)	Lat.	Lon.	Taxonomic remarks
Anthomasttus ritteri		DQ302816	K. McFadeen		1998	Off Pebble Beach: Californa: USA	300	36.58	-122.10	
Anthomastus cf grandiflorus		KC984603								
Anthomastus robustus delta		KC984604								
Anthomastus sp	p4		WHOI	p4	2011	Patagonian shelf				
Chelidonisis aurantiaca mexicana		KC788274								
Corallium elatius		AB700134								
Corallium elatius		AB700135								
Corallium japonicum		AB595189								
Corallium kishinouyei		GQ293300	USNM	1072441	2003	Off Laysan Island: Hawaii: USA	1490	25.70	-171.45	
Corallium kishinouyei		KC782353								
Corallium konojoi		AB595190								
Corallium regale		AF385321								
Corallium rubrum		AB700136								
Corallium secundum		GQ293303	USNM	1010758	2001	Off Maui: Hawaii: USA	240	20.88	-156.73	
Corallium secundum		KC782347								
Corallium sp	86121		NIWA	86121	2012	Kermadec Ridge; Colville ridge volcano; TAN1213/18	380	-30.19	179.72	
Corallium sp	Coralliumsp56807		USNM	56807	1978	USA, Hawaii, Oahu Island, Makapuu Point	366	21.30	-157.53	
Corallium sp		KC788270								
Corallium tortuosum		GQ293306	USNM	1089600	2003	New Caledonia	470-621	-23.71	168.26	
Hemicorallium ducale		DQ297416								
Hemicorallium ducale		EF060050								
Hemicorallium ducale		EU293805								
Hemicorallium imperiale		KC782352								
Hemicorallium imperiale		KC782355								
Hemicorallium imperiale-laauense	64642		NIWA	64642	2010	Kermadec Ridge; Rumble II East seamount cone; TAN1007/97	1050	-35.42	178.65	
Hemicorallium imperiale-laauense	82674		NIWA	82674	2012	Kermadec Ridge; Site SM2aa, summit of Whakatane Seamoun	878	-36.81	177.47	
Hemicorallium imperiale-laauense	83361		NIWA	83361	2012	Bay of Plenty; Site SM1b, Matatara Knoll; TAN1206/168	948	-37.19	176.98	
Hemicorallium imperiale-laauense	86232		NIWA	86232	2012	Kermadec Ridge; Northeast pimple volcano; TAN1213/22	483	-30.08	179.82	
Hemicorallium imperiale-laauense	86234		NIWA	86234	2012	Kermadec Ridge; Northeast pimple volcano; TAN1213/22	483	-30.08	179.82	
Hemicorallium imperiale-laauense	86286		NIWA	86286	2012	Kermadec Ridge; Havre volcano; TAN1213/30	860	-31.13	-179.05	
Hemicorallium imperiale-laauense	Claauense1072452		USNM	1072452	2003	USA, Hawaii, Laysan Island	1509	25.70	-171.44	
Hemicorallium imperiale-laauense		GQ293301	USNM	1071433	2004	Off Keahole Point: Hawaii Island: Hawaii: USA	867	19.80	-156.13	
Hemicorallium imperiale-laauense		GQ293302	USNM	1075800	2004	Pratt Seamount: Alaska: USA	1627	56.32	-142.44	
Hemicorallium laauense		KC782348								
Hemicorallium niobe		EF060051								
Hemicorallium niobe		KC788267								
Heteropolypus sp	69654		NIWA	69654	2011	Chatam Rise; TRIP3306/78	495	-44.33	-177.22	
Heteropolypus sp	85040		NIWA	85040	2012	Chatam Rise; TAN1208/22	2098	-42.59	179.42	
Heteropolypus sp	85662		NIWA	85662	2012	Chatam Rise; TAN1208/61	1931	-42.59	179.59	
Ideogorgia capensis		GQ342502								
Paragorgia alisonae	66097		NIWA	66097	2008	Cambell Plateau; TRIP2718/50	875	-50.02	175.00	
Paragorgia alisonae	Palisonae3312	JX128349	NIWA	3312	1998	New Zealand, Otara Hill, 1171/25, Z9596	980	-48.02	166.08	type

Paragorgia alisonae	Palisonae3313		NIWA	3313	1998	New Zealand, Otara Hill, 1171/25, Z9596	980	-48.02	166.08	type
Paragorgia alisonae	Palisonae3315		NIWA	3315	1998	New Zealand, Otara Hill, 1171/24, Z9595	940	-48.02	166.10	type
Paragorgia alisonae	Palisonae3316		NIWA	3316	1997	New Zealand, TAN9713/037, Z8981	1041	-44.96	174.19	type
Paragorgia alisonae	Palisonae3317		NIWA	3317	1998	New Zealand, TRIP1171/12, Z9583	935	-48.03	166.10	type
Paragorgia alisonae	Palisonae42002		NIWA	42002	2008	New Zealand, TRIP2551/258	930.0	-44.7850	-176.5833333333	
Paragorgia alisonae	Palisonae44606		NIWA	44606	2007	New Zealand, TRIP2521/9	1068	-44.7433	-177.0533333	
Paragorgia alisonae	ParagospValpo046			Valpo046		Chile, Valparaiso, deep seamount				
Paragorgia aotearoa		GQ293305	NIWA	3325	1996	New Zealand, Mt. Muck (Box Hill Complex), TAN9609/40	700	-42.83	176.92	type
Paragorgia arborea	0672		RBCM	011-00067-002	2004	British Columbia; VE14280		53.33	-135.66	
Paragorgia arborea	1601		RBCM	011-00160-001	2009	British Columbia; VE14444	695	53.31	-135.58	
Paragorgia arborea	41780	JX124558	NIWA	41870	2008	New Zealand, TRIP2617/120	600	-44.52	175.78	
Paragorgia arborea	1007340	JX124556	USNM	1007340	2001	British Columbia; Vancouver Island	1168	48.44	-126.38	
Paragorgia arborea	Paragosp1014919	JX124578	USNM	1014919	2003	Davidson Seamount, California, USA	1313	35.70	-122.70	Sanchez 2005
Paragorgia arborea	Paragosp1027060	JX124560	USNM	1027060	2003	Pioneer Seamount, South of farallon Islands, California, USA	1712	37.40	-123.44	
Paragorgia arborea	Paragosp1075738	JX124526	USNM	1075738	2004	Dickins Seamount, Gulf of Alaska, USA	760	54.55	-136.84	
Paragorgia arborea	Paragosp1075744	JX124565	USNM	1075744	2004	Dickins Seamount, Gulf of Alaska, USA	851	54.51	-136.91	
Paragorgia arborea	Paragosp1075745	JX124602	USNM	1075745	2004	Dickins Seamount, Gulf of Alaska, USA	849	54.51	-136.91	
Paragorgia arborea	Paragosp1075746	JX124540	USNM	1075746	2004	Welker Seamount, Gulf of Alaska, USA	780	55.05	-140.31	
Paragorgia arborea	Paragosp1075753	JX124527	USNM	1075753	2004	Welker Seamount, Gulf of Alaska, USA	1112	55.07	-140.41	
Paragorgia arborea	Paragosp1075754	JX124593	USNM	1075754	2004	Welker Seamount. Gulf of Alaska, USA	1084	55.07	-140.41	
Paragorgia arborea	Paragosp1075760	JX124530	USNM	1075760	2004	Pratt Seamount, Gulf of Alaska, USA	959	56.17	-142.70	
Paragorgia arborea	Paragosp1075761	JX124576	USNM	1075761	2004	Pratt Seamount, Gulf of Alaska, USA	941	56.17	-142.70	
Paragorgia arborea	Paragosp1075766	JX124541	USNM	1075766	2004	Welker Seamount, Gulf of Alaska, USA	1114	55.07	-140.41	
Paragorgia arborea	Paragosp17971	JX124592	NIWA	17971	2004	New Zealand, 1172/06, Z9566	1235	-44.80	-177.12	
Paragorgia arborea	Paragosp200701	JX124587	B. Stone	20070178B01	2004	Gulf of Alaska, USA	867	55.91	-154.02	
Paragorgia arborea	Paragosp44156	JX124563	NIWA	44156	2007	New Zealand, TRIP2416/54	720-741	-47.47	177.02	
Paragorgia arborea	Paragosp46314	JX124594	NIWA	46314	2008	New Zealand, TRIP2324/48	843-998	-50.05	174.73	
Paragorgia arborea	Paragosp46315	JX124569	NIWA	46315	2008	New Zealand, TRIP2571/65	888-101	-47.55	177.86	
Paragorgia arborea	Paragosp46316	JX124573	NIWA	46316	2007	New Zealand, TRIP2617/120	600	-44.52	175.77	
Paragorgia arborea	Paragosp46317	JX124603	NIWA	46317	2008	New Zealand, TRIP2494/13	931-102	-47.58	177.78	
Paragorgia arborea	Paragosp46318	JX124583	NIWA	46318	2008	New Zealand, TRIP2551/254	794-987	-44.73	-177.04	
Paragorgia arborea	Paragosp46319	JX124595	NIWA	46319	2006	New Zealand, TRIP2614		-49.50	176.00	
Paragorgia arborea	Paragosp46377	JX124549	NIWA	46377	1927	New Zealand, TRIP2571/53	952-111	-50.00	176.06	
Paragorgia arborea	Paragosp56389		USNM	56389	1962	USA, Hawaii, Bushnell Seamount	1920	18.55	-155.44	
Paragorgia arborea	ParagospDAVI1	JX124577	A. Andre	DAVI1	2002	Davidson Seamount, California, USA	1313	35.75	-122.70	
Paragorgia arborea	ParagospDAVI3	JX124542	A. Andre	DAVI3	2004	Davidson Seamount, California, USA	1313	35.75	-122.70	
Paragorgia arborea	Paragospnizinski	JX124547	USNM	1120444	2008	off Maryland, USA	400	37.06	-74.62	
Paragorgia arborea	Parborea100758	JX124584	USNM	100758	1994	Aleutian Islands		52.00	-170.00	
Paragorgia arborea	Parborea100817	JX124539	USNM	100817	1994	Atka Island, Andreanof Islands, Aleutian Islands		53.00	-174.00	
Paragorgia arborea	Parborea100818	JX124597	USNM	100818	1994	Semisopochnoi Island, Rat Islands, Aleutian Islands		52.17	179.72	
Paragorgia arborea	Parborea100843	JX124529	USNM	100843	1994	Tanaga Island, Andreanof Islands, Aleutian Islands		52.00	-178.00	
Paragorgia arborea	Parborea100846	JX124538	USNM	100846	1994	Yunaska Island, Islands of Four Mountains, Aleutian Islands		53.00	-171.00	
Paragorgia arborea	Parborea1010787	JX124528	USNM	1010787	2000	Norfolk Canyon, Virginia, USA	375-489	37.07	-74.66	
Paragorgia arborea	Parborea1011097	JX124545	USNM	1011097	2002	Buldir Reef, Rat Islands, Aleutian Islands	160	51.96	176.83	
Paragorgia arborea	Parborea1011360	JX124582	USNM	1011360	2001	off Umnak Island, Fox Islands, Aleutian Islands	102	53.68	-169.11	

Paragorgia arborea	Parborea1092764	JX124589	USNM	1092764	2000	East of Virginia Beach, Virginia, USA	375-489	37.07	-74.66	
Paragorgia arborea	Parborea1092765	JX124520	USNM	1092765	2000	East of Virginia Beach, Virginia, USA	375-489	37.07	-74.66	
Paragorgia arborea	Parborea1092766	JX124554	USNM	1092766	2000	East of Virginia Beach, Virginia, USA	375-489	37.07	-74.66	
Paragorgia arborea	Parborea17969	JX124607	NIWA	17969	2002	New Zealand, 1621/18, Z11010	900	-44.74	-177.19	
Paragorgia arborea	Parborea17970	JX124605	NIWA	17970	2001	New Zealand, AEX0101/80, Z10956	753	-44.74	-177.19	
Paragorgia arborea	Parborea25527	JX124548	NIWA	25527	2004	New Zealand, TAN0408/23	826	-42.83	177.42	
Paragorgia arborea	Parborea28123	JX124546	NIWA	28123	2000	New Zealand, 1390/12, Z11161	872	-47.31	165.83	
Paragorgia arborea	Parborea28154	JX124553	NIWA	28154	1981	New Zealand, T16	427	-43.35	178.66	
Paragorgia arborea	Parborea28156	JX124610	NIWA	28156	2002	New Zealand, 1621/08, Z11008	920	-33.92	167.92	
Paragorgia arborea	Parborea28157	JX124531	NIWA	28157	1999	New Zealand, TRIP1223/29	959	-44.58	-177.88	
Paragorgia arborea	Parborea28158	JX124566	NIWA	28158	2001	New Zealand, AEX0101/80, Z10907	753	-44.74	-177.18	
Paragorgia arborea	Parborea28160	JX124523	NIWA	28160	2001	New Zealand				
Paragorgia arborea	Parborea28161	JX124568	NIWA	28161	2001	New Zealand, AEX0101/80, Z10920	753	-44.74	-176.81	Sanchez 2005
Paragorgia arborea	Parborea28392	JX124575	NIWA	28392	2001	New Zealand, AEX0101/80, Z10956	753	-44.74	-177.19	
Paragorgia arborea	Parborea28422	JX124601	NIWA	28422	2001	New Zealand, AEX0101/80, Z10920	753	-44.74	-176.81	
Paragorgia arborea	Parborea28425	JX124550	NIWA	28425	1997	New Zealand, TAN9713/52, Z8979	858	-44.45	-179.96	
Paragorgia arborea	Parborea3308	JX124552	NIWA	3308	2002	New Zealand, Z10987	1225	-33.93	167.92	Sanchez 2005
Paragorgia arborea	Parborea3309	JX124535	NIWA	3309	2002	New Zealand, Z11009	955	-33.93	167.91	Sanchez 2005
Paragorgia arborea	Parborea3310	GQ293311	NIWA	3310	1999	New Zealand, Z9862	687	-44.75	174.82	Sanchez 2005
Paragorgia arborea	Parborea3311	JX124525	NIWA	3311	1996	New Zealand, Southern Havre trough, X700	1525	-35.84	177.91	Sanchez 2005
Paragorgia arborea	Parborea33559	JX124559	USNM	33559	1878	Fishing Banks, North Carolina, USA	457	36.00	-74.00	
Paragorgia arborea	Parborea33561	JX124562	USNM	33561		Off NE North America, USA				
Paragorgia arborea	Parborea4089	JX124591	USNM	4089		Sable Island, 50 Mile E Of E Light, Nova Scotia, Canada	512	43.90	-58.80	
Paragorgia arborea	Parborea4091	JX124579	USNM	4091	1879	Banquereau Bank, Nova Scotia, Canada	366	44.58	-57.68	
Paragorgia arborea	Parborea4178B	JX124524	USNM	1123932	2002	South of Trinity Islands, Aleutian Islands	746	55.87	-154.06	
Paragorgia arborea	Parborea41829	JX124585	NIWA	41829	2006	New Zealand, TRIP2324/76	1044	-47.25	178.33	
Paragorgia arborea	Parborea41854	JX124611	NIWA	41854	2007	New Zealand, TRIP2494/10	867-986	-47.53	177.87	
Paragorgia arborea	Parborea41999	JX124588	NIWA	41999	2007	New Zealand, TRIP2551/50	1203-12	-44.50	-174.79	
Paragorgia arborea	Parborea42001	JX124574	NIWA	42001	2007	New Zealand, TRIP2551/55	1283-13	-44.50	-174.82	
Paragorgia arborea	Parborea4238	JX124608	USNM	4238	1879	Banquereau Bank, South Of, Nova Scotia, Canada	457	43.90	-58.67	
Paragorgia arborea	Parborea4242	JX124567	USNM	4242	1879	Grand Banks, W Part Of, Newfoundland, Canada		45.00	-54.00	
Paragorgia arborea	Parborea44608	JX124537	NIWA	44608	2007	New Zealand, TRIP2506/81	1106-13	-46.91	171.88	
Paragorgia arborea	Parborea44609	JX124534	NIWA	44609	2007	New Zealand, TRIP2506/135	870-967	-47.53	177.92	
Paragorgia arborea	Parborea4569	JX124596	USNM	4569	1879	Sable Island Bank, Nova Scotia, Canada	457	43.42	-60.00	
Paragorgia arborea	Parborea46320	JX124570	NIWA	46320	2008	New Zealand, TRIP2320/70	750-855	-46.48	170.60	
Paragorgia arborea	Parborea50890	JX124521	USNM	50890	1927	Burdwood Bank, S Of Falkland Islands, Scotia Sea		-54.50	-59.10	
Paragorgia arborea	Parborea80838	JX124544	USNM	80838	1979	Baltimore Canyon, Off Eastern Shore, Maryland, USA	480	38.17	-73.84	
Paragorgia arborea	Parborea80936	JX124543	USNM	80936	1979	Lydonia Canyon, Massachusetts, USA	680-370	40.38	-67.66	
Paragorgia arborea	Parborea80937	GQ293312	USNM	80937	1979	Lydonia Canyon, Massachusetts, USA	613-430	40.38	-67.66	
Paragorgia arborea	ParboreaJ2095271	JX124532	USNM	1123936	2004	Amlia Island, Andreanof Islands, Aleutian Islands	843	51.81	-173.83	
Paragorgia arborea	ParboreaJ2095272	JX124580	USNM	1123935	2004	Amlia Island, Andreanof Islands, Aleutian Islands	843	51.81	-173.83	
Paragorgia arborea	ParboreaJ2099211	JX124604	USNM	1123937	2004	Adak Canyon, Andreanof Islands, Aleutian Islands	1269	51.51	-177.04	
Paragorgia arborea	ParboreaJ210441	JX124536	USNM	1123938	2004	Amchitka Pass, Andreanof Islands, Aleutian Islands	747	51.72	-179.58	
Paragorgia arborea	ParboreaJ210462	JX124555	USNM	1123934	2004	Amchitka Pass, Andreanof Islands, Aleutian Islands	857	51.68	-179.58	
Paragorgia arborea	ParboreaMCZ1572	JX124606	MCZ	15721	2002	Atlantic Ocean	480	37.67	-74.65	

Paragorgia arborea	ParboreaMCZ2805	JX124551	MCZ	28057	2001	Nantucket Island, Massachussetts, USA	156	41.97	-65.87	
Paragorgia arborea	ParboreaMCZ2869	JX124561	MCZ	28697	1878	off Maine, USA	245	42.60	-65.73	
Paragorgia arborea	ParboreaMCZ5124	JX124600	MCZ	51244	1979	Georges Bank, Massachusetts, USA		41.00	-67.00	
Paragorgia arborea	ParboreaMNHN0411		MNHN	0411	1997	France, North Altlantic Ocean, France	700	48.83	-11.33	
Paragorgia arborea	ParboreaMNHN04	JX124609	MNHN	412		off Norway		66.70	11.60	
Paragorgia arborea	ParboreaMNHN04	JX124564	MNHN	422	2002	Trondhjems Fjord, Norway		63.50	10.50	
Paragorgia arborea	Parboreap33560	JX124572	USNM	33560		Fishing Banks, North Carolina, USA		45.00	-53.50	
Paragorgia arborea	ParboreaT661A10	JX124590	USNM	1122237	2002	Rodriguez Seamount, California, USA	894.5	34.06	-121.08	
Paragorgia arborea	ParboreaT662A28	JX124522	USNM	1122240	2002	San Juan Seamount, California, USA	1362.9	32.97	-121.04	
Paragorgia arborea	ParboreaT662A29	JX124586	USNM	1122233	2002	San Juan Seamount, California, USA	1360.8	32.97	-121.04	
Paragorgia arborea	ParboreaZ11166	JX124533	NIWA	76238	2007	New Zeland	891	-44.44	175.54	
Paragorgia arborea	ParboreaZC0706	JX124598	B. Stone	ZC0706ROV01		Zhemchung Canyon, Bering Sea	171	-47.54	177.93	
Paragorgia arborea	PcfarboreJapan	JX124557	WPMNH		2005	Off Yaizu-shi, Shijuoka Prof., Japan	760-800	33.00	138.40	
Paragorgia arborea	Ppacifica1016320	JX124581	USNM	1016320	2002	British Columbia, Canada	1152-11	53.70	-133.42	
Paragorgia arborea	TC16_03		WHOI	HB1302_TC016	2013	NE US Canyons; Munson Canyon; TowCam 16	540	40.54	-67.01	
Paragorgia cf dendroides	Pcfdendro98788		USNM	98788	1996	USA, Hawaii, Lanai Island, Keanapapa Point, SW of Point	1007	20.78	-157.15	
Paragorgia cf dendroides	Pcfdendro98792		USNM	98792	1996	USA, Hawaii, Lanai Island, Keanapapa Point, SW of Point	1018	20.78	-157.15	
Paragorgia cf dendroides	PcfdendroT662A30		USNM	1122228	200?	USA, California, San Juan Seamount	1237.6	32.97	-121.03	
Paragorgia cf johnsoni		KC984606								
Paragorgia cf regalis	Pcfregalis1072337		USNM	1072337	2003	USA, Hawaii, Pioneer Bank	1211	25.81	-173.50	
Paragorgia coralloides	06390		WHOI	REH112-5	2005	New England Seamounts; Rehobot Seamount; H13	1821	37.46	-59.95	
Paragorgia coralloides	12016		WHOI	leftover sieving	2003	New England Seamounts; Manning Seamount, station 4; AD3	2000	38.23	-60.46	
Paragorgia coralloides	Pcfcorallo98785	JX128350	USNM	98785	1995	East Pacific Rise, off Mexico	1950	12.73	-102.60	
Paragorgia dendroides	Pdendroid1072362		USNM	1072362	2003	USA, Hawaii, Necker Island, Seamount East of Island	1536	23.30	-163.70	
Paragorgia johnsoni	20136	KC984607	TU-WH	0909-Oct1	2009	Gulf of Mexico; MC751; J2-464	438	28.19	-89.80	
Paragorgia johnsoni	Paragosp100898		USNM	100898	2000	USA, North Atlantic Ocean, South of Georges Bank		39.86	-67.42	Sanchez 2005
Paragorgia johnsoni	Pjohnsoni73767	JX128348	USNM	73767	1984	Little Bahama Bank, Bahamas	608	27.10	-79.70	Sanchez 2005
Paragorgia johnsoni		KC788262								
Paragorgia kaupeka	72152		NIWA	72152	2011	Kermadec Ridge; Clark Seamount, chimney field, north cone;	877	-36.45	177.84	
Paragorgia kaupeka	82260		NIWA	82260	2012	Kermadec Ridge; Site SM3a, summit of Clark Seamount; TAN	850	-36.45	177.84	
Paragorgia kaupeka	82342		NIWA	82342	2012	Kermadec Ridge; Site SM3a, Clark Seamount; TAN1206/40	1100	-36.45	177.84	
Paragorgia kaupeka	84804		NIWA	84804	2005	Kermadec Ridge; Clark Seamount; KOK0506/12	870	-36.45	177.84	
Paragorgia kaupeka	Pkaupeka3320	GQ293313	NIWA	3320	1989	New Zealand, X152	820	-36.16	176.81	type
Paragorgia maunga	64980		NIWA	64980	2010	Kermadec Ridge; Silent II seamount; TAN1007/120	772	-35.17	178.89	
Paragorgia maunga	Pmaunga28393		NIWA	28393	1999	New Zealand, Z9779	1121	-34.12	174.90	
Paragorgia maunga	Pmaunga3322		NIWA	3322	2002	New Zealand, Wanganella Bank, TRIP, Z10989	1082	-33.89	167.94	type
Paragorgia maunga	Pmaunga3323		NIWA	3323	1999	New Zealand, Z9779	1121	-34.12	174.90	type
Paragorgia regalis	ParagospT629A6		USNM	1122239	200?	USA, California, Rodriguez Seamount	1843.8	33.95	-121.14	
Paragorgia regalis	Pcfdendro98789		USNM	98789	1996	USA, Hawaii, Lanai Island, Keanapapa Point, SW of Point	1018	20.78	-157.15	
Paragorgia regalis	PdendroidT630A5		USNM	1122192	200?	USA, California, Rodriguez Seamount	1031.8	34.00	-121.10	
Paragorgia regalis	Pregalis1027063		USNM	1027063	2003	USA, California, Rodriguez Seamount, West of San Miguel Pa	1840	33.95	-121.14	
Paragorgia regalis	Pregalis1072338		USNM	1072338	2003	USA, Hawaii, Laysan Island, SE of Island	1136	25.67	-171.41	
Paragorgia regalis	Pregalis1072339		USNM	1072339	2003	USA, Hawaii, Pioneer Bank	1743.7	25.57	-173.51	
Paragorgia regalis	Pregalis1072340		USNM	1072340	2004	USA, Hawaii, Pioneer Bank	1744.7	25.57	-173.51	
Paragorgia regalis		GQ293307	USNM	1014743	2003	Cross Seamount: Hawaii: USA	452	19.74	-158.30	Sanchez 2005

Paragorgia regalis		JQ241244							
Paragorgia sp	Paragosp1007354		USNM	1007354	2001	Canada, British Columbia, Vancouver Island	1168	48.44	-126.38
Paragorgia sp	Paragosp1011093		USNM	1011093	2002	USA, Bering Sea, Forrester Island	601-800	55.40	-134.83
Paragorgia sp	Paragosp1011094		USNM	1011094	2002	USA, Bering Sea, East of Kodiak Island	601-836	58.22	-148.70
Paragorgia sp	Paragosp1011095		USNM	1011095	2002	USA, Bering Sea, Alaska, Alexander Archipielago, Baranof Isl	601-715	57.19	-136.24
Paragorgia sp	Paragosp1027079		USNM	1027079		USA, California, Rodriguez Seamount, West of San Miguel Pa	1030	34.05	-121.10
Paragorgia sp	Paragosp1071233		USNM	1071233	2004	USA, Hawaii, Cross Seamount	410	18.73	-158.26
Paragorgia sp	Paragosp1071440		USNM	1071440	2004	USA, Hawaii, Cross Seamount	427.51	18.73	-158.26
Paragorgia sp	Paragosp1071441		USNM	1071441	2004	USA, Hawaii, Cross Seamount	427.51	18.73	-158.26
Paragorgia sp	Paragosp1072336		USNM	1072336	2003	USA, Hawaii, Raita Bank	573	25.63	-169.32
Paragorgia sp	Paragosp1075392		USNM	1075392	2004	USA, Gulf of Alaska, Alaska, Giacomini Seamount	733	56.42	-146.37
Paragorgia sp	Paragosp1075750		USNM	1075750	2004	USA, Gulf of Alaska, Alaska, Welker Seamount	718	55.05	-140.31
Paragorgia sp	Paragosp1075751		USNM	1075751	2004	USA, Gulf of Alaska, Alaska, Welker Seamount	782	55.05	-140.31
Paragorgia sp	Paragosp1075752		USNM	1075752	2004	USA, Gulf of Alaska, Alaska, Welker Seamount	1119	55.06	-140.41
Paragorgia sp	Paragosp1075755		USNM	1075755	2004	USA, Gulf of Alaska, Alaska, Welker Seamount	1050	55.06	-140.41
Paragorgia sp	Paragosp1075756		USNM	1075756	2004	USA, Gulf of Alaska, Alaska, Welker Seamount	1050	55.06	-140.41
Paragorgia sp	Paragosp1075757		USNM	1075757	2004	USA, Gulf of Alaska, Alaska, Pratt Seamount	1093	56.30	-142.47
Paragorgia sp	Paragosp1075759		USNM	1075759	2004	USA, Gulf of Alaska, Alaska, Pratt Seamount	1069	56.17	-142.70
Paragorgia sp	Paragosp1075764		USNM	1075764	2004	USA, Gulf of Alaska, Alaska, Pratt Seamount	920	56.17	-142.70
Paragorgia sp	Paragosp1075769		USNM	1075769	2004	USA, Gulf of Alaska, Alaska, Giacomini Seamount	730	56.42	-146.37
Paragorgia sp	Paragosp1082595		USNM	1082595	2002	USA, Gulf of Alaska, Alaska, Murray Seamount	855	53.89	-148.53
Paragorgia sp	Paragosp1082600		USNM	1082600	2002	USA, Gulf of Alaska, Alaska, Murray Seamount	1376	53.99	-148.50
Paragorgia sp	Paragosp1082644		USNM	1082644	2002	USA, Washington, Warwick Seamount	768	48.05	-132.74
Paragorgia sp	Paragosp1200601		Stone-Sa	1200601	2006	Gulf of Alaska	741	53.98	-162.85
Paragorgia sp	Paragosp121	JX128347	USNM	1122305	2004	British Columbia, Canada		51.20	-130.14
Paragorgia sp	Paragosp1310601		Stone-Sa	1310601	2006	Gulf of Alaska	549	53.73	-163.98
Paragorgia sp	Paragosp159		USNM	1122302	2003	Canada, British Columbia			
Paragorgia sp	Paragosp165		USNM	1122303	2003	Canada, British Columbia, Brooks Penninsula			
Paragorgia sp	Paragosp27294		YPM	27294	2000	Bear Seamount		39.92	-67.44
Paragorgia sp	Paragosp28905		YPM	28905	2003	USA, Atlantic Ocean, Muir Seamount	1713	33.85	-62.66
Paragorgia sp	Paragosp28911		YPM	28911	2003	USA, Atlantic Ocean, Manning Seamount	1550	38.22	-60.46
Paragorgia sp	Paragosp28921		YPM	28921	2003	USA, Atlantic Ocean, Manning Seamount	1579	38.21	-60.53
Paragorgia sp	Paragosp28946		YPM	28946	2003	USA, Atlantic Ocean, Manning Seamount	1597	38.20	-60.53
Paragorgia sp	Paragosp291		USNM	1122306	2004	Canada, British Columbia			
Paragorgia sp	Paragosp34786		YPM	34786	2004	Atlantic Ocean, Manning Seamount, on summit	1336	38.22	-60.51
Paragorgia sp	Paragosp35381		YPM	35381	2004	USA, Manning Seamount, on summit	1337	38.22	-60.51
Paragorgia sp	Paragosp36781		YPM	36781	2004	Bear Seamount, South rim of table top peak	1428-16	39.87	-67.36
Paragorgia sp	Paragosp36910C		YPM	36910C	2004	Bear Seamount, South rim of table top peak	1428-16	39.87	-67.36
Paragorgia sp	Paragosp37148		YPM	37148	2003	USA, Manning Seamount	1483	38.26	-60.55
Paragorgia sp	Paragosp38636		YPM	38636	2004	Bear Seamount, South rim of table top peak	1428-16	39.87	-67.36
Paragorgia sp	Paragosp41849		NIWA	41849	2007	New Zealand, TRIP2494/97	1155.0	-47.0100	175.5800000000
Paragorgia sp	Paragosp44607		NIWA	44607	2007	New Zealand, TRIP2506/24	858.0	-50.0516	174.7150000000
Paragorgia sp	Paragosp54830		USNM	54830	1974	USA, Florida, Straits of Florida, Off Delray Beach	743-761	26.38	-79.60
Paragorgia sp	Paragosp56615		USNM	56615	1974	USA, Florida, Straits of Florida, Off Mile Beach	770-660	25.71	-79.79
Paragorgia sp	Paragosp56690		USNM	56690		USA, Hawaii, Oahu Island, Makapuu Point	366		

Paragorgia sp	Paragospn32830		NIWA	32830	2005	New Zealand, Tangaroa Seamount, P629-4B, KOK0507/4	790	-36.3284	178.0359955	
Paragorgia sp	ParagospT665A3		USNM	1122225	200?	USA, California, San Juan Seamount	706.6	33.11	-120.96	
Paragorgia sp	Parborea28423		NIWA	28423	1998	New Zealand, 1152/48, Z9275	660	-34.18	162.65	
Paragorgia sp	Parborea28717		YPM	28717	2003	Manning Seamount	1372	38.22	-61.51	
Paragorgia sp	PcfaoteaT668A3		USNM	1122226	200?	USA, California, Little Joe Seamount	2397.8	31.89	-120.05	
Paragorgia sp	PcfaoteaT669A15		USNM	1122227	200?	USA, California, San Marcos Seamount	2061.7	32.64	-121.51	
Paragorgia sp	PcfstepheT665A4		USNM	1122235	200?	USA, California, San Juan Seamount	720.5	33.11	-120.96	
Paragorgia sp	PcfwahineT627A3		USNM	1122238	200?	USA, California, Pioneer Seamount	1771.5	37.40	-123.44	
Paragorgia sp	PcfwahineT630A6		USNM	1122234	200?	USA, California, Rodriguez Seamount	1031.9	34.00	-121.10	
Paragorgia sp	PcfyutliT663A16J4		USNM	1122231	200?	USA, California, Northeast Bank	546.6	32.32	-119.61	
Paragorgia sp		KC788261								
Paragorgia stephencairnsi	2344		RBCM	010-00234-004	2004	British Columbia; VE13978	1194	53.37	-133.31	
Paragorgia stephencairnsi	101010		USNM	1157074	2008	California; Piggy Bank, southern California; DW-026-02	283	33.92	-119.47	
Paragorgia stephencairnsi	1007316		USNM	1007316	2001	British Columbia; Vancouver Island	1168	48.44	-126.38	
Paragorgia stephencairnsi	1124300		USNM	1124300	2006	British Columbia; Vancouver Island, Ohiat Island; OC 06/952	188	48.83	-125.13	
Paragorgia stephencairnsi	Agam		WHOI	Agam	2012	British Columbia; Agamemnon Channel	32	49.72	-124.05	
Paragorgia stephencairnsi	C02		WHOI	C02	2013	British Columbia; Agamemnon Channel; Dive02	41	49.74	-124.03	
Paragorgia stephencairnsi	C03		WHOI	C03	2013	British Columbia; Agamemnon Channel; Dive02	41	49.74	-124.03	
Paragorgia stephencairnsi	C04		WHOI	C04	2013	British Columbia; Agamemnon Channel; Dive02	41	49.74	-124.03	
Paragorgia stephencairnsi	C05		WHOI	C05	2013	British Columbia; Agamemnon Channel; Dive02	41	49.74	-124.03	
Paragorgia stephencairnsi	C100		WHOI	C100	2013	British Columbia; Vancouver Island, Tahsis Inlet; Dive07	40	49.86	-126.67	
Paragorgia stephencairnsi	C101		WHOI	C101	2013	British Columbia; Vancouver Island, Tahsis Inlet; Dive07	40	49.86	-126.67	
Paragorgia stephencairnsi	C102		WHOI	C102	2013	British Columbia; Vancouver Island, Tahsis Inlet; Dive07	40	49.86	-126.67	
Paragorgia stephencairnsi	C104		WHOI	C104	2013	British Columbia; Vancouver Island, Tahsis Inlet; Dive07	40	49.86	-126.67	
Paragorgia stephencairnsi	FOC25		DFO	25	2012	British Columbia; W of Graham Island; 2012-65	204	53.31	-133.03	
Paragorgia stephencairnsi	FOC26		DFO	26	2012	British Columbia; W of Graham Island; 2012-65	221	53.30	-133.04	
Paragorgia stephencairnsi	FOC30		DFO	30	2012	British Columbia; W of Graham Island; 2012-65	318	53.48	-133.07	
Paragorgia stephencairnsi	FOC5		DFO	5	2009	British Columbia; E of Graham Island; 2009-47	201	52.13	-128.90	
Paragorgia stephencairnsi	Paragosp1075741		USNM	1075741	2004	USA, Gulf of Alaska, Alaska, Dickins Seamount	751	54.55	-136.84	
Paragorgia stephencairnsi	Paragosp1092785		USNM	1092785	2005	USA, North Pacific Ocean, Alexander Archipielago, Baranof I	171	56.19	-135.10	
Paragorgia stephencairnsi	Paragosp200601		Stone-Sa	200601106B01	2006	Gulf of Alaska	479	55.40	-134.83	
Paragorgia stephencairnsi	Paragosp77		USNM	1122304	2004	Canada, British Columbia, Brooks Penninsula		49.94	-128.06	
Paragorgia stephencairnsi	Parborea41106		USNM	1123931	2004	USA, Bering Sea, GOA	417	55.37	-134.78	
Paragorgia stephencairnsi	Parborea411081		USNM	1123930	2004	USA, Bering Sea, GOA	427	54.47	-133.97	
Paragorgia stephencairnsi	ParboreaNew100		USNM	1124301	2006	Canada, British Columbia, Vancouver Island, Barkley Sound	270.22	48.26	-125.01	
Paragorgia stephencairnsi	ParboreaNew545		USNM	1124298	2006	Canada, British Columbia, Vancouver Island, Barkley Sound	309.5	48.15	-125.07	
Paragorgia stephencairnsi			CAS	190438	2010	California, Farallon Escarpement	424	37.74	-123.19	
Paragorgia wahine	Pwahine3326	GQ293314	NIWA	3326	2001	New Zealand, Diabolical seamount, TAN0104/113	900	-42.79	179.99	type
Paragorgia whero	Pwhero3436		NIWA	3436	1998	New Zealand, TRIP1171/12, Z9583	935	-48.03	166.10	type
Paragorgia yutlinux	Pyutlinux1073480	GQ293315	USNM	1073480	2003	Off Vancouver Isl., British Columbia, Canada	846–861	50.23	-128.58	type
Paraminabea aldersladei		JX203767								
Paraminabea aldersladei		KF915662								
Paraminabea aldersladei		KF915665								
Sibogagorgia cauliflora	2036	KC984605	TU-WHC	2036-Oct1	2009	Gulf of Mexico; DC583; J2-454	2440	28.39	-87.39	
Sibogagorgia cauliflora	1122230	GQ293310	USNM	1122230	2006	California; Davidson seamount; dive 945	2502	35.83	-122.61	type

Sibogagorgia cauliflora	Sibogasp1T947A9	GQ293317	USNM	1122229	2006	Davidson Seamount, California, USA	3042.4	35.63	-122.83	
Sibogagorgia cauliflora		GQ293308	USNM	1081143	2004	Derickson Seamount: Alaska: USA	2766	52.98	-161.25	type
Sibogagorgia cauliflora		GQ293309	USNM	54831	1968	Straits of Florida: Havana: Cuba	1638-17	23.55	-82.78	type
Sibogagorgia dennisgordoni		GQ293316	NIWA	3329	1998	New Zealand, 1124/70, Z9228	820	-36.69	176.46	type
Sphaerasclera flammicerebra		JX203765								

Acronyms as follows: National Museum of Natural History, Smithsonian Institution, USA (USNM); The National Institute of Water and Atmospheric Research, New Zealand (NIWA); Museum of Comparative Zoology, Harvard University, USA (MCZ); Muséum National d'Histoire Naturelle, Paris, France (MNHN); Senckenberg Research Institute And Natural History Museum Frankfurt, Germany (SMF); Uppsala University Evolutionsmuseet, Sweden (UUZM); Wakayama Prefectural Museum of Natural History, Japan (WPMNH); Yale Peabody Museum of Natural History, USA (YPM), Department of Fisheries and Oceans Canada (DFO), Woods Hole Oceanographic Institution (WHOI), Temple University (TU), Royal British Columbia Museum (RBCM), California Academy of Scieneces (CAS)

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