



NIWA
Taihoro Nukurangi

INT2023-05: High-resolution estimation of species diversity for a protected coral family commonly occurring as trawl bycatch

Final Report

*Prepared for Conservation Services Programme
Department of Conservation*

June 2024



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NIWA CLIENT REPORT No: 2024188WN
Report date: June 2024
NIWA Project: DOC24302

Revision	Description	Date
Version 0.1	Draft in preparation/in review	24 June 2024
Version 1.0	Final version sent to client	26 June 2024
Version 1.1	Revised version sent to client	10 September 2024

Quality Assurance Statement		
	Reviewed by:	Felix Vaux
	Formatting checked by:	Jess Moffat
	Approved for release by:	Judy Sutherland

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Contents

Executive summary	4
1 Background	5
2 Methods.....	7
3 Results	8
4 Discussion	16
5 Recommendations.....	19
6 Acknowledgements	20
7 References.....	21
Appendix A Sample Collection Metadata.....	24
Appendix B Target-bait enrichment DNA sequencing summary.....	29

Figures

Figure 1:	Location of Paramuriceidae specimens housed in the NIC.	6
Figure 2:	Map of locations from which sequenced paramuriceid samples were obtained.	8
Figure 3:	Phylogenetic outgroup analysis using 50%-occupancy alignment of UCE dataset.	10
Figure 4:	Phylogenetic outgroup analysis using 75%-occupancy alignment of UCE dataset.	11
Figure 5:	Phylogenetic analysis of Paramuriceidae using 50%-occupancy alignment of UCE dataset.	13
Figure 6:	Phylogenetic analysis of Paramuriceidae using 75%-occupancy alignment of UCE dataset.	14
Figure 7:	Cladogram of 50%-occupancy UCE alignment showing specimen origin details.	15
Figure 8:	Representatives of gorgonian specimens included for sequencing.	17

Executive summary

Theme 1 of the Conservation Services Programme's Medium-Term Research Plan (MTRP) for protected corals highlights the requirements for species-level data in determinations of susceptibility to commercial fishing impacts. However, most protected gorgonian corals (Cnidaria: Anthozoa: Octocorallia) known to occur in the territorial seas of Aotearoa New Zealand (NZ) have not been documented at the species-level and many represent undescribed taxa that lack formal scientific names. The ubiquitous sea-fans of the family Paramuriceidae are recognised as one of the most diverse groups of gorgonians within the EEZ but there are currently only two records of described species from this group in NZ, despite being a regular component of bottom trawl bycatch in commercial fishing operations.

This research addresses Biodiversity and Distribution aspects of Theme 1 of the coral MTRP, using genetic delimitation of species that are distributed across spatially explicit regions of relevance to commercial fishing activities. Genomic DNA sequencing using target-bait enrichment was applied to available specimens of the Paramuriceidae held in the NIWA Invertebrate Collection, which were obtained from both bycatch and non-bycatch sources on the Chatham Rise (Fisheries Management Area (FMA4) and Campbell and Bounty plateaus (FMA6). DNA sequencing was used to objectively determine the number of distinct taxa present in each region and examine fishing-related impacts on protected coral diversity. Fifty-one specimens were successfully sequenced, and 32 distinct taxa were discriminated, including a distinct genetic lineage that may represent a new family of protected gorgonians. The two FMA regions shared representatives of most major lineages, but one lineage was confined to the Chatham Rise and the members of the tentative new family were confined to the Campbell and Bounty plateaus, where they were obtained primarily as bycatch. Overall, all major genetic lineages were represented among sampled bycatch, with the exception of the lineage found only on the Chatham Rise. In combination with previous genetic characterisation efforts, the current project brings the total number of protected octocorals represented in trawl bycatch that have been genetically detected or confirmed to 43 taxa (genera and species) distributed amongst at least seven families (five described, one in question, one undescribed).

The continued use of genomic approaches in the characterisation of coral bycatch is warranted by its ability to produce large amounts of informative data that can resolve relationships at multiple taxonomic levels, and its resilience to highly degraded sample sources that include dried and aged archival tissues. While the current and previous studies support target-bait enrichment as an effective means of characterising the identity and relationships of bycatch specimens, recent advances and cost reductions in whole genome 'skimming' make it an attractive method for generating comparable genomic and genetic data for protected corals, which can be implemented within NZ, without need for specialised overseas providers.

1 Background

Corals belonging to the Class Octocorallia assume a wide range of morphological forms, including sea-fans and sea-whips, soft corals, sea pens, and encrusting colonies. Most of the erect and tree-like families of octocorals (commonly called 'gorgonians') were originally classified in the Order Gorgonacea, with soft corals and other fleshy forms contained in the Alcyonacea (Hickson 1930). The Gorgonacea included bubblegum corals (F. Paragorgiidae), bamboo corals (Isididae), golden corals (Chrysogorgiidae), and primnoid (Primnoidae), plexaurid (Plexauridae) and acanthogorgiid (Acanthogorgiidae) sea-fans. The Order was the only octocoral group protected under the Schedule 7A addition to the Wildlife Act of New Zealand in 2010 but by then it had been recognised as an invalid taxonomic assemblage by the scientific community (Berntson et al. 2001; Daly et al. 2007), leaving its constituent families without a valid higher-level classification. Recently, McFadden et al. (2022) revised the higher classification of the Octocorallia, with families of the former Gorgonacea divided into the O. Malacalcyonea and O. Scleralcyonacea. Several families were also revised, including the grouping of the Plexauridae and Acanthogorgiidae into a single family – the Paramuriceidae. However, few families have been the subject of revision of genera and species; only the bubblegum corals (*Paragorgia* and *Sibogorgia* – now placed in the family Coralliidae) and Primnoidae have received species-level reviews (Sanchez 2005; Cairns 2012, 2016, 2021), with the bamboo corals (now placed in the Keratoisididae and Mopseidae) still undergoing genus-level revisions (Heestand-Saucier et al. 2021; Watling & France 2021).

Theme 1 of the Conservation Services Programme's (CSP) Medium-Term Research Plan (MTRP) for protected corals highlights the requirements for species-level data in determinations of susceptibility to commercial fishing impacts. However, most protected gorgonian corals (Cnidaria: Anthozoa: Octocorallia) in Aotearoa New Zealand (NZ) have not been documented at the species-level and many represent undescribed species in need of formal taxonomic descriptions (Tracey & Hjørvarsdóttir 2019). In particular, the ubiquitous sea-fans of the family Paramuriceidae consist of at least 13 genera in NZ and is likely one of the most diverse groups of gorgonians within its territorial seas (Macpherson et al. 2023). However, a lack of applicable species names has precluded inclusion of any members of this family in the NZ Threat Classification System (Funnell et al. 2023), and there are currently only two NZ records of described species from this group (Macpherson et al. 2023). The remaining records constitute undescribed or unidentified species.

Bottom-trawling fisheries (including those targeting orange roughy, oreos, hoki, hake and scampi) produce the most gorgonian coral bycatch amongst New Zealand deep-water fisheries (Tracey et al. 2011; Meyer 2023). A previous DOC CSP project (INT2019-05: Bilewitch & Tracey 2020) included 19 paramuriceid bycatch samples in an EEZ-wide examination, finding 12 'operational taxonomic units' (OTUs) divided across 10 genera, but the applied genetic barcoding methods were unable to confidently recognise or delineate species. As such, there is currently no knowledge of the breadth of species-level diversity that is being impacted by commercial fishing activity for this frequently encountered and commonly occurring group. This project sought to address this shortfall by producing the first high-resolution estimate of the number of Paramuriceidae species present within the Chatham Rise and Campbell and Bounty plateaus fisheries management areas (FMAs 4 and 6 respectively), since these represent regions of relevance for bottom trawling activities (Baird & Mules 2021), and both have sufficient sample numbers available within the NIWA Invertebrate Collection (NIC) (Figure 1).

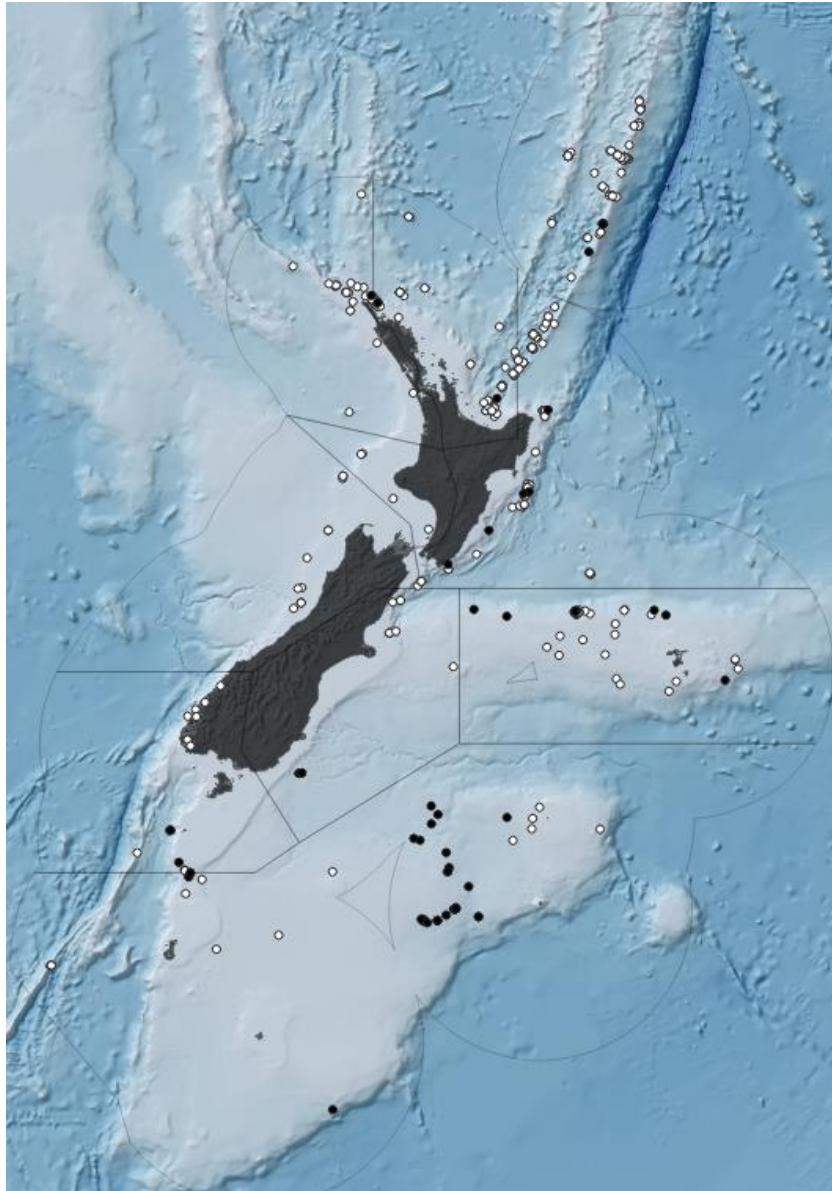


Figure 1: Location of Paramuriceidae specimens housed in the NIC. Dots indicate collection locations for bycatch (black; n=59) and non-bycatch (white; n=416) NIC specimens. Black lines denote FMA boundaries.

High-resolution genomic target-capture methods (otherwise known as ‘ultraconserved elements’ or ‘UCEs’) have been developed for octocorals overseas (Quattrini et al. 2017), where they have demonstrated an ability to resolve relationships at varying taxonomic levels – from higher-level orders and families (McFadden et al. 2022), to genera and species (Morrisey et al. 2023), and even at intra-specific (population) levels (Glon et al. 2021). The DOC project BCBC2020-26 (Bilewitch 2022) tested these methods on a subset of Chatham Rise Primnoidae and found that they resolved closely related genera and species, which could not be distinguished using older genetic barcoding methods. As such, the target-capture approach (hereafter UCEs) was used to describe the extent of species-level diversity and genetic relationships of paramuriceid gorgonian coral samples available in the NIWA Invertebrate Collection, which originated from both bycatch and research voyages in FMA4 and FMA6.

