

# Cryptic diversity, erroneous species identification, sequencing errors, or shadows of nuclear mitochondrial DNA segments (NUMTs)? A case study in *Panulirus* lobsters

Seinen Chow<sup>1,2\*</sup>, Philippe Borsa<sup>3</sup>

<sup>1</sup>Fisheries Resources Institute, 2-12-4 Fukuura, Yokohama, Kanagawa 236-8648, Japan. <sup>2</sup>Aquos Institute, 3-2153-79 Motohachioji, Hachioji, Tokyo 193-0826 Japan. <sup>3</sup>Institut de recherche pour le développement - UMR 250 S, Nouméa, New Caledonia and Montpellier, France.

\*Corresponding author, email: kaiyoeell@gmail.com

## Abstract

DNA barcoding using mitochondrial DNA relies on accurate DNA databases. However, it is not uncommon for these DNA databases to contain nucleotide sequences with erroneous species labels. Furthermore, sequencing errors and nuclear mitochondrial DNA (NUMT) sequences may impact DNA barcoding. In the present study, we applied phylogenetic, nucleotide sequence, and deduced amino-acid sequence analyses to the 1,157 unique COI haplotypes of the 22 spiny lobster species of the genus *Panulirus* collected from the GenBank database and from a published article. Based on the large number of amino-acid substitutions, large nucleotide-sequence divergence, and odd phylogenetic placement, one haplotype was found to be misidentified to species. Of 55 haplotypes determined to be problematic, 27 haplotypes obtained from cloned amplicons were presumed to be NUMTs and 28 haplotypes obtained by direct nucleotide sequencing were presumed to be NUMT-contaminated sequences. A small number of non-synonymous nucleotide substitutions were observed in 96 other haplotypes, possibly caused by sequencing errors or slight NUMT-contamination. The unnoticed inclusion of NUMTs or NUMT-contaminated sequences in nucleotide sequence datasets may inadvertently inflate intraspecific genetic diversity and distort phylogenies.

**Key words:** COI; quality control; spiny lobster; pseudogene

## Introduction

The mitochondrial DNA (mtDNA) cytochrome c oxidase subunit I (COI) gene, which has been proposed as a universal DNA barcode for species identification (Hebert et al. 2003a), is now the most sequenced gene in animals (Pentinsaari et al. 2016). Genetic information from accurately identified specimens is essential to building a reliable DNA barcode database (e. g. BOLD: Ratnasingham and Hebert 2007; GenBank: Benson et al. 2017). However, species misidentification is not rare, and species entries in barcode databases may include distinct species, cryptic or not (Meiklejohn et al. 2019; Pentinsaari et al. 2020; Stein and Gailing 2025). Minor errors in nucleotide sequencing generally do not have a major impact on species identification but can bias

estimates of genetic diversity. MtDNA copies inserted in the nuclear genome have been observed in a wide variety of eukaryotes (Hazkani-Covo et al. 2010). These mtDNA copies, coined as NUMTs (Lopez et al. 1994), may be co-amplified with authentic mtDNA, which can complicate the process of DNA barcoding and affect the outcomes of population genetic and evolutionary studies. The unusual placement of haplotypes with long branches in a phylogenetic tree may thus indicate cryptic diversity, sequencing error, erroneous species identification, or NUMT contamination. The unnoticed incorporation of NUMTs has led to erroneous conclusions regarding species identity, population genetic structure, and phylogenetic reconstruction (Song et al. 2008; Buhay 2009). Fukuda et al. (1985) have been the first to report

several hundreds of NUMTs in the human nuclear genome. NUMTs are now considered common in animals (Bensasson et al. 2001; Song et al. 2008; Hebert et al. 2023).

Buhay (2009) presented several nucleotide-sequence chromatograms for the partial COI gene of crayfish, in which NUMT contamination was thought to have generated sloppiness. This author also reported problematic crustacean COI-like sequences deposited in the GenBank database that contained NUMT signals. Using clone-library-based nucleotide sequence analysis, NUMTs of the COI gene were also detected in the Japanese spiny lobster (*Panulirus japonicus*) and in the Aesop slipper lobster (*Scyllarides haanii*) (Chow et al. 2021, 2024b). In these two species, chromatograms obtained by direct nucleotide sequencing showed admixture of signals from authentic mtDNA and NUMTs. Even when the peaks of the chromatograms obtained through direct nucleotide sequencing were readable, NUMT contamination occasionally impacted our ability to obtain nucleotide sequences from authentic mtDNA (Chow et al. 2021, 2024a, b).

The number of NUMTs differs considerably among animal taxa and is strongly correlated with genome size (Pamilo et al. 2007; Hazkani-Covo et al. 2010), while the number of putative NUMTs detected can also vary among conspecific individuals and with the tissue from which the DNA is extracted (Bensasson et al. 2001; Gíslason et al. 2013; Chow et al. 2024a). In many cases, the signal peaks of authentic mtDNA largely dominate over those possibly contributed by NUMTs. However, there are cases where the signal peaks of both types of amplicons are comparable, and sometimes the intensity of the peaks contributed by NUMTs exceeds that of the authentic mtDNA amplicon (e. g. Sorenson and Fleischer 1996; Parr et al. 2006; Chow et al. 2021, 2024b). Contamination by NUMTs with indels results in deteriorated chromatograms. It empirically seems that double peaked or deteriorated chromatograms are more

frequently encountered in crustaceans than in fishes or mollusks (Williams and Knowlton 2001; Buhay 2009; Baeza and Fuentes 2013; Gíslason et al. 2013; Iacchei et al. 2014; Woodings et al. 2019; Chow et al. 2021, 2024a, b).