2 Methods

Paramuriceidae from FMAs 4 and 6 were selected for potential inclusion in this study by overlaying FMA boundaries onto a map of coordinates from NIC specimens identified as Paramuriceidae (or the former Plexauridae and Acanthogorgiidae). Specimens were obtained from both the dry and ethanol-preserved collections and were examined prior to sampling for genetic analysis. DNA was extracted from approximately 10mg of coral tissue using a DNeasy Blood & Tissue kit, following the manufacturer's protocol (Qiagen), except that two final elutions of 40 μ l were used. DNA extracts were quantified using Quant-iT PicoGreen (Invitrogen) and samples with a concentration of >1ng/ μ l were considered for UCE sequencing.

Extracted DNA was submitted for UCE sequencing at a commercial facility (Arbor Biosciences, USA) using the *octocoral-v2* baitset (Erickson et al. 2020). Project INT2022-03 provided support for the inclusion of an additional eight paramuriceid DNA extracts obtained during INT2019-05 plus 16 reference octocoral specimens for outgroup comparisons: two Chrysogorgiidae specimens (NIWA 88693 & 131891) and two Keratoisididae specimens (NIWA 65999 & 88879) extracted during INT2019-05 (Bilewitch & Tracey 2020), three DNA extracts of Primnoidae (NIWA 42522, 53309 & 102618) and eight extracts of a putative new family from BCBC2020-26 (Bilewitch 2022), plus a specimen of an inshore encrusting soft coral (NIWA 41313: cf. *Callipodium*). These 'outgroup' specimens represented other octocoral families and were used as references to resolve ancestral phylogenetic relationships for the paramuriceid 'ingroup' samples that were the focus of this study.

UCE sequencing data was trimmed of adapter sequences and low-quality bases using the *Illuminiprocessor* package in *phyluce* v1.7.3 (Faircloth 2016), followed by assembly of cleaned reads using *SPAdes* v3.15.4 (Bankevich et al. 2012) with the *--careful* and *--cov-cutoff=2* options enabled. Assembled contigs were matched to UCE loci and extracted using *phyluce*. UCE loci were aligned with edge-trimming using the *phyluce_align_seqcap_align* option. The resulting alignments were subset to remove UCE loci containing high levels of missing data, which can mislead or reduce the resolution of phylogenetic reconstructions. The alignment was filtered to include only loci that either contained DNA sequences for at least 50%- or 75%- of all paramuriceid taxa (50%- or 75%- 'occupancy'). A separate UCE analysis including outgroup taxa was also conducted on 50%- and 75%-occupancy alignments. Bayesian phylogenetic analyses using *ExaBayes* v1.5.1 (Aberer et al. 2014) were conducted with four chains run for 1x10⁶ generations, sampled every 1000 generations with the first 1x10⁵ generations discarded as burn-in. Chain parameters were inspected for convergence using *Tracer* (Rambaut et al. 2018) and posterior results were summarised into a consensus tree.

3 Results

A total of 92 paramuriceid specimens were initially identified among ethanol-preserved and dried collections, but further inspection determined that 15 of these were misidentified non-paramuriceid taxa (primarily *Iciligorgia* and *Anthothela*) and warranted exclusion. A further five specimens could not be located, and 10 specimens lacked any suitable tissues for genetic sampling (*ie.* they were denuded branches). Among the remaining material, 42 originated from FMA4 (including four bycatch specimens) and 20 originated from FMA6 (including 15 bycatch specimens) and all of these were sampled for DNA extraction (Figure 2).

Six samples produced $<1\text{ng}/\mu\text{l}$ of extracted DNA, a concentration usually too low for analysis, but two of these (NIWA 14435, 47780) were included nonetheless, to keep the FMA6 sample size similar to FMA4. Five specimens from FMA4 produced $<2\text{ng}/\mu\text{l}$ of DNA and were excluded since they represented morphological forms that were represented by other specimens with higher yield. A further seven FMA4 specimens were excluded since they were morphotypes represented by other specimens and logistics prevented inclusion of all successfully extracted samples. Ultimately, 46 samples were submitted for UCE sequencing, along with eight additional paramuriceids and eight reference samples from previous studies. A total of 59 samples were successfully DNA sequenced by the commercial sequencing provider, however they advised that they were unable to generate DNA sequence libraries for three of the reference specimens (two paramuriceids and one keratoisidid).

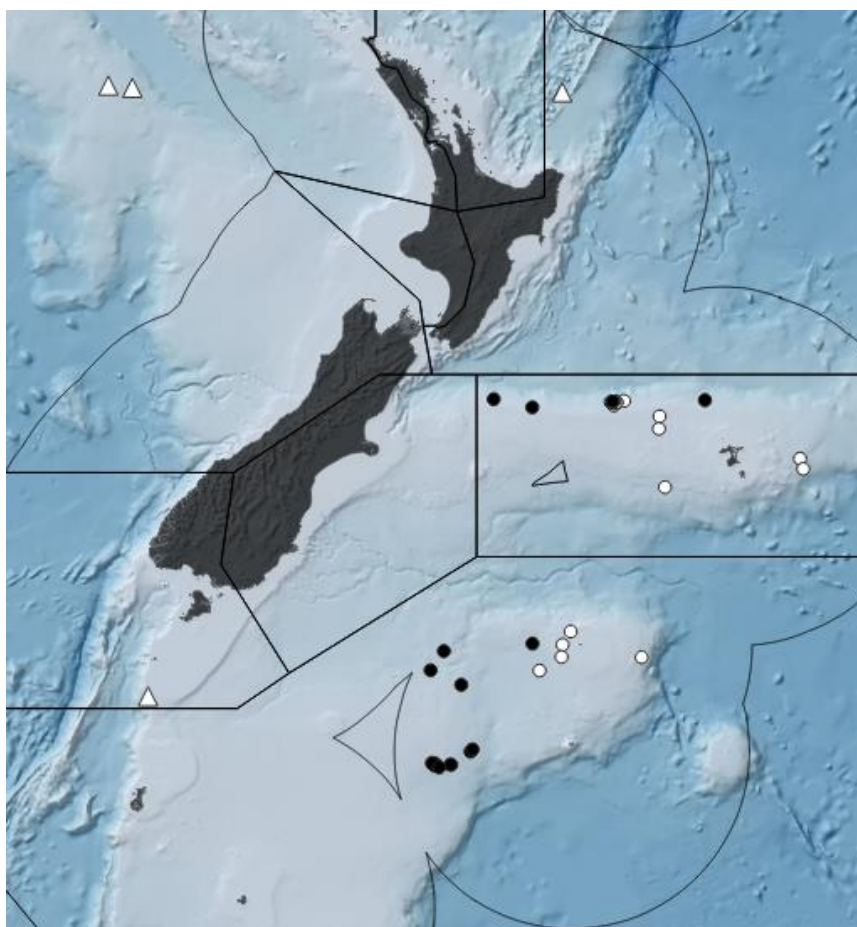


Figure 2: Map of locations from which sequenced paramuriceid samples were obtained. Each marker indicates a single sample: White circles = non-bycatch samples (n=32); Black circles = bycatch samples (n=18); Triangles = additional reference samples (n=4). Black lines indicate FMA boundaries. Non-paramuriceid outgroup samples are not shown.

After quality filtering, sequence data returned by the commercial provider contained over 650 million reads, within an average read length of 124 base pairs (bp) (SE=1.78) and 11.2 million reads per sample (SE=8.05x10⁵). Assembly of the cleaned reads resulted in an average of 50,289 contiguous sequences (contigs) per sample (SE=7377) with an average contig length of 253bp (SE=24.9). The smallest contig for paramuriceids was 47bp (NIWA 14481) and the largest was 57,432bp (NIWA 42496), with 460 contigs per sample exceeding 1,000bp in length, on average. One paramuriceid sample (NIWA 69604) and one primnoid reference sample (NIWA 42522) each had <100 total contigs and were therefore excluded from further analysis. Matching assembled contigs to UCE loci resulted in an average of 1686 contigs (UCE loci) per sample (SE=103) with an average length of 466bp (SE=22.8). UCE contig lengths ranged from 58bp (NIWA 25401) to 15,611bp (NIWA 42514), with each sample having an average of 57 contigs exceeding 1,000bp in length. Three paramuriceid samples (NIWA 14459, 42550, 69555) matched fewer than 100 UCE contigs and were also excluded from further analysis.

Alignment of the final ingroup (*ie.* Paramuriceidae-only) dataset consisting of 51 samples resulted in a total of 2874 UCE loci, totalling 1,009,311bp with 167,680bp representing informative sites. Concatenation of all loci into subsets resulted in 2348 UCE loci (845,759bp) for the 50%-occupancy dataset and 156 UCE loci (68,081bp) for the 75%-occupancy dataset. Alignment of the final outgroup+ingroup dataset consisting of 67 samples resulted in a total of 2999 UCE loci, totalling 664,837bp with 271,548bp representing informative positions. Concatenation resulted in 2229 loci (490,614bp) for the 50% dataset and 99 loci (24,312bp) for the 75% dataset.

Bayesian analysis of the four concatenated datasets (two ingroup, two ingroup+outgroup) stabilised after 100,000 generations, indicating convergence to a singular phylogenetic result. The 75% and 50% datasets for the outgroup analysis differed in their placement of the single included soft coral specimen (*Callipodium*), placing it as basal to the Primnoidae, Keratoisididae and Chrysogorgiidae in the 50% dataset, and as a sister group to a basal lineage of paramuriceids (Clade A) in the 75% dataset (Figures 3 & 4, respectively). The relationships of the primary lineages of paramuriceids were otherwise identical between both outgroup analyses and consistently identified a group of 10 specimens as a basal sister group (Clade A) to all other paramuriceids. The genetic distance (*ie.* branch lengths) observed between this lineage and all other paramuriceids was similar to – or exceeded – that seen between different families in the outgroup (*eg.* the divergence between the Primnoidae and the Chrysogorgiidae was lesser).

Among the Paramuriceidae (*ie.* the ingroup), phylogenetic analysis produced trees with identical relationships for the major (ancestral) lineages (Clades A-D), but the 50% and 75% datasets differed in their resolution at the terminal tips of each tree (Figures 5 & 6). The 50% tree discriminated more taxa with high confidence than the 75% tree, particularly among an assemblage of 16 samples of *Acanthogorgia* where the 50% tree resolved 12 taxa with high confidence (99-100% posterior probability: pp) while the 75% tree resolved only three taxa with 100% pp. Overall, the 50% dataset discriminated 33 taxa at 97-100% pp while the 75% dataset supported 16 taxa at 100% pp. The 50% tree topology was favoured as a hypothesis to explain the relationships among paramuriceids over the 75% topology since the results of the two trees were not incongruent and the latter contained less than 10% of the amount of sequence data as the former – also a likely explanation for the lower resolution of the 75% tree. However, both ingroup trees resolved and supported a basal sister lineage of 10 unidentified 'Plexauridae' samples. The genetic distance between this lineage (Clade A) and all other paramuriceids (Clade '*') exceeded that seen between different families included in the outgroup analyses (Figs. 3 & 4) and was interpreted as support for a separate family of paramuriceid-like gorgonians (see Discussion).

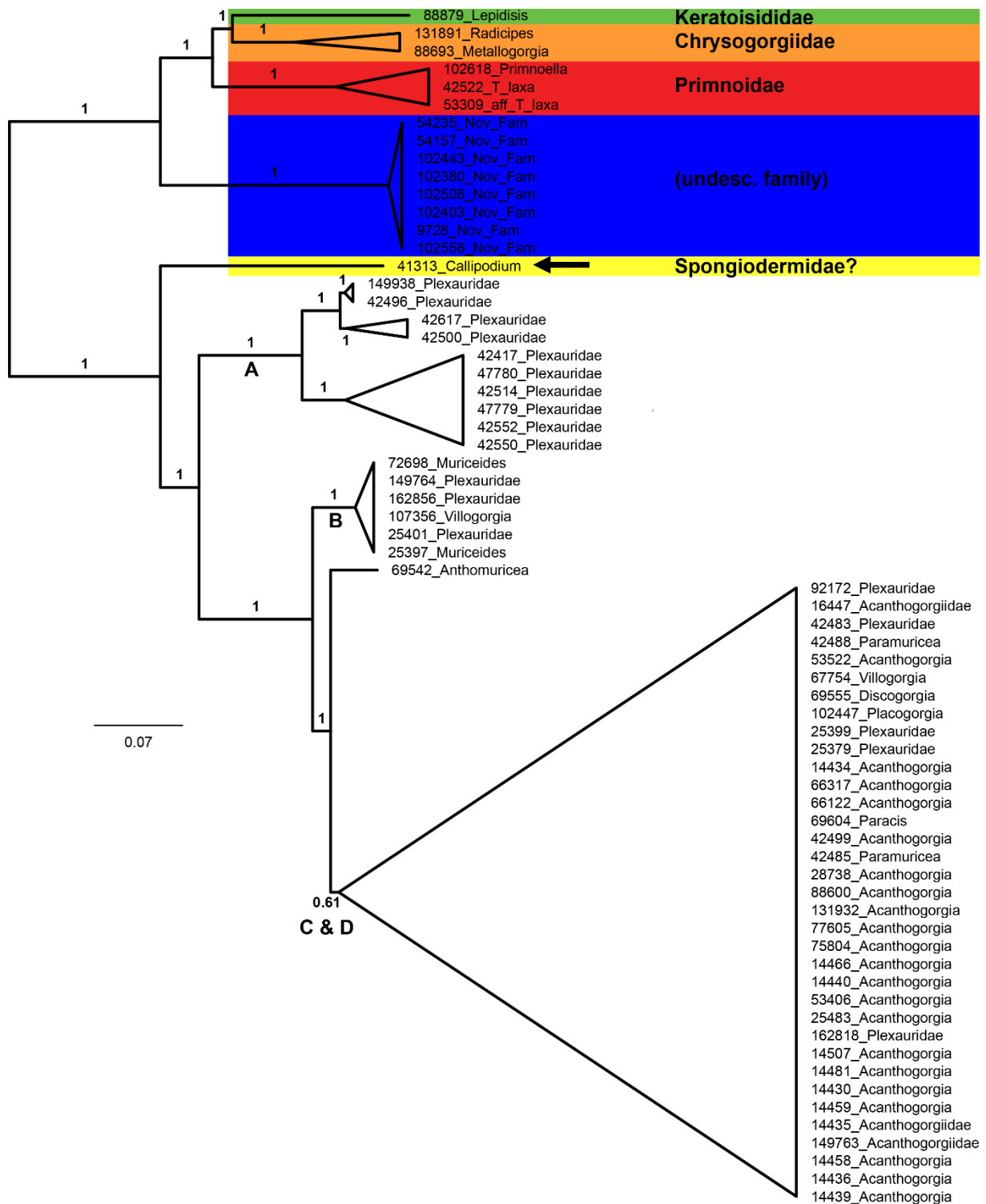


Figure 3: Phylogenetic outgroup analysis using 50%-occupancy alignment of UCE dataset. The families of outgroup specimens are indicated in coloured blocks. Major ingroup (Paramuriceidae) lineages are indicated with letters (A-D), for reference in the text. Note that relationships among Paramuriceidae are only shown for major lineages. Detail of ingroup (Paramuriceidae) genetic variation is shown in Figure 5. Branch numbers indicate posterior probability support values (expressed as a proportion) for the corresponding lineages. Branch tip labels indicate the NIC catalogue number and corresponding identification for included specimens. The position of outgroup sample 41313 differed between 50% and 75% trees (black arrow; compare to Fig. 4).

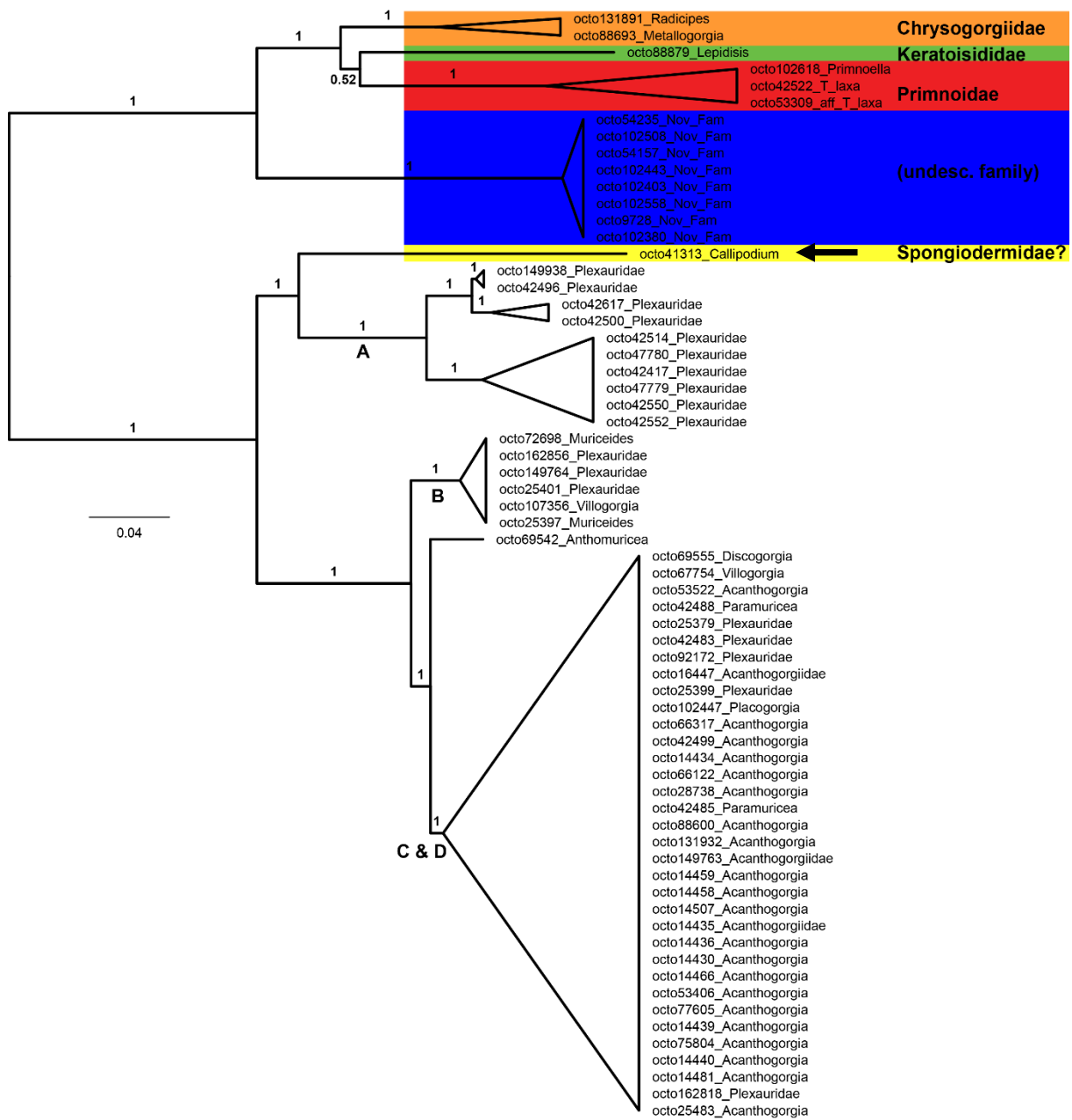


Figure 4: Phylogenetic outgroup analysis using 75%-occupancy alignment of UCE dataset. The families of outgroup specimens are indicated in coloured blocks. Major ingroup Paramuriceidae lineages are indicated with letters (A-D), for reference in the text. Note that relationships among ingroup Paramuriceidae are only shown for major lineages. Detail of ingroup genetic variation is shown in Figure 6. Branch numbers indicate posterior probability support values (expressed as a proportion) for the corresponding lineages. Branch tip labels indicate the NIC catalogue number and original identification for included specimens. The position of outgroup sample 41313 differed between 50% and 75% trees (black arrow; compare to Fig. 3).

Analyses of patterns of regional and bycatch-vs.-non-bycatch diversity were conducted using a cladogram representation of the 50%-occupancy tree (Figure 7). The unidentified family of gorgonians (Clade A in Figure 7) was further subdivided into at least four lineages – all of which were sampled exclusively from FMA6 and all except one originated as bottom-trawl bycatch. The single non-bycatch sample of this lineage (NIWA 149938) was collected in 1979 and the most recent bycatch sample was collected by observers in 2008, indicating this group has been present in the Bounty/Campbell Rise region for at least 29 years. Another lineage of samples with mixed identifications (Clade B) originated solely from more northern regions, with five samples originating from the Graveyard Seamount Complex of the Chatham Rise and a sixth from the southern end of the Kermadec Ridge in FMA2; none of which originated as trawl bycatch but all of which occurred at trawlable depths (757-1076m). The sole included specimen of *Anthomuricea* (NIWA 69542) originated as a trawl bycatch sample from the northwestern edge of the Chatham Rise and represented a distinct lineage without other close relatives among the sequence specimens. Clade C consisted of a large and genetically diverse assemblage of *Acanthogorgia* specimens originating as both bycatch and non-bycatch samples, from a broad geographical range (FMAs 4, 5, 6 and the SPRFMO region to the west of FMA 9 – see Figure 2). This clade was subdivided into a minimum of 13 taxa (see also Fig. 5) and contained 24 of the 57 ingroup samples (including reference material) sequenced in this study. The fifth major lineage (Clade D) among the ingroup samples contained a diverse array of samples primarily originating from the Chatham Rise, except for one sample (NIWA 42483) from the eastern end of the Campbell Plateau and a reference specimen (NIWA 69555) from outside the EEZ on the West Norfolk Ridge. At least seven taxa were represented within this clade and four of these contained bycatch samples.

Although recently collected and ethanol-preserved specimens are often required for traditional barcoding analyses requiring PCR amplification of marker regions, target-bait enrichment produced high-quality DNA sequence data for a variety of sample ages and preservation methods. UCE sequencing was successful in producing genome-scale data from seven ethanol-preserved historical specimens that were over 40 years old, including one collected in 1963 (NIWA 77605). The results also indicate that desiccated specimens are amenable to UCE sequencing, with 13 specimens producing viable sequence data from dried tissue samples collected in 2007 and 2008. For Clade A, dried specimens represented nearly the entirety of available and sequenced material (8/10 specimens).

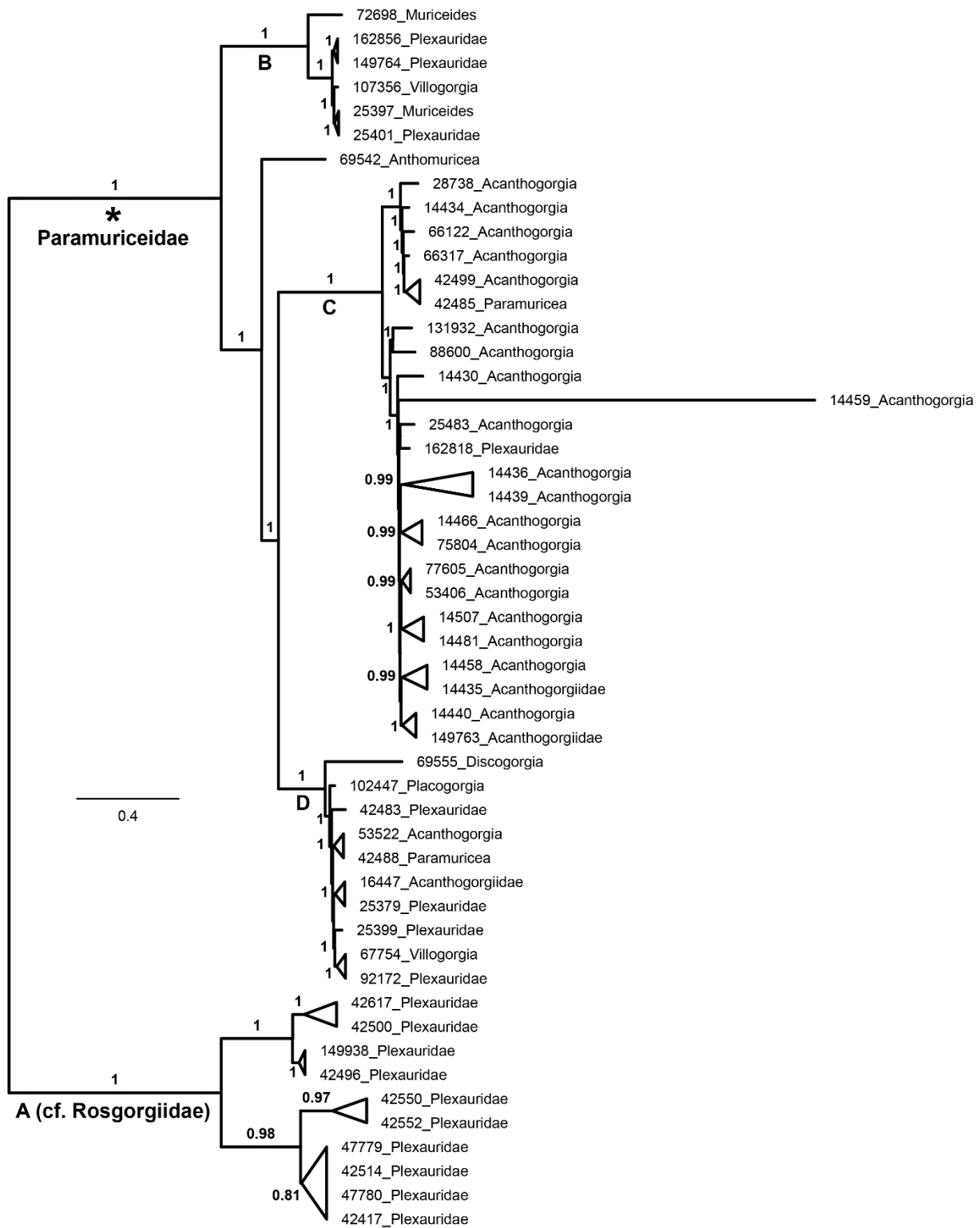


Figure 5: Phylogenetic analysis of Paramuriceidae using 50%-occupancy alignment of UCE dataset. Major lineages are labelled (A-D, *) for reference in the text; clade A was originally considered part of the paramuriceid ingroup but was subsequently determined to be part of a separate family (cf. Rosgorgiidae – see Discussion). The asterisk indicates the clade corresponding to actual Paramuriceidae. Branch numbers indicate posterior probability (expressed as a proportion) support values for the corresponding lineages. Branch tip labels indicate the NIC catalogue number and corresponding identification for included specimens.

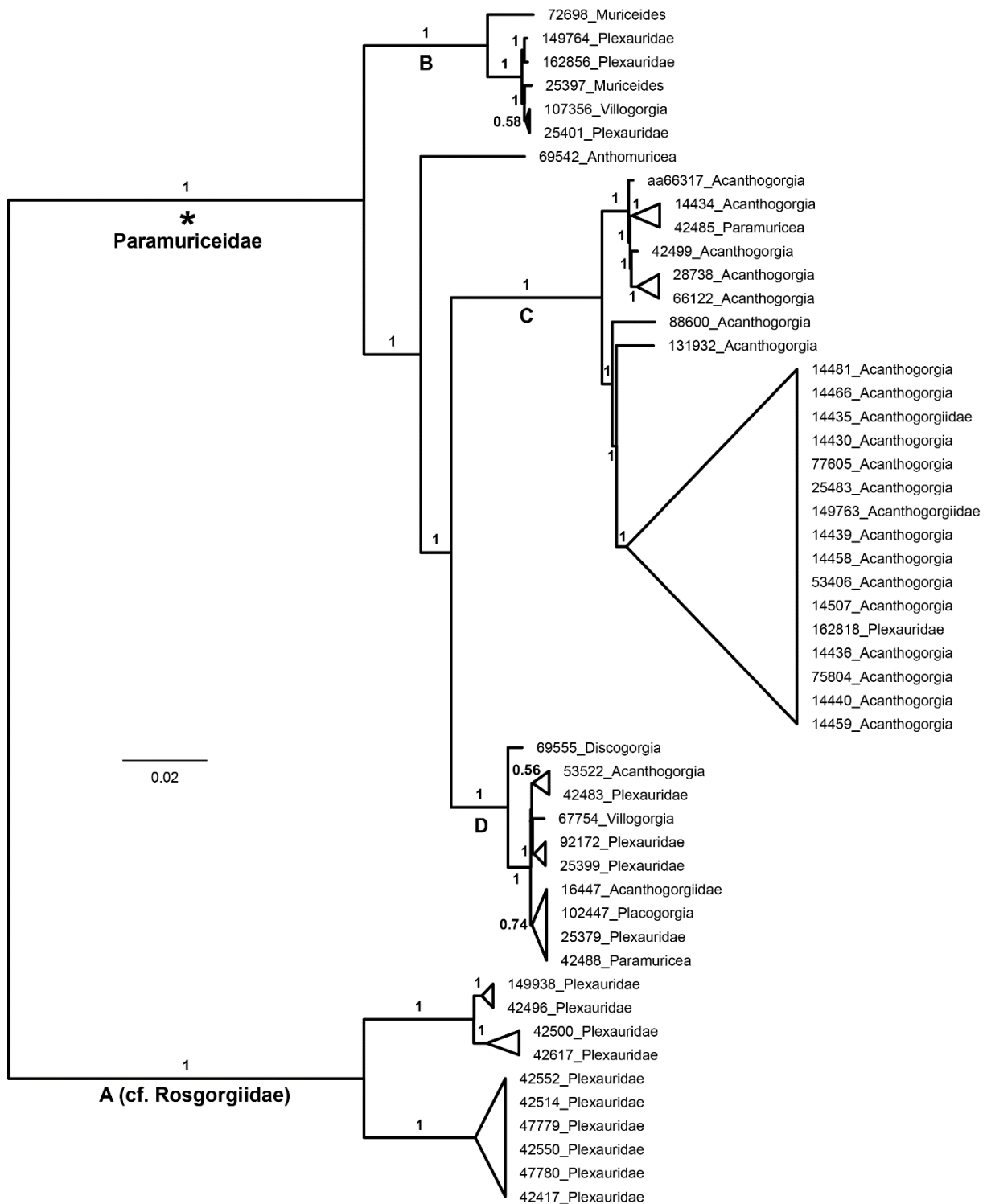


Figure 6: Phylogenetic analysis of Paramuriceidae using 75%-occupancy alignment of UCE dataset. Major lineages are labelled (A-D, *) for reference in the text; clade A was originally considered part of the paramuriceid ingroup but was subsequently determined to be part of a separate family (cf. Rosgorgiidae – see Discussion). The asterisk indicates the clade corresponding to actual Paramuriceidae. Branch numbers indicate posterior probability (expressed as a proportion) support values for the corresponding lineages. Branch tip labels indicate the NIC catalogue number and corresponding identification for included specimens.

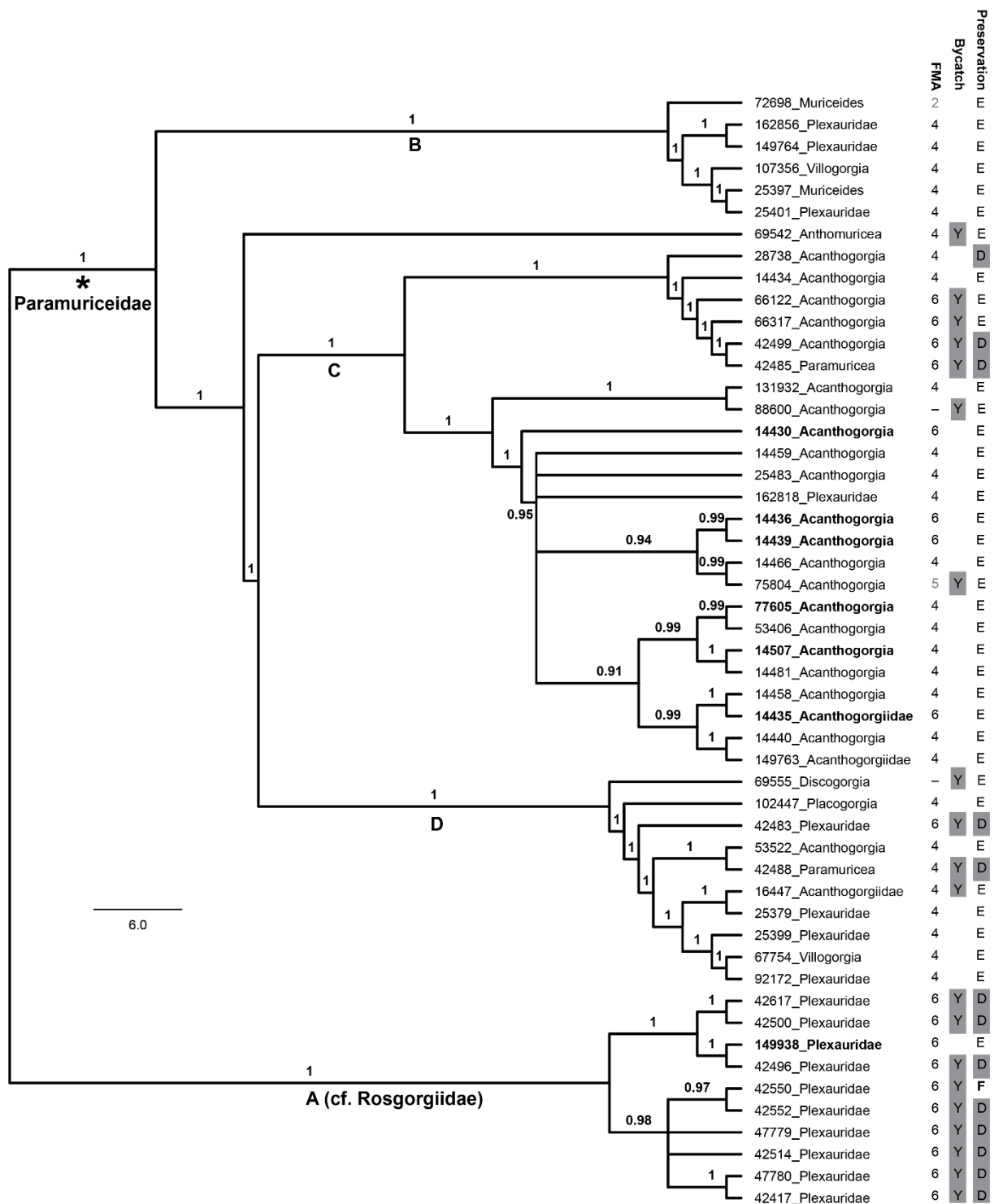


Figure 7: Cladogram of 50%-occupancy UCE alignment showing specimen origin details. Major lineages are labelled (A-D, *) for reference in the text. Branch values indicate Bayesian posterior probability support values, indicated as a proportion. For each specimen, the FMA region from which it originated (2=Central-East, 4=Chatham Rise, 5=Southland, 6=Bounty Rise, '-' = outside EEZ), its status as a trawl bycatch specimen (Y=bycatch), and the long-term preservation method used to archive the specimen (E=ethanol, D=dry, F=frozen) are indicated in the right-hand columns. Noteworthy values are shaded grey (see text for discussion). Specimen names in boldface were collected prior to 1980 (1963-1979).

4 Discussion

These results represent the first in-depth examination of genetic diversity among the Paramuriceidae (formerly the Plexauridae and Acanthogorgiidae) in the world and will be of interest internationally as the family is a common inhabitant of seamounts and other hard-bottom habitats in the deep sea. Although the Paramuriceidae is thought to be one of the most diverse families of protected gorgonian corals in New Zealand (Macpherson et al. 2023), there had been no prior assessment of how many species it represents within the EEZ. The current study represents the first focused examination of diversity for the family, although it lacks representatives of several nominal genera recorded from New Zealand (*Paracis*, *Bebryce*, *Paraplexaura*), and utilises samples from a limited geographic area.

At least 32 taxa that were distributed broadly across two distinct geographic regions were detected with high confidence among seven nominal genera (although some of these genera may be mis-represented as incorrectly identified specimens – see below). The taxonomic status of these Operational Taxonomic Units (OTUs, or genetic variants) remains uncertain, with morphological examinations of the sequenced specimens and comparison to existing species and genera descriptions required to establish whether genetic differences delineate different genera, species, or populations of a single species. *Acanthogorgia* (Clade C, Fig. 7) was noteworthy as it included at least 13 OTUs, although specimens of the genus appear outwardly similar in colony form (Figure 8). As a commonly encountered genus in deep-sea regions across New Zealand, these results suggest that *Acanthogorgia* may comprise a complex of multiple species rather than a single cosmopolitan taxon, and further study of these OTUs is warranted to determine the boundaries of their geographic distributions.

Comparisons of paramuriceid specimens obtained from different regions showed that most major lineages were shared between the Chatham Rise (FMA4) and the Campbell and Bounty plateaus (FMA6). While some OTUs were only found in a single region, their nearest relatives often occurred in the other sampled region. However, in most cases each OTU was represented by a single specimen, thus biogeographical patterns for taxa should be interpreted with caution. Due to the small sample sizes, comparisons should focus on broader lineages containing multiple taxa. Two such notable lineages were Clade B (Fig. 7), which consisted of three OTUs (from five specimens) found only on the Chatham Rise, and Clade A, which represents a distinct family recorded only from the Campbell and Bounty plateaus region (ten specimens) (Figure 8).

Originally suspected to be a new family, the morphologies of the paramuriceid-like gorgonians of Clade A were examined by Dr. Kirrily Moore (Tasmanian Museum and Art Gallery), as part of the DOC-CSP project INT2023-07 (Mills et al. 2024). The ten specimens were tentatively identified as *Rosgorgia* sp. – a member of the family Rosgorgiidae, which is a rare group of gorgonians usually confined to the Antarctic region (Lopez-Gonzalez & Gili 2001). More detailed morphological characterisation and comparison to reference material are required to confirm the identification, but the available UCE sequence data may also be analysed in a broader context (eg. Quattrini et al. 2023) to determine the genetic affinities and relationships of this distinct lineage. Similar work is currently underway for another tentative new gorgonian family first identified in additional DOC projects BCBC2020-26 (Bilewitch 2022) and subsequently examined in INT2022-03 (Connell et al. 2024).

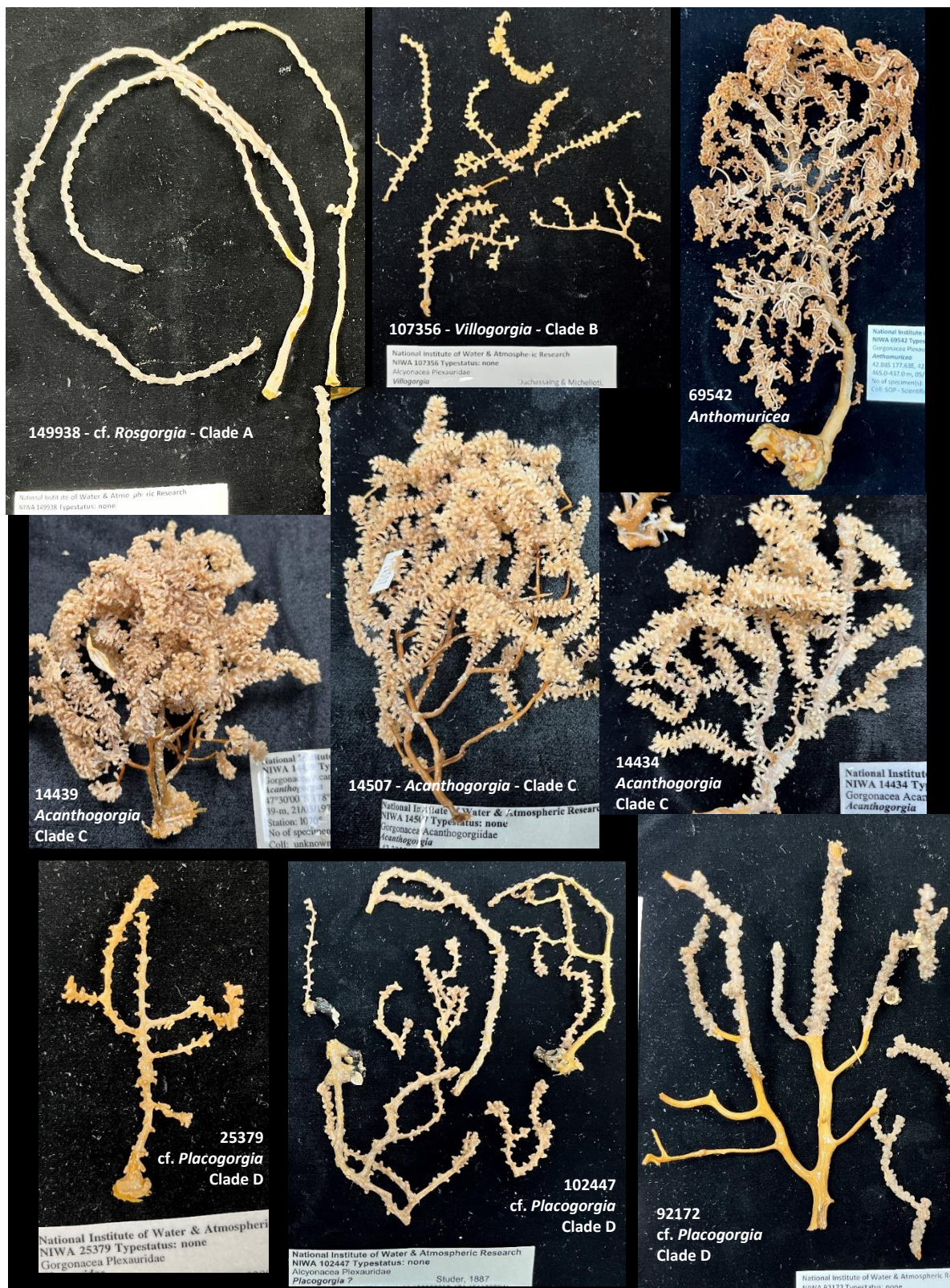


Figure 8: Representatives of gorgonian specimens included for sequencing. Each inset lists the NIC catalogue number, the identification, and the corresponding clade designator within the phylogenetic results. The clade corresponding to *Anthomuricea* specimen (69542) was not given a designator.

This study included representatives of only three paramuriceid genera that were identified by taxonomists or octocoral experts with high confidence: *Acanthogorgia*, *Anthomuricea*, and *Discogorgia* (the latter was included only as a reference). The remaining samples included records of *Muriceides*, *Paramuricea*, *Placogorgia*, and *Villogorgia* and were suspect as they were both identified by individuals not familiar with the Paramuriceidae and they were found as mixed (polyphyletic) assemblages in the resulting phylogenetic trees (in particular, see Clades B & D in Fig. 7). Further morphological examination of these sequenced specimens is required to determine if these genera have been mis-identified or if their associated lineages represent multiple genera. A prior examination of the accuracy of expert morphological identifications for gorgonian corals found that it disagreed with- or was less precise than DNA barcoding results for 21% of cases (Bilewitch & Tracey 2020), illustrating the difficulty and subjectivity inherent in identifying cryptic gorgonian taxa, including the Paramuriceidae.

Regardless of uncertain taxonomic status, the presented results provide a baseline of genetic diversity that is impacted by commercial bottom-trawling. At least 15 of the 32 distinct taxa detected here were represented by specimens returned to the surface by bottom trawling vessels. This compares to previous findings where Bilewitch (2022) found 10 of 13 taxa of Primnoidae present on the Chatham Rise were found among fisheries bycatch, and Bilewitch & Tracey (2020) where 32 of 34 sequenced taxa among five gorgonian families (including seven paramuriceids) across the EEZ were represented as bycatch. Collectively, these studies have discriminated at least 43 distinct taxa among seven families of protected gorgonian corals that are recorded as bycatch from commercial bottom-trawling. However, this count underestimates octocoral diversity in commercial bycatch since it does not include taxa that were identified only by morphology (without genetics; *eg.* Tracey et al. 2017).

The use of genome-scale DNA sequencing methods in the delineation of species and evolutionary relationships ('phylogenomics') has become common-place due to cost reductions in high-throughput sequencing applications combined with their ability to generate large amounts of informative data. In comparison to barcoding methods, for a subset of barcoded gorgonian corals UCE sequencing resulted in more data generation per dollar spent, plus improved resolution and precision of reconstructed species relationships for the Primnoidae (Bilewitch 2022). UCEs also allow for inter-lab comparisons, provided similar sequencing approaches (baitsets, sequencing depth) are followed. Such high-resolution applications are necessary for documenting protected coral diversity since morphology can overlook cryptic diversity (McFadden et al. 2006) and routine DNA barcoding may not fully resolve species-level relationships with high confidence or accuracy (Quattrini et al. 2023). Genomic methods using small DNA fragments such as UCE-sequencing can also take advantage of archival tissues that are of insufficient integrity for barcoding and morphological study. In the current study, we obtained high quality UCE data from seven ethanol-preserved paramuriceid specimens collected in the 1960s and 1970s, plus 13 specimens that were held as air-dried specimens in the NIC for over 20 years. Although still preferable for barcoding methods, these results obviate the requirement for observers and collection managers to rely on freezing and high-grade ethanol preservation to maintain genetically viable voucher material. Furthermore, a recent examination of genome skimming (low-coverage whole-genome DNA sequencing) has shown that it is more effective at recovering UCE loci than target-bait enrichment and carries additional benefits of producing whole (or nearly so-) mitochondrial and ribosomal cistron sequences, which allows reference to older barcoding datasets that are not otherwise comparable to UCE datasets (Quattrini et al. 2024). As a potentially more cost-effective approach that can be implemented without the use of specialised commercial providers overseas, genome skimming will likely replace target-bait enrichment in our future coral diversity studies, including for bycatch characterisation.

5 Recommendations

The following recommendations are made for further research improvements in bycatch documentation and characterisation:

- Incorporation of larger UCE and genome skimming datasets from international studies, to resolve the taxonomic status of OTUs and place unrecognised families of protected gorgonians in a broader context. Datasets have already been obtained and are ready for use (A. Quattrini, pers. comm.)
- Genetic characterisation of more reference and bycatch specimens of Paramuriceidae, to represent the breadth of genus-level diversity for the in New Zealand and improve distributional records for OTUs.
- Summarisation of distribution and taxonomic status (described vs. undescribed taxa) for well-documented taxa of gorgonian octocorals, and identification of taxa in need of further characterisation/study.
- Investigation of the feasibility of incorporating biomass, frequency and genetic characterisation into bycatch documentation and associated considerations of fishing impacts.

The following recommendations are made for operational considerations in bycatch characterisation and potential mitigation:

- Incorporation of uncertainty and potential for undocumented or cryptic species diversity in risk assessments and habitat suitability and hotspot modelling.
- Consideration of diversity (documented and cryptic) and potential for regional endemism when assessing fisheries impacts on protected gorgonian corals.
- Promoting increased coral bycatch sampling by fisheries observers and exploring means for reducing sampling logistics (*eg.* eliminating cold-chain requirements in favour of dried specimens and ethanol-preserved vouchers/sub-samples).

6 Acknowledgements

This research would not have been possible without the efforts of Fisheries Observers, who have been returning valuable coral bycatch specimens to NIWA for more than 20 years. Funding for this project was provided by the Conservation Services Programme at the Department of Conservation and I thank Lyndsey Holland and Katie Clemens-Seeley at DOC for their support and advice. Sample processing and data generation was assisted by Lisa Smith and Amber Brooks in the NIWA Molecular Biology facility, as well as by the NIWA Invertebrate Collections team: Sadie Mills, Diana Macpherson and Amelia Connell. I also acknowledge Arbor Biosciences (USA) and Dr. Andrea Quattrini (USNM) for their advice on UCE sequencing and thank Felix Vaux (NIWA) and the CSP Technical Working Group for reviewing and improving this report.

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Appendix A Sample Collection Metadata

Collection information for examined material from NIWA Invertebrate Collection. Found = sample located and sighted within collections; Sampled = sample subsampled for genetics; [gDNA] = concentration of gDNA extracted from approximately 10mg of tissue; Sequenced = submitted for UCE sequencing; FMA = Chatham Rise (4) or Campbell/Bounty Rise (6); Catalogue = NIC catalogue number; Label = name used in phylogenetic analysis; Date = date of collection; Depth = metres; Determiner = surname of identifier; Preservative = current preservative medium. Noteworthy values are indicated in boldface (*eg.* old, dried, bycatch or misidentified specimens).

Found	Sampled	[gDNA]	Sequenced	FMA	Catalogue	Family	Genus	Label	Station	Date	Lat	Long	Depth	Gear	Determiner	Preservative
Y	Y	0.7		4	9657	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/337	20/04/2001	-42.7	-179.9	970	Sled		Dry
Y	Y	3.3	Y	6	14430	Paramuriceidae	Acanthogorgia	Acanthogorgia	S70	26/09/1978	-47.7	178.5	353	Trawl, Agassiz	Sanchez	Ethanol
Y	Y	23.5	Y	4	14434	Paramuriceidae	Acanthogorgia	Acanthogorgia	X484	4/07/1994	-42.7	-179.9	899	Trawl, Otter	Sanchez	Ethanol
Y	Y	0.8	Y	6	14435	Paramuriceidae		Acanthogorgiidae	G888	14/12/1970	-48.2	177.8	1020	Trawl, Otter		Ethanol
Y	Y	71.0	Y	6	14436	Paramuriceidae	Acanthogorgia	Acanthogorgia	I704	21/03/1979	-48	178.4	475	Trawl, Agassiz	Sanchez	Ethanol
				4	14437	Paramuriceidae	Acanthogorgia	Acanthogorgia	D90	17/05/1963	-43.8	-179	399	Trawl, Agassiz		Ethanol
Y	Y	6.1	Y	6	14439	Paramuriceidae	Acanthogorgia	Acanthogorgia	I705	21/03/1973	-47.5	178.7	390	Dredge		Ethanol
Y	Y	118.1	Y	4	14440	Paramuriceidae	Acanthogorgia	Acanthogorgia	X493	6/07/1994	-43.9	-174.5	665	Trawl, Otter		Ethanol
Y	Y	0.8		4	14447	Paramuriceidae		Acanthogorgiidae	TAN9812/87	26/10/1998	-44.4	-178.6	843	Trawl, Fish, Bottom		Ethanol
Y	Y	5.4	Y	4	14458	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/113	17/04/2001	-42.7	-179.9	900	Sled	Sanchez	Ethanol
Y	Y	40.1	Y	4	14459	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/115	17/04/2001	-42.8	179.9	1013	Sled		Ethanol
Y	Y	1.1		4	14460	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/393	21/04/2001	-42.7	179.9	1009	Sled	Sanchez	Ethanol
Y	Y	1.8		4	14461	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/47	16/04/2001	-42.7	-179.9	950	Sled	Sanchez	Ethanol
Y	Y	0.8		4	14463	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/393	21/04/2001	-42.7	179.9	1009	Sled	Sanchez	Ethanol
Y	Y	5.1	Y	4	14466	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/198	18/04/2001	-42.7	179.9	1058	Sled	Sanchez	Ethanol
Y				4	14470	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/194	18/04/2001	-42.7	-179.9	1042	Sled		Ethanol

Found	Sampled	[gDNA]	Sequenced	FMA	Catalogue	Family	Genus	Label	Station	Date	Lat	Long	Depth	Gear	Determiner	Preservative
Y	Y	16.5	Y	4	14481	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0104/48	16/04/2001	-42.7	-179.9	993	Sled	Sanchez	Ethanol
Y	Y	1.3		4	14483	Paramuriceidae	Acanthogorgia	Acanthogorgia	AEX0101/80	1/11/2001	-44.7	-176.8	753			Ethanol
Y	Y	1.1		4	14493	Paramuriceidae	Acanthogorgia	Acanthogorgia	Q343	14/11/1979	-44.1	175.7	500	Dredge	Sanchez	Ethanol
Y	Y	8.8	Y	4	14507	Paramuriceidae	Acanthogorgia	Acanthogorgia	D871	24/03/1969	-43.3	-178.6	454	Trawl, Agassiz	Sanchez	Ethanol
Y	Y	1.4	Y	4	16447	Paramuriceidae	Villogorgia?	Acanthogorgiidae	TRIP1288/158	4/12/1999	-42.7	176.5	650	Trawl, Fish, Bottom		Ethanol
Y	Y	41.0	Y	4	25379	Paramuriceidae		Plexauridae	TAN0604/6	28/05/2006	-42.7	-179.9	1040	Sled	Sanchez	Ethanol
Y	Y	31.2	Y	4	25397	Paramuriceidae	Muriceides	Muriceides	TAN0604/30	30/05/2006	-42.7	-179.9	951	Sled	Sanchez	Ethanol
Y	Y	78.7	Y	4	25399	Paramuriceidae		Plexauridae	TAN0604/30	30/05/2006	-42.7	-179.9	951	Sled	Sanchez	Ethanol
Y	Y	56.8	Y	4	25401	Paramuriceidae		Plexauridae	TAN0604/31	30/05/2006	-42.7	179.9	1020	Sled	Sanchez	Ethanol
Y	Y	9.8		4	25421	Paramuriceidae		Plexauridae	TAN0604/99	4/06/2006	-42.7	-179.9	890	Sled	Sanchez	Ethanol
Y	Y	22.4	Y	4	25483	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0604/118	7/06/2006	-42.7	179.9	925	Sled	Sanchez	Ethanol
Y	Y	1.1		4	25493	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0408/23	13/07/2004	-42.8	-177.4	826	Trawl, Fish, Bottom	Matsumoto	Ethanol
Y	Y	1.6	Y	4	28738	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0705/94	10/04/2007	-44.5	-178.4	1110	Sled	Sanchez	Dry
Y				4	28778	Paramuriceidae		Plexauridae	TAN0705/189	19/04/2007	-42.7	-178.3	997	Trawl, Beam	Sanchez	Ethanol
Y				4	28785	Paramuriceidae		Plexauridae	TAN0705/191	19/04/2007	-42.7	-178.3	900	Trawl, Beam	Sanchez	Ethanol
Y	Y	8.0		6	41851	(Plexauridae)	(Swiftia)	Swiftia	TRIP2571/122	13/03/2008	-49.9	176.6	839	Trawl, Fish, Bottom	Rowley	Ethanol
Y	Y	1.0	Y	6	42417	Paramuriceidae		Plexauridae	TRIP2571/225	31/03/2008	-49.8	175.8	1038	Trawl, Fish, Bottom	France, S. & Watling	Dry
Y	Y	10.4		4	42481	Paramuriceidae		Plexauridae	TRIP2626/144	14/06/2008	-42.7	179.9	790	Trawl, Fish, Bottom	Sanchez	Dry
Y	Y	6.6	Y	6	42483	Paramuriceidae		Plexauridae	TRIP2653/207	10/08/2008	-50	174.7	911	Trawl, Fish, Bottom	France, S. & Watling	Dry
Y				4	42484	Paramuriceidae		Plexauridae	TRIP2626/98	5/06/2008	-42.7	179.9	869	Trawl, Fish, Bottom	France, S. & Watling	Dry
Y	Y	11.0	Y	6	42485	Paramuriceidae	Paramuricea	Paramuricea	TRIP2653/125	30/07/2008	-50	174.7	859	Trawl, Fish, Bottom	France, S. & Watling	Dry
Y				6	42486	Paramuriceidae	Acanthogorgia	Acanthogorgia	TRIP2653/47	17/07/2008	-49.9	175.5	900	Trawl, Fish, Bottom	Alderslade	Dry
Y	Y	1.8	Y	4	42488	Paramuriceidae	Paramuricea	Paramuricea	TRIP2626/127	12/06/2008	-42.7	179.9	773	Trawl, Fish, Bottom	France, S. & Watling	Dry

Found	Sampled	[gDNA]	Sequenced	FMA	Catalogue	Family	Genus	Label	Station	Date	Lat	Long	Depth	Gear	Determiner	Preservative
Y				6	42492	(Melithaeidae)	(Iciligorgia)	Plexauridae	TRIP2653/111	27/07/2008	-49.8	175.8	1041	Trawl, Fish, Bottom	France, S. & Watling	Frozen
Y	Y	2.3	Y	6	42496	Paramuriceidae		Plexauridae	TRIP2653/40	16/07/2008	-50	174.7	880	Trawl, Fish, Bottom	France, S. & Watling	Dry
Y	Y	32.2	Y	6	42499	Paramuriceidae	Acanthogorgia	Acanthogorgia	TRIP2653/102	27/07/2008	-49.7	175.8	998	Trawl, Fish, Bottom	Moore	Dry
Y	Y	1.5	Y	6	42500	Paramuriceidae		Plexauridae	TRIP2653/43	17/07/2008	-50	175.2	1024	Trawl, Fish, Bottom	France, S. & Watling	Dry
Y				6	42513	(Melithaeidae)	(Iciligorgia)	Plexauridae	TRIP2614/39	12/04/2008	-49.8	175.8	870	Trawl, Fish, Bottom	France, S. & Watling	Frozen
Y	Y	1.8	Y	6	42514	Paramuriceidae		Plexauridae	TRIP2614/127	29/04/2008	-48.5	175.5	920	Trawl, Fish, Bottom	France, S. & Watling	Dry
Y	Y	16.8	Y	6	42550	Paramuriceidae		Plexauridae	TRIP2494/27	4/09/2007	-47.8	175	1057	Trawl, Fish, Bottom	France	Frozen
Y	Y	0.2		6	42551	Paramuriceidae		Plexauridae	TRIP2614/38	12/04/2008	-49.8	175.8	1010	Trawl, Fish, Bottom	France	Dry
Y	Y	10	Y	6	42552	Paramuriceidae		Plexauridae	TRIP2614/124	28/04/2008	-47.7	177.6	950	Trawl, Fish, Bottom	France	Dry
Y	Y	0.9		6	42559	(Plexauridae)	(Swiftia)	Swiftia	TRIP2571/122	13/03/2008	-49.9	176.6	839	Trawl, Fish, Bottom	France	Dry
Y	Y	10.3	Y	6	42617	Paramuriceidae		Plexauridae	TRIP2614/65	16/04/2008	-50	174.7	910	Trawl, Fish, Bottom	Matsumoto	Dry
Y	Y	4.8		4	45310	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0801/16	30/12/2007	-43.4	-179.7	416	Trawl, Fish, Bottom	Sanchez	Ethanol
Y				6	45882	Paramuriceidae		Plexauridae	TRIP2320/116	5/11/2006	-47.6	175.2	818	Trawl, Fish, Bottom	Tracey, Di & Sanchez	Ethanol
Y				6	47778	(Melithaeidae)	(Iciligorgia)	Plexauridae	TRIP2614/39	12/04/2008	-49.8	175.8	870	Trawl, Fish, Bottom	France	Ethanol
Y	Y	1.0	Y	6	47779	Paramuriceidae		Plexauridae	TRIP2494/34	5/09/2007	-48.2	174.6	1048	Trawl, Fish, Bottom	France	Dry
Y	Y	0.4	Y	6	47780	Paramuriceidae		Plexauridae	TRIP2614/48	13/04/2008	-50	175.2	1020	Trawl, Fish, Bottom	France	Dry
Y				6	47781	Paramuriceidae		Plexauridae	TRIP2614/232	12/05/2008	-50	175.2	1020	Trawl, Fish, Bottom	France	Dry
Y				6	47782	Paramuriceidae		Plexauridae	TRIP2614/147	1/05/2008	-49.3	176.3	1210	Trawl, Fish, Bottom	France	Dry
Y				4	47785	(Alcyoniidae)	(Anthothela)	Plexauridae	TRIP2699/17	2/10/2008	-44.4	-174.8	1008	Trawl, Fish, Bottom	France	Frozen
Y				6	47786	Paramuriceidae		Plexauridae	TRIP2614/145	1/05/2008	-48.8	175.6	915	Trawl, Fish, Bottom	Matsumoto	Dry
				4	53162	Paramuriceidae		Plexauridae	TAN0905/42	18/06/2009	-42.7	-179.9	1051	Sled	Sanchez	Ethanol
Y	Y	73.2		4	53323	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0905/60	20/06/2009	-42.8	-179.5	1251	Sled	Sanchez	Dry
Y	Y	70.8	Y	4	53406	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0905/66	22/06/2009	-42.8	-179.9	795	Sled	Cairns	Ethanol

Found	Sampled	[gDNA]	Sequenced	FMA	Catalogue	Family	Genus	Label	Station	Date	Lat	Long	Depth	Gear	Determiner	Preservative
Y	Y	88.4	Y	4	53522	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN0905/71	22/06/2009	-42.7	-179.6	820	Sled	Alderslade	Ethanol
Y				6	65539	(Melithaeidae)	(Iciligorgia)	Plexauridae	TRIP2832/75	5/05/2009	-47.4	175	1061	Trawl, Fish, Bottom	Sanchez	Ethanol
				6	65922	Paramuriceidae		Plexauridae	TRIP2718/23	13/11/2008	-48.9	175.6	869	Trawl, Fish, Bottom	Fenwick	Frozen
Y	Y	116.1	Y	6	66122	Paramuriceidae	Acanthogorgia	Acanthogorgia	TRIP2718/59	17/11/2008	-50	174.7	857	Trawl, fish, bottom	Schnabel	Ethanol
				4	66296	Paramuriceidae		Acanthogorgiidae	TRIP2895/56	10/07/2009	-42.8	-176.9	689	Trawl, Fish, Bottom	Sanchez	Frozen
Y	Y	68.6	Y	6	66317	Paramuriceidae	Acanthogorgia	Acanthogorgia	TRIP2920/96	28/09/2009	-50.1	174.9	1005	Trawl, Fish, Bottom	Sanchez	Ethanol
Y				4	67752	Paramuriceidae	Villogorgia	Villogorgia	TAN0104/396	21/04/2001	-42.7	-179.9	1040	Sled	Sanchez	Ethanol
Y	Y	13.1	Y	4	67754	Paramuriceidae	Villogorgia	Villogorgia	TAN0604/30	30/05/2006	-42.7	-179.9	951	Sled	Sanchez	Ethanol
Y	Y	5.1	Y	4	69542	Paramuriceidae	Anthomuricea	Anthomuricea	TRIP3235/23	5/12/2010	-42.8	177.6	465	Trawl, fish, bottom	Alderslade	Ethanol
Y	Y	83.9	Y	4	77605	Paramuriceidae	Acanthogorgia	Acanthogorgia	A910	13/09/1963	-43	-178.6	549	Dredge	Cairns	Ethanol
Y	Y	159.2		6	90000	(Plexauridae)	(Swiftia)	Swiftia	Z10334	16/10/2000	-48.2	174.4	1050		Mills	Dry
Y	Y	1.9	Y	4	92172	Paramuriceidae	Anthomuricea?	Plexauridae	X483	4/07/1994	-42.7	-179.9	890		Alderslade	Ethanol
Y	Y	2.7		4	102330	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN1503/56	3/04/2015	-42.7	-179.9	918	Sled		Ethanol
Y	Y	21.2		4	102344	Paramuriceidae	Acanthogorgia	Acanthogorgia	TAN1503/67	4/04/2015	-42.7	179.9	936	Sled		Ethanol
Y	Y	156.1	Y	4	102447	Paramuriceidae	Placogorgia	Placogorgia	TAN1503/103	9/04/2015	-44.1	-174.4	1099	Sled	Moore	Ethanol
Y	Y	1.2	Y	4	107356	Paramuriceidae	Villogorgia	Villogorgia	TAN0104/394	21/04/2001	-42.7	-179.9	920	Sled	Bilewitch	Ethanol
Y	Y	3.2		4	123399	Paramuriceidae		Plexauridae	X492	6/07/1994	-43.9	-174.5	662		Cairns	Dry
				4	131932	Paramuriceidae	Acanthogorgia	Acanthogorgia	TRIP5844/37	4/12/2019	-42.7	-177.3	1182	Trawl, fish, bottom	Tracey	Ethanol
Y				4	140314	(Anthothelidae)		Plexauridae	TAN1903/106	21/06/2019	-43.3	179.4	396	Sled		Ethanol
Y	Y	2.9	Y	4	149763	Paramuriceidae		Acanthogorgiidae	TAN0104/48	16/04/2001	-42.7	-179.9	993	Sled		Ethanol
Y	Y	26.6	Y	4	149764	Paramuriceidae	Villogorgia?	Plexauridae	TAN0104/2	15/04/2001	-42.7	-179.9	875	Sled		Ethanol
Y				4	149842	(Primnoidae)	(Thouarella)	Acanthogorgiidae	V373	13/09/1989	-43.6	179	392	Trawl, Agassiz		Ethanol
Y	Y	193.6	Y	6	149938	Paramuriceidae	Clematissa?	Plexauridae	I674	14/03/1979	-48	-179.1	750	Trawl, Agassiz		Ethanol

Found	Sampled	[gDNA]	Sequenced	FMA	Catalogue	Family	Genus	Label	Station	Date	Lat	Long	Depth	Gear	Determiner	Preservative
Y	Y	107.1	Y	4	162818	Paramuriceidae	Acanthogorgia?	Plexauridae	X486	4/07/1994	-42.7	-179.9	910	Trawl, Otter		Ethanol
Y				4	162825	(Alcyoniidae)	(Anthothela)	Plexauridae	X486	4/07/1994	-42.7	-179.9	910	Trawl, Otter		Ethanol
Y				4	162828	(Alcyoniidae)	(Anthothela)	Plexauridae	X486	4/07/1994	-42.7	-179.9	910	Trawl, Otter		Ethanol
Y	Y	37.5	Y	4	162856	Paramuriceidae	Bebryce?	Plexauridae	TAN0104/288	19/04/2001	-42.7	-179.9	972	Sled		Ethanol
Y				4	162865	(Alcyoniidae)	(Anthothela)	Plexauridae	J59	20/05/1970	-43.8	179.4	309	Dredge		Ethanol
Y				6	162870	(Alcyoniidae)	(Anthothela)	Plexauridae	Z9343	29/09/1998	-48.9	175.5	910			Ethanol
Y				4	162899	(Alcyoniidae)	(Anthothela)	Plexauridae	Q24	22/03/1978	-44.4	-176.5	320	Dredge, rock		Ethanol
Y	Y	-	Y		75804	Paramuriceidae	Acanthogorgia sp2	Acanthogorgia	TRIP3426/57	14/01/2012	-48.7	166.3	608	Trawl, fish, bottom	Alderslade	Ethanol
Y	Y	-	Y		131932	Paramuriceidae	Acanthogorgia sp2	Acanthogorgia	TRIP5844/37	04/12/2019	-42.7	-177.3	1182	Trawl, fish, bottom		Ethanol
Y	Y	-			106577	Paramuriceidae	Plexauridae sp1		TRIP5117/16	02/09/2017	-40.6	177	228	Trawl, fish, bottom		Ethanol
Y	Y	-	Y		69604	Paramuriceidae	Paracis squamata	Paracis	TRIP3248/17	15/12/2010	-32.5	166.7	367	Bottom longline	Schnabel	Ethanol
Y	Y	-	Y		72698	Paramuriceidae	Muriceides	Muriceides	TAN1104/102	17/03/2011	-35.7	178.5	440	Sled	Alderslade	Ethanol
Y	Y	-	Y		69555	Paramuriceidae	Discogorgia	Discogorgia	TRIP3246/23	31/12/2010	-35.6	165.9	747	Trawl, fish, bottom	Schnabel	Ethanol
Y	Y	-			95114	Paramuriceidae	Acanthogorgia sp1	Acanthogorgia	TRIP4255/43	02/12/2014	-34.2	172.9	137	Trawl, fish, bottom	Schnabel	Ethanol
Y	Y	-	Y		88600	Paramuriceidae	Acanthogorgia sp1	Acanthogorgia	TRIP3812/20	12/07/2013	-35.5	165.2	928	Trawl, fish, bottom	Schnabel	Ethanol

Appendix B Target-bait enrichment DNA sequencing summary

Summary of outputs from UCE bioinformatic workflow, including results of DNA sequencing post-quality control (Sequencing), after de novo assembly (Assembly), and after matching to UCE loci (UCEs). Reads = read counts; total bp = sum of sequence lengths; mean length = mean read or contig length in bp; SE length = standard error of mean length; contigs = count of assembled contigs; 95% CI length = \pm length in bp; min length, max length = length in bp; contigs >1kb = count of contigs exceeding 1000bp in length.

sample	Sequencing				Assembly							UCEs						
	reads	total bp	mean length	SE length	contigs	total bp	mean length	95% CI length	min length	max length	contigs >1kb	contigs	total bp	mean length	95% CI length	min length	max length	contigs >1kb
102447_Placogorgia	3.05E+06	4.55E+08	149	0.004	7.24E+03	3.51E+06	485	4.802	56	7283	595	2.25E+03	1.91E+06	849	4.321	148	2596	334
107356_Villogorgia	8.64E+06	9.31E+08	108	0.009	3.01E+04	5.09E+06	169	1.271	55	18747	167	2.25E+03	9.44E+05	419	2.826	206	2673	13
131932_Acanthogorgia	1.61E+06	2.07E+08	129	0.023	1.01E+05	8.67E+06	86	0.325	54	19101	127	1.05E+03	3.30E+05	314	3.133	65	1172	3
14430_Acanthogorgia	1.79E+06	1.77E+08	99	0.020	1.09E+04	9.62E+05	88	0.775	54	1810	11	2.21E+02	5.61E+04	254	5.559	155	1222	1
14434_Acanthogorgia	7.84E+06	9.14E+08	117	0.011	4.20E+04	6.86E+06	163	1.171	54	19019	436	2.24E+03	1.17E+06	521	4.400	102	3119	56
14435_Acanthogorgiidae	8.66E+06	9.59E+08	111	0.010	1.84E+05	2.42E+07	132	0.369	51	19114	744	2.50E+03	9.80E+05	392	2.193	163	1400	6
14436_Acanthogorgia	8.61E+06	1.01E+09	117	0.010	4.50E+04	1.05E+07	233	1.742	69	13634	1261	1.72E+03	8.10E+05	472	13.892	228	13634	95
14439_Acanthogorgia	8.60E+06	8.86E+08	103	0.009	1.89E+05	1.60E+07	85	0.208	51	18480	210	1.48E+03	4.71E+05	319	4.187	78	1851	22
14440_Acanthogorgia	8.65E+06	1.11E+09	129	0.009	5.87E+04	9.98E+06	170	0.950	52	7623	669	1.94E+03	1.20E+06	617	5.259	117	3672	74
14458_Acanthogorgia	8.63E+06	9.47E+08	110	0.010	3.07E+04	5.61E+06	183	1.204	71	18727	225	1.88E+03	7.27E+05	386	4.916	155	5646	14
14459_Acanthogorgia	2.71E+06	3.06E+08	113	0.018	8.36E+04	9.35E+06	112	0.639	51	11268	429	8.90E+01	2.59E+04	291	13.042	90	874	0
14466_Acanthogorgia	8.62E+06	9.49E+08	110	0.010	2.91E+04	6.54E+06	225	1.503	72	19136	351	1.88E+03	8.17E+05	435	4.069	160	2475	29
14481_Acanthogorgia	8.62E+06	9.03E+08	105	0.010	1.26E+05	1.54E+07	122	0.500	47	13493	706	2.29E+03	9.62E+05	420	3.552	78	5481	17
14507_Acanthogorgia	8.58E+06	9.34E+08	109	0.010	3.57E+04	7.25E+06	203	1.806	72	19136	598	1.51E+03	4.53E+05	301	2.261	228	2214	3
149763_Acanthogorgiidae	4.90E+06	5.66E+08	115	0.013	8.12E+04	9.34E+06	115	0.491	51	5825	291	2.42E+03	8.98E+05	371	2.469	108	2048	12
149764_Plexauridae	8.63E+06	1.18E+09	136	0.008	2.11E+04	5.94E+06	281	2.176	50	18747	492	2.42E+03	1.57E+06	648	4.205	111	3598	97
149938_Plexauridae	2.38E+06	3.55E+08	149	0.005	4.64E+03	2.81E+06	605	6.056	56	6973	504	2.21E+03	1.84E+06	832	4.185	221	1839	297

sample	Sequencing				Assembly							UCEs						
	reads	total bp	mean length	SE length	contigs	total bp	mean length	95% CI length	min length	max length	contigs >1kb	contigs	total bp	mean length	95% CI length	min length	max length	contigs >1kb
162818_Plexauridae	8.67E+06	1.16E+09	134	0.008	2.08E+04	4.50E+06	217	1.551	53	3542	213	1.82E+03	1.12E+06	613	4.078	62	2249	49
162856_Plexauridae	1.73E+07	2.17E+09	125	0.007	6.71E+04	9.48E+06	141	0.769	54	19456	436	2.45E+03	1.38E+06	565	3.132	217	1746	46
16447_Acanthogorgiidae	1.72E+07	1.94E+09	113	0.007	2.06E+05	2.35E+07	114	0.316	49	10065	828	2.36E+03	8.25E+05	349	2.264	61	2890	6
25379_Plexauridae	1.73E+07	2.26E+09	131	0.006	1.03E+05	1.28E+07	125	0.536	53	6434	676	2.28E+03	1.44E+06	631	4.926	103	3136	153
25397_Muriceides	1.31E+07	1.66E+09	127	0.007	1.29E+04	3.60E+06	279	2.480	56	8765	224	2.28E+03	1.28E+06	559	3.889	106	3926	52
25399_Plexauridae	1.73E+07	2.31E+09	133	0.006	1.39E+04	4.39E+06	316	2.446	55	5343	371	2.25E+03	1.48E+06	657	4.343	112	2762	121
25401_Plexauridae	1.72E+07	2.26E+09	132	0.006	5.81E+04	1.14E+07	197	1.157	54	18747	996	2.33E+03	1.45E+06	621	5.052	58	3610	107
25483_Acanthogorgia	1.72E+07	2.06E+09	120	0.007	2.17E+05	2.04E+07	94	0.233	48	19114	348	1.99E+03	8.94E+05	449	3.844	80	3126	22
28738_Acanthogorgia	1.72E+07	1.78E+09	103	0.007	4.14E+04	4.71E+06	114	0.698	69	19041	112	8.88E+02	2.69E+05	303	4.008	86	2251	5
42417_Plexauridae	1.72E+07	1.67E+09	97	0.006	9.08E+04	1.07E+07	118	0.370	68	18791	48	1.84E+03	6.29E+05	342	1.661	197	1022	1
42483_Plexauridae	1.72E+07	1.90E+09	111	0.007	1.89E+04	4.35E+06	230	1.607	77	19062	122	2.18E+03	9.00E+05	414	2.526	88	1515	13
42485_Paramuricea	1.72E+07	1.84E+09	106	0.007	1.54E+04	4.29E+06	278	2.223	77	13442	236	1.88E+03	8.23E+05	437	4.884	181	6182	20
42488_Paramuricea	1.73E+07	2.02E+09	117	0.007	1.32E+05	1.51E+07	115	0.190	72	1174	4	7.31E+02	2.85E+05	389	3.362	78	832	0
42496_Plexauridae	8.43E+06	1.06E+09	126	0.010	2.38E+04	8.83E+06	372	3.850	78	57432	542	2.01E+03	8.40E+05	419	3.972	228	2767	31
42499_Acanthogorgia	1.73E+07	1.93E+09	112	0.007	2.23E+04	6.71E+06	300	2.175	70	19041	548	1.89E+03	9.38E+05	497	5.598	230	4084	48
42500_Plexauridae	1.73E+07	2.02E+09	117	0.007	6.88E+04	1.45E+07	211	0.703	75	5964	403	1.82E+03	7.42E+05	409	2.923	186	2244	4
42514_Plexauridae	9.71E+06	1.18E+09	121	0.009	9.26E+03	6.32E+06	682	12.869	78	35249	1242	5.73E+02	1.83E+05	320	27.710	95	15611	5
42550_Plexauridae	2.74E+06	3.65E+08	134	0.015	2.70E+04	1.01E+07	373	2.393	56	6905	1643	2.00E+01	5.95E+03	298	49.277	96	1178	1
42552_Plexauridae	1.27E+07	1.52E+09	120	0.008	1.14E+05	2.10E+07	184	0.482	75	9952	380	2.80E+02	7.74E+04	276	3.918	83	720	0
42617_Plexauridae	9.88E+06	1.28E+09	129	0.009	4.91E+04	1.29E+07	263	1.363	78	18752	1221	1.79E+02	5.30E+04	296	11.180	154	1651	2
47779_Plexauridae	1.73E+07	1.82E+09	105	0.007	2.03E+04	6.61E+06	326	2.134	77	18788	467	2.14E+03	8.35E+05	391	4.225	169	6278	18
47780_Plexauridae	1.73E+07	1.85E+09	106	0.006	8.75E+04	1.48E+07	170	0.658	70	18791	376	2.06E+03	7.17E+05	347	1.678	160	1288	1

sample	Sequencing				Assembly							UCEs						
	reads	total bp	mean length	SE length	contigs	total bp	mean length	95% CI length	min length	max length	contigs >1kb	contigs	total bp	mean length	95% CI length	min length	max length	contigs >1kb
53406_Acanthogorgia	1.73E+07	2.43E+09	141	0.005	1.17E+04	4.56E+06	391	3.791	78	7343	712	1.32E+03	1.03E+06	776	8.823	95	3910	168
53522_Acanthogorgia	1.73E+07	2.28E+09	132	0.006	9.63E+03	3.89E+06	404	3.591	78	6140	532	1.43E+03	9.88E+05	692	7.535	92	3708	132
66122_Acanthogorgia	1.73E+07	2.20E+09	127	0.007	9.90E+03	3.79E+06	383	4.252	77	9417	598	1.43E+03	9.46E+05	663	8.890	80	3828	130
66317_Acanthogorgia	1.73E+07	2.20E+09	127	0.006	2.39E+04	5.56E+06	233	1.891	54	13807	451	2.46E+03	1.42E+06	577	4.034	193	2953	76
67754_Villoorgia	1.72E+07	2.12E+09	123	0.007	1.13E+04	4.50E+06	399	4.322	77	19062	581	2.03E+03	1.11E+06	546	6.161	91	4984	70
69542_Anthomuricea	2.87E+06	3.43E+08	120	0.017	3.57E+03	9.47E+05	266	5.180	77	17709	5	1.63E+03	4.85E+05	297	1.616	99	1451	2
69555_Discogorgia	8.76E+05	9.56E+07	109	0.036	5.95E+02	1.10E+05	185	22.558	78	12565	4	4.70E+01	1.30E+04	277	6.886	228	414	0
69604_Paracis	1.65E+05	1.89E+07	114	0.105	7.10E+01	9.24E+03	130	12.268	78	812	0	-	-	-	-	-	-	-
72698_Muriceides	1.40E+07	2.09E+09	149	0.002	1.81E+04	4.83E+06	267	2.295	67	4119	669	2.13E+03	1.82E+06	853	4.853	180	1998	434
75804_Acanthogorgia	6.69E+06	8.49E+08	127	0.011	1.76E+04	4.67E+06	265	2.855	72	19136	485	1.37E+03	4.80E+05	351	5.099	228	3982	16
77605_Acanthogorgia	1.73E+07	2.22E+09	128	0.006	2.36E+04	5.46E+06	231	1.733	53	5519	432	2.14E+03	1.27E+06	592	4.720	59	2911	76
88600_Acanthogorgia	1.74E+06	2.10E+08	121	0.026	3.31E+04	3.49E+06	106	0.669	52	4619	93	9.74E+02	2.96E+05	303	3.333	69	1388	3
92172_Plexauridae	1.72E+07	1.85E+09	107	0.007	3.02E+04	4.78E+06	158	1.084	54	10338	142	2.47E+03	9.93E+05	402	2.523	102	2156	18
102618_Primnoella	5.78E+06	8.60E+08	149	0.004	1.58E+04	4.30E+06	272	2.512	55	4125	686	-	-	-	-	-	-	-
131891_Radicipes	2.09E+06	2.95E+08	141	0.014	4.07E+03	5.43E+06	1334	61.401	56	59212	557	-	-	-	-	-	-	-
41313_Callipodium	1.74E+07	2.47E+09	142	0.004	6.12E+03	2.84E+06	464	4.339	77	5136	281	-	-	-	-	-	-	-
42522_T_laxa	5.24E+04	6.00E+06	115	0.186	7.50E+01	1.68E+04	224	19.183	79	822	0	-	-	-	-	-	-	-
53309_aff_T_laxa	1.72E+07	2.37E+09	137	0.005	2.05E+05	2.04E+07	100	0.142	71	11998	20	-	-	-	-	-	-	-
88693_Metallogorgia	1.72E+07	2.39E+09	139	0.005	3.15E+04	5.21E+06	165	0.997	54	4139	164	-	-	-	-	-	-	-
88879_Lepidisis	1.73E+07	2.47E+09	143	0.004	1.29E+04	3.29E+06	255	2.597	50	19284	170	-	-	-	-	-	-	-