The amino-acid sequence provides several clues to assess the quality of the nucleotide sequence. The highly conserved COI gene encodes a key enzyme of the respiratory chain of mitochondria (Castresana et al. 1994). At the COI locus, as for other conservative protein-coding genes, there should be no stop codons and indels should be rare, even among distantly related animal taxa. One also expects few amino-acid differences among conspecific individuals or even among closely related taxa.

In this study, we critically examined publicly available nucleotide sequences at the COI locus of all spiny lobster species in the genus *Panulirus*. The lobster genus *Panulirus* currently comprises 22 species (DecaNet eds. 2025), of which 16 are distributed in the Indo-Pacific and the remaining six in the Atlantic. We propose criteria to evaluate the quality of COI-gene sequences and to eventually detect NUMTs in this genus, chosen as a model case.

## Materials and Methods

The nucleotide sequences of the COI gene of all 22 *Panulirus* species were obtained from the GenBank database (<https://www.ncbi.nlm.nih.gov/nucleotide/>; last accessed 09 September 2025) and from a published article (Table S1). These sequences included the 5' end of the gene, corresponding to the "Folmer region" (Folmer et al. 1994) used for DNA barcoding. Alignments of nucleotide sequences and their translation into amino-acid sequences using the invertebrate mitochondrial code were performed using GENETYX ver. 12 (GENETYX Co., Tokyo). The most Common Amino-acid Sequences within a species was abbreviated as "CAS". In the case of identical nucleotide sequences in a species, only the longest sequence was retained as the sequence defining the

corresponding haplotype. COI-gene sequences shorter than 400 bp and those containing ambiguous nucleotides were not used. The Kimura 2-parameter (K2P) distance between nucleotide sequences and the number of residue differences and the p-distance between deduced amino-acid sequences were calculated using MEGA 6 (Tamura et al. 2013). Model selection and the construction of phylogenetic trees were also done using MEGA 6.

Buhay (2009) flagged problematic COI nucleotide sequences of decapod crustaceans in the GenBank database. Here, we separated these flagged sequences into two categories. The first category, symbolized by one red flag, included haplotypes that differed by one or two amino-acid residues from the CAS and/or that showed comparable or slightly larger K2P distance than the lowest threshold (2% to 3%) reported between congeneric animal species (Hebert et al. 2003a, b). This K2P threshold is reasonable for lobster, as the COI nucleotide sequence divergence between putative subspecies of *Panulirus homarus* ranges from 1.3 % to 9.3 % and that within subspecies is smaller than 2.0 % (Lavery et al. 2014). Ptacek et al. (2001) reported the COI nucleotide sequence divergence between the two subspecies of *Panulirus longipes* (*P. longipes longipes* and *P. l. bispinosus*) to be 6.0 % and those between good species of the genus *Panulirus* to be larger than 12.4 %. The second category, symbolized by two red flags, included haplotypes that differed by three or more amino-acid residues from the CAS and/or that were separated by much larger K2P distances than the 2 % to 3 % threshold. It cannot be known whether the haplotypes of the first category represent true sequences, or have sequencing errors, or suffer from slight contamination by NUMTs. The haplotypes of the second category may either be NUMTs, or sequences heavily contaminated by NUMTs including chimera formation, or cryptic species, or they may represent misidentified individuals from another species. Since most of the nucleotide sequences were obtained by

direct sequencing, the sequences identified as problematic may contain sequencing errors, signals from non-specific PCR products, or contamination by NUMTs, but cannot represent a single NUMT. Haplotype sequences with two red flags were entered as queries into a BLAST search in GenBank (<https://blast.ncbi.nlm.nih.gov/>) to find the most similar homologous sequences present in the database while excluding self-matches.

## Results and Discussion

Of 2,049 COI-gene sequences examined, 1,157 were retained after initial screening (Table S1).

### Indo-Pacific *Panulirus* species

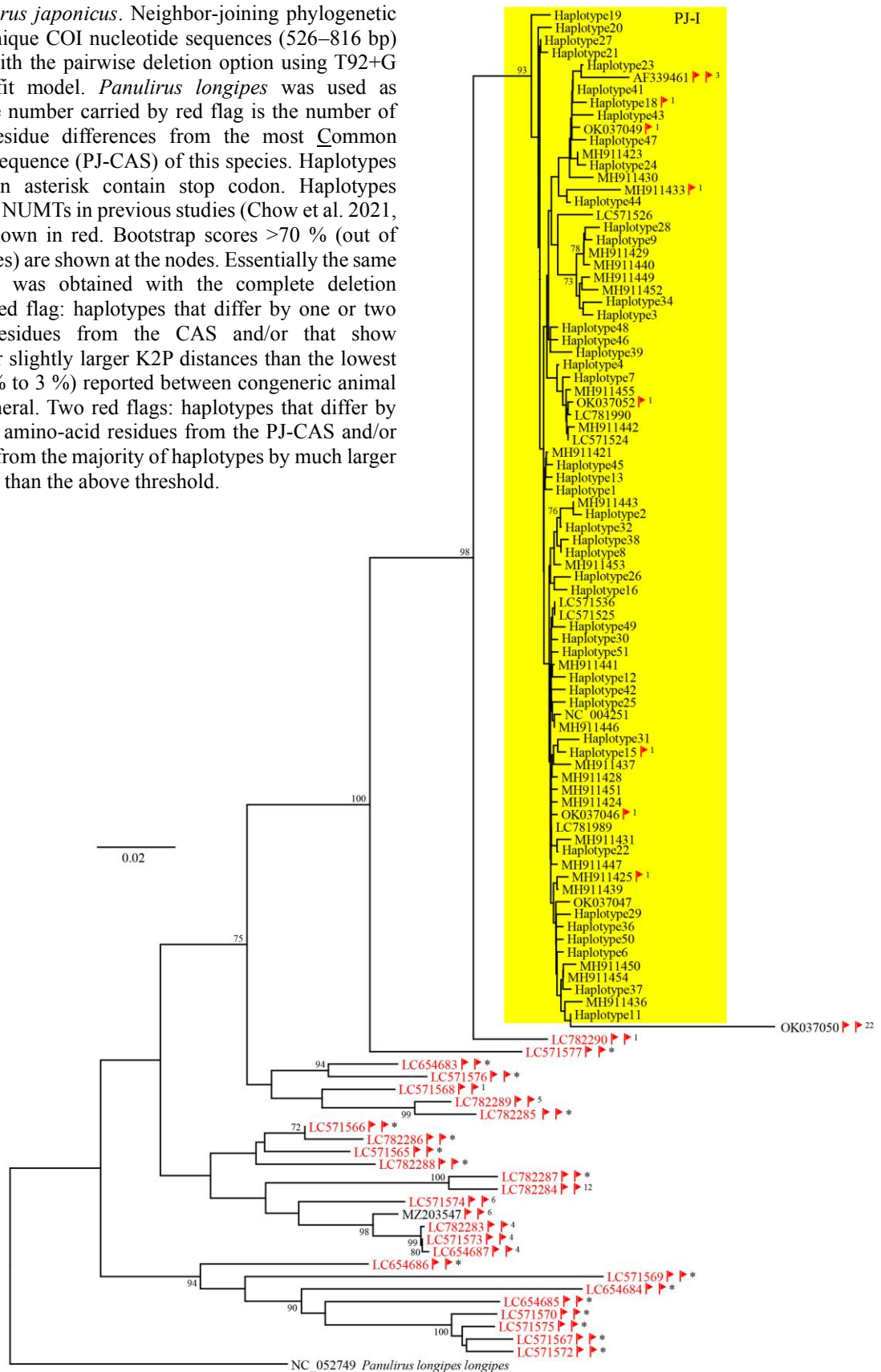
#### *Panulirus japonicus* (Fig. 1)

This species is confined to subtropical to temperate regions in the western North Pacific, including Japan, Korea, China, and Taiwan (Holthuis 1991), and no genetic population structuring has been reported (Inoue et al. 2007; Chang et al. 2019).

Of 146 sequences collected from GenBank and a published article, 108 unique haplotypes were retained for phylogenetic analysis (Table S1). Most of the haplotypes (n=81) belonged to one large haplogroup (coined “PJ-I”), in which the K2P distance between haplotypes ranged from 0.2 % to 3.5 %. The deduced amino-acid sequences of 73 out of the 81 haplotypes in PJ-I were identical: they represented the CAS (“PJ-CAS”) in this species. The deduced amino-acid sequences of the remaining eight haplotypes in PJ-I differed from the PJ-CAS by one to three amino-acid residues and differed from one another. Two red flags were given to one haplotype (AF339461) in PJ-I, as the K2P distance between this and the other haplotypes in PJ-I ranged from 1.3 % to 3.5 % and the amino-acid sequence deduced from this haplotype differed from the PJ-CAS by three residues.

All remaining 27 haplotypes strongly diverged from PJ-I were given two red flags. Twenty five were previously presumed to be NUMTs (Chow et al. 2021,

Fig. 1. *Panulirus japonicus*. Neighbor-joining phylogenetic tree of 108 unique COI nucleotide sequences (526–816 bp) constructed with the pairwise deletion option using T92+G as the best fit model. *Panulirus longipes* was used as outgroup. The number carried by red flag is the number of amino-acid residue differences from the most Common Amino-acid Sequences (PJ-CAS) of this species. Haplotypes marked by an asterisk contain stop codon. Haplotypes determined as NUMTs in previous studies (Chow et al. 2021, 2024a) are shown in red. Bootstrap scores >70 % (out of 1,050 replicates) are shown at the nodes. Essentially the same tree topology was obtained with the complete deletion option. One red flag: haplotypes that differ by one or two amino-acid residues from the CAS and/or that show comparable or slightly larger K2P distances than the lowest threshold (2 % to 3 %) reported between congeneric animal species in general. Two red flags: haplotypes that differ by three or more amino-acid residues from the PJ-CAS and/or are separated from the majority of haplotypes by much larger K2P distances than the above threshold.



2024a) and are shown in red. Of these putative NUMTs, 17 haplotypes contained one or more stop codons, and the amino-acid sequences deduced from the eight others differed from the PJ-CAS by one to 12 amino-acid residues and also differed from one another. The K2P distance between these putative NUMTs and haplotypes in PJ-I ranged from 3.2 % to 25.3 %. Two red flags were also given to two strongly diverged haplotypes (OK037050, MZ203547) produced by Hettiarachchi et al. (2022). The deduced amino-acid sequences of these two haplotypes differed from the PJ-CAS by six and 22 amino-acid residues, respectively. The K2P distance between OK037050 and haplotypes in PJ-I ranged from 7.6 % to 11.0 %, and that between MZ203547 and haplotypes in PJ-I ranged from 15.4 % to 17.6 %. A BLAST search for OK037050 pointed to COI-gene sequences of *P. japonicus* but homology scores were lower than 95.0 %, and that for MZ203547 nominated putative NUMTs of *P. japonicus* with relatively high homology score (98.1 %). All these 27 haplotypes were obtained from cloned PCR amplicons (Chow et al. 2021, 2024a; Hettiarachchi et al. 2022), suggesting these were NUMTs or chimeric molecules. Hettiarachchi et al. (2022) unknowingly included these problematic sequences in their analysis, thereby inadvertently inflating intraspecific genetic diversity in this species.

### ***Panulirus longipes* group (Fig. 2)**

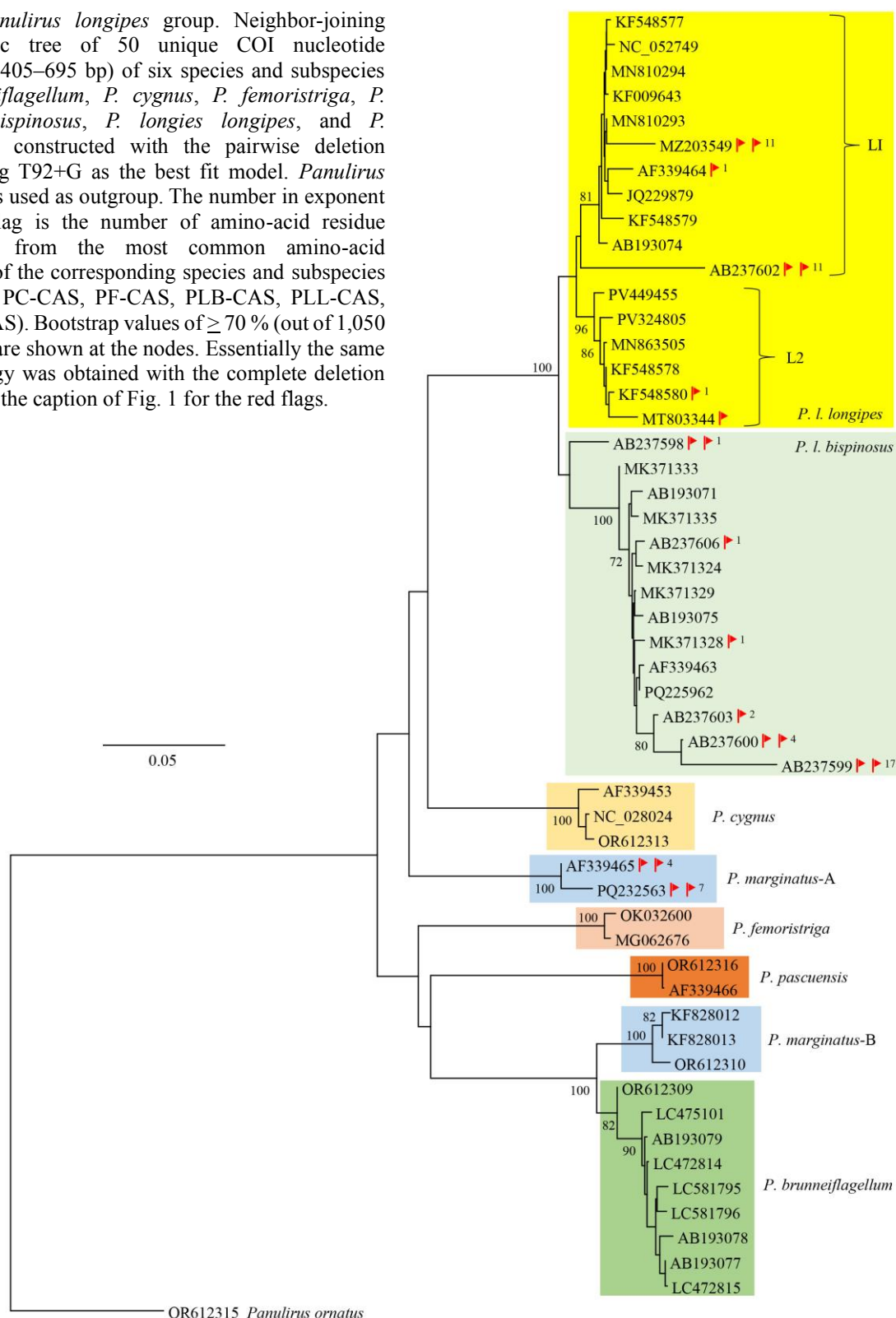
Six species (*P. brunneiflagellum*, *P. cygnus*, *P. femoristriga*, *P. longipes*, *P. marginatus*, and *P. pascuensis*) are now considered to belong to this group (Holthuis 1991; Chang and Ng 2001; Ravago and Juinio-Meñez 2003; Sekiguchi and George 2005). *Panulirus brunneiflagellum*, *P. cygnus*, *P. marginatus*, and *P. pascuensis* are endemic in the Ogasawara Islands, Western Australia, the Hawaiian Islands, and Easter Island, respectively, whereas *P. femoristriga* and *P. longipes* show Indo-Central Pacific wide distribution. *Panulirus longipes* is further subdivided into two subspecies, *P. l. longipes* and *P. l. bispinosus*

(Ravago and Juinio-Meñez 2003).

Of 70 sequences collected from GenBank, 51 unique haplotypes (with two to 31 haplotypes per species) were retained for phylogenetic analysis (Table S1).

Two main lineages were observed in *P. longipes*, corresponding to the two subspecies (*P. l. longipes* and *P. l. bispinosus*), and two sub-lineages (I and II) were observed within *P. l. longipes*. The K2P distance between haplotypes ranged from 0.2 % to 8.1 % in *P. l. longipes* and from 0.2 % to 9.5 % in *P. l. bispinosus*. The haplotypes with red flags appeared to be responsible for the inflated genetic diversity in both subspecies. The CAS of *P. l. longipes* (PLL-CAS) differed from that of *P. l. bispinosus* (PLB-CAS) by one residue. The deduced amino-acid sequences of two haplotypes in *P. l. longipes* (AF339464, KF548580) differed from the PLL-CAS by one residue and also differed from one another. Two red flags were given to two other haplotypes in *P. l. longipes* (AB237602, MZ203549), as these two differed from the PLL-CAS by 11 residues, and the K2P distance between these and the other haplotypes in *P. l. longipes* ranged from 2.0 % to 8.1 %. A BLAST search for these haplotypes nominated COI-gene sequences of *P. longipes* but homology scores were lower than 95 % for AB237602 and lower than 98 % for MZ203549. MZ203549 was obtained from cloned PCR amplicon (Hettiarachchi et al. 2022), suggesting this was a NUMT or chimeric molecule. Three haplotypes in *P. l. bispinosus* (AB237603, AB237606, MK371328) differed from the PLB-CAS by one or two residues and also differed from one another. Two red flags were given to three haplotypes (AB237598–AB237600) in *P. l. bispinosus*, as these differed from the PLB-CAS by one to 17 residues and the K2P distance between these and the other haplotypes in *P. l. bispinosus* ranged from 2.3 % to 6.2 %. A BLAST search for these haplotypes nominated COI-gene sequences of *P. longipes* but homology scores were lower than 98 %. These five haplotypes with two red flags were determined to be problematic sequences,

Fig. 2. *Panulirus longipes* group. Neighbor-joining phylogenetic tree of 50 unique COI nucleotide sequences (405–695 bp) of six species and subspecies (*P. brunneiflagellum*, *P. cygnus*, *P. femoristriga*, *P. longipes bispinosus*, *P. longipes longipes*, and *P. pascuensis*) constructed with the pairwise deletion option using T92+G as the best fit model. *Panulirus ornatus* was used as outgroup. The number in exponent to a red flag is the number of amino-acid residue differences from the most common amino-acid sequences of the corresponding species and subspecies (PBr-CAS, PC-CAS, PF-CAS, PLB-CAS, PLL-CAS, and PPa-CAS). Bootstrap values of  $\geq 70\%$  (out of 1,050 replicates) are shown at the nodes. Essentially the same tree topology was obtained with the complete deletion option. See the caption of Fig. 1 for the red flags.



possibly influenced by NUMTs.

Five haplotypes of *P. marginatus* were assigned to two distinct lineages (designated *P. marginatus-A* and

*P. marginatus-B*), differing by a K2P distance between 15.3 % and 17.2 %. The CAS of the *P. marginatus-B* lineage (PMaB-CAS) shared an amino-acid sequence

identical with PLB-CAS. As two haplotypes in the *P. marginatus*-A lineage differed from the PMaB-CAS by four and seven amino-acid residues, respectively, two red flags were given to these. Since no highly homologous sequence for *P. marginatus*-A haplotypes was nominated following a BLAST search, these two haplotypes were determined to be problematic sequences, possibly contaminated by NUMTs. Iacchei et al. (2014) reported slight genetic structuring in *P. marginatus*, but this was smaller than the divergence between the A and B lineages.

No haplotype was given two red flags in the other species of the *P. longipes* group.

### ***Panulirus penicillatus* (Fig. 3)**

*Panulirus penicillatus* is the only spiny lobster species distributed throughout the Indo-Pacific Ocean (Holthuis 1991), but the recent molecular analyses have revealed substantial genetic differentiation between Indo-central Pacific and Eastern Pacific populations (Chow et al. 2011; Abdulah et al. 2014a; Iacchei et al. 2016).

Of 468 sequences collected from GenBank, 277 unique haplotypes were retained for phylogenetic analysis (Table S1). Two main lineages (I and II) with a distinct sub-lineage (Ia) within lineage I were observed, corresponding to the Indo-West Pacific, the Eastern Pacific, and the Red Sea populations, respectively (see Chow et al. 2011; Abdulah et al. 2014a, b; Iacchei et al. 2016). The Indo-central and Eastern Pacific populations shared the same CAS (PPe-CAS). All five haplotypes (KT954881–KT9548885) from the Red Sea population shared the same amino-acid sequence (PPer-CAS) but differed from the PPe-CAS by one residue. The deduced amino-acid sequences of the nine haplotypes that were given one red flag in lineage I (excluding lineage Ia) differed from the PPe-CAS by one residue. Eight of these differed from one another. The deduced amino-acid sequences of the 15 haplotypes that were given one red flag in lineage II differed from the PPe-CAS

by one or two residues, which were assigned to four groups. The K2P distance ranged from 0.1 % to 2.9 % between haplotypes in lineage I (excluding lineage Ia) and from 0.1 % to 2.3 % between haplotypes in lineage II. One haplotype (MT750276) sampled from Indonesia was placed at a phylogenetically intermediate position between the two main lineages. The K2P distance between MT750276 and the haplotypes in lineages I and II ranged from 1.3 % to 2.9 %, and one red flag was given to this haplotype. Two red flags were given to two highly divergent haplotypes (KT954772, MT533488), which differed from the PPe-CAS by two and nine residues, respectively. The K2P distance between KT954772 and the haplotypes in lineages I and II ranged from 8.9 % to 10.4 %. A BLAST search for this haplotype returned the COI gene of *P. penicillatus* as the closest match, but the homology score was lower than 92 %. This haplotype was therefore determined to be a problematic sequence, possibly contaminated by NUMTs. The K2P distance between MT533488 and the haplotypes in lineages I and II ranged from 22.1 % to 26.3 %. A BLAST search for this haplotype returned a COI-gene sequence of *Panulirus inflatus* (OR612314) as the closest match with a 99 % homology score. On the other hand, the two other haplotypes of *Panulirus inflatus* in the database (AF339459, FJ174964) substantially diverged from MT533488 (13.6 to 14.0 % in K2P). Both Jeena et al. (2024) and Chow and Borsa (2024) questioned whether MT533488 was *P. penicillatus*.

### ***Panulirus homarus* (Fig. 4)**

This species is widely distributed in the Indo-West Pacific (Holthuis 1991). Based on the morphology, coloration, and geographic distribution, three subspecies (*P. h. homarus*, *P. h. megalculpta*, and *P. h. rubellus*) have been described (Berry 1974; Holthuis 1991; George 2006).

Of 483 sequences collected from GenBank, 169 unique haplotypes were retained for phylogenetic

Fig. 3. *Panulirus penicillatus*. Neighbor-joining phylogenetic tree of 277 unique COI nucleotide sequences (460–673 bp) constructed with the pairwise deletion option using TN93+G as the best fit model. *Panulirus japonicus* was used as outgroup. The number in exponent to a red flag is the number of amino-acid residue differences from the most common amino acid sequence of this species (PPe-CAS). Bootstrap scores of  $\geq 70\%$  (out of 1,050 replicates) are shown at the nodes. Essentially the same tree topology was obtained with the complete deletion option. See the caption of Fig. 1 for the red flags. Phylogenetic tree with accession numbers of all haplotypes used can be obtained [here](#).

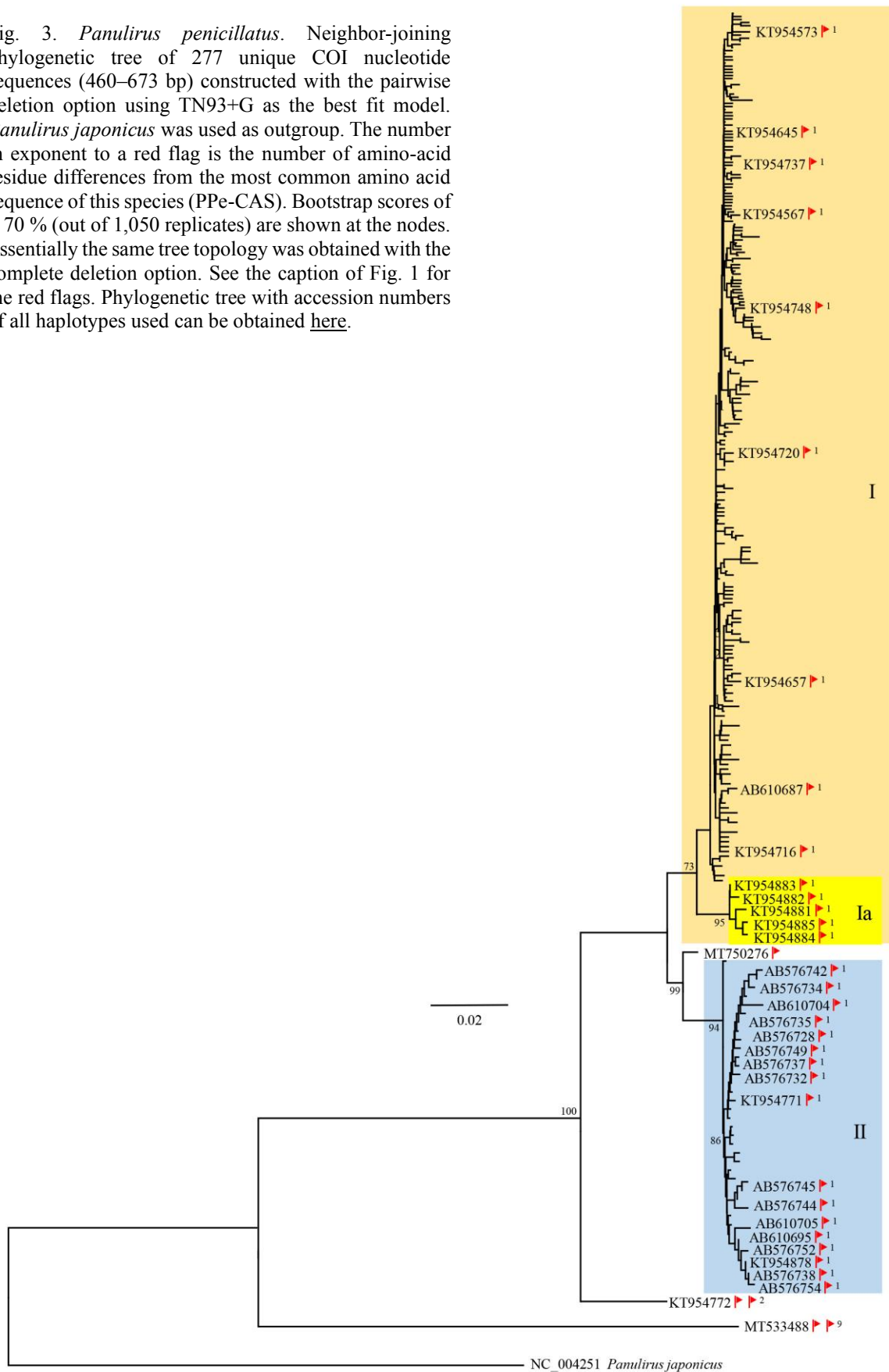
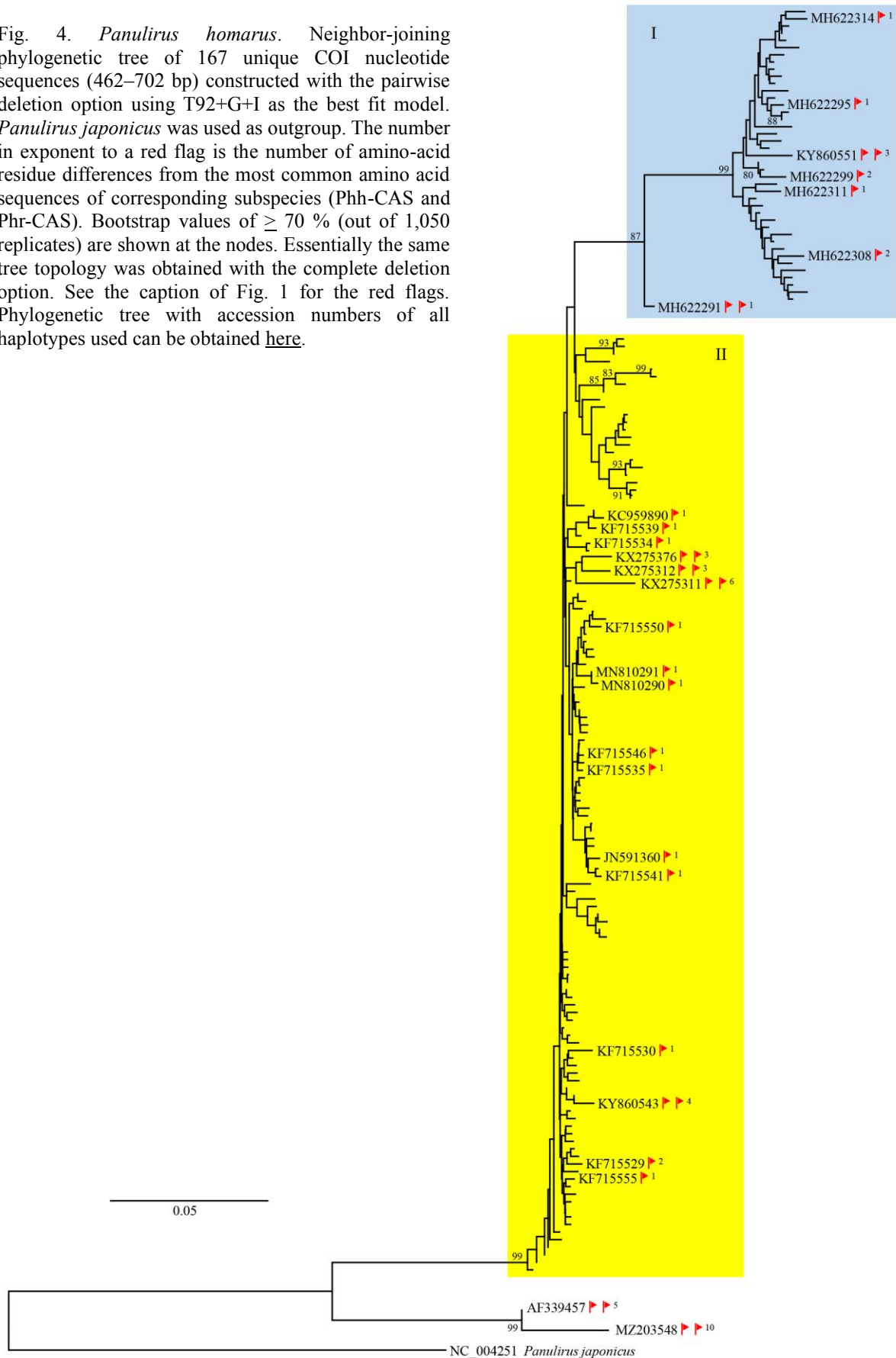


Fig. 4. *Panulirus homarus*. Neighbor-joining phylogenetic tree of 167 unique COI nucleotide sequences (462–702 bp) constructed with the pairwise deletion option using T92+G+I as the best fit model. *Panulirus japonicus* was used as outgroup. The number in exponent to a red flag is the number of amino-acid residue differences from the most common amino acid sequences of corresponding subspecies (Phh-CAS and Phr-CAS). Bootstrap values of  $\geq 70\%$  (out of 1,050 replicates) are shown at the nodes. Essentially the same tree topology was obtained with the complete deletion option. See the caption of Fig. 1 for the red flags. Phylogenetic tree with accession numbers of all haplotypes used can be obtained [here](#).



analysis (Table S1). COI-like sequences (KJ671893–KJ672073) reported by Reddy et al. (2014) possessing numerous indels and stop codons were not included in this study. Two main lineages (I and II) were observed. Lineage I represented *P. h. rubellus*. No segregation for the two other subspecies was observed within lineage II, as previously reported (Lavery et al. 2014). The CAS of *P. h. rubellus* (PHr-CAS) differed by one residue from that of *P. h. homarus* and *P. h. megasculpta* (PHh-CAS). Five haplotypes in lineage I were given one red flag as their deduced amino-acid sequences differed from PHr-CAS by one or two residues and also differed from one another. Two red flags were given to two haplotypes (KY860551, MH622291) in lineage I, which differed from PHr-CAS by three and one residues, respectively. The K2P distance ranged from 1.4 % to 3.6 % between KY860551 and the other haplotypes in lineage I, and from 3.7 % to 5.4 % between MH622291 and the other haplotypes in lineage I. A BLAST search for these haplotypes nominated COI-gene sequences of *P. homarus* as the best matches, but the homology scores were lower than 98.6 %. The deduced amino-acid sequences of 13 haplotypes in lineage II differed from the PHh-CAS by one or two residues and also differed from one another. One red flag was given to each of these 13 haplotypes. Two red flags were given to four haplotypes (KX275311, KX275312, KX275376, KY860543) in lineage II, which differed from PHh-CAS by three to six residues. A BLAST search for these four haplotypes nominated COI-gene sequences of *P. homarus* as the best matches, with homology scores lower than 99.1 %. These four haplotypes with two red flags were determined to be problematic sequences, possibly contaminated by NUMTs. The deduced amino-acid sequences for two divergent haplotypes (MZ203548, AF339457) differed from the PHr-CAS and PHh-CAS by five and 10 residues, respectively. The K2P distance between these two haplotypes and all the other haplotypes in the lineages I and II ranged from 10.5 % to 16.1 % and from 12.8 %

to 18.6 %, respectively. A BLAST search for these two haplotypes detected no highly homologous sequence. These two haplotypes were determined to be problematic sequences. MZ203548 was obtained from a cloned PCR amplicon (Hettiarachchi et al. 2022), suggesting this was a NUMT or a chimeric molecule, whereas AF339457 obtained by direct nucleotide sequencing is possibly contaminated by NUMTs. Lavery et al. (2014) suspected AF339457 to be a NUMT.

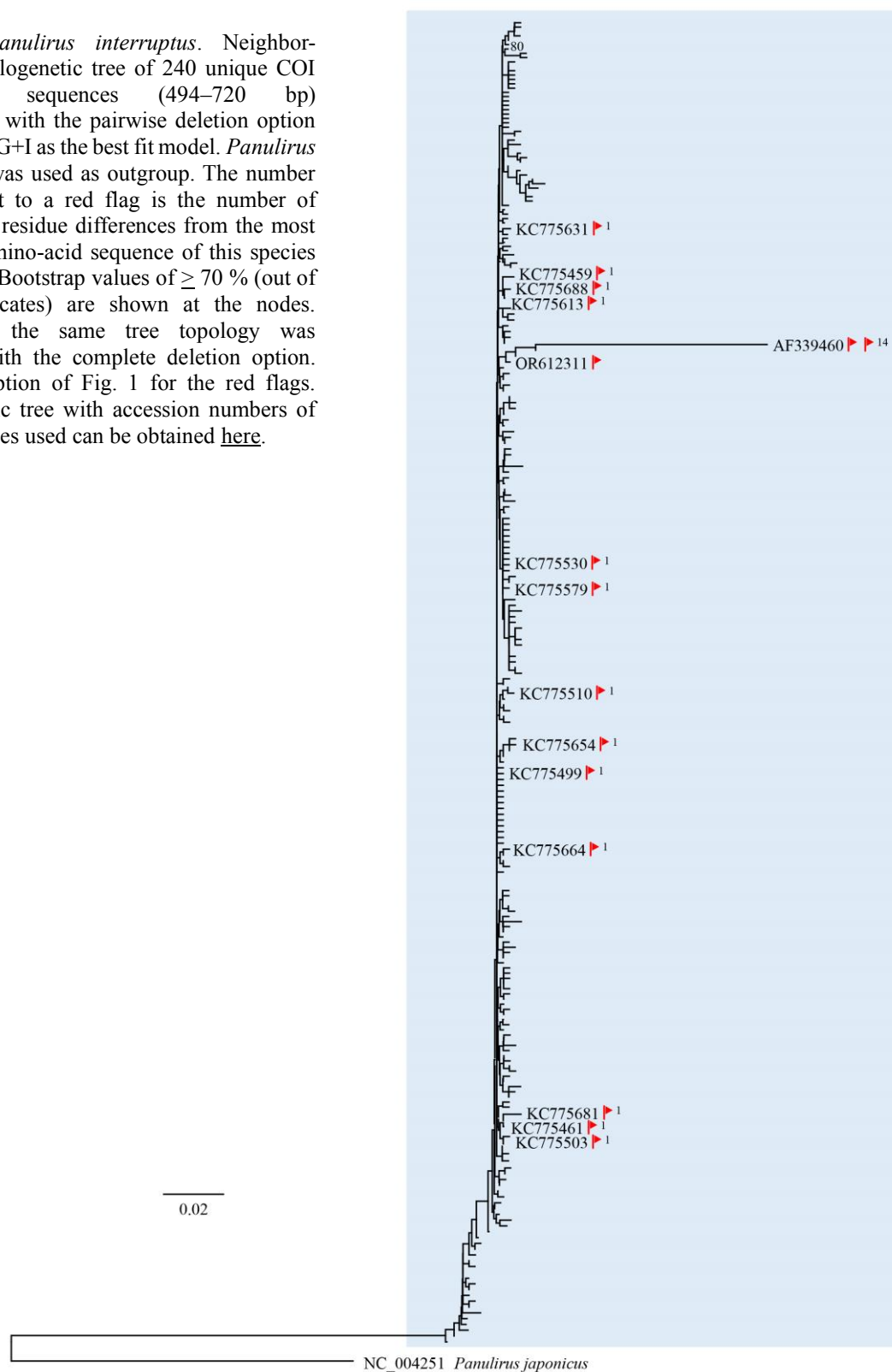
#### ***Panulirus ornatus* and *P. versicolor* (Fig. 5)**

These species are distributed in the Indo-West Pacific region (Holthuis 1991). Molecular analyses have revealed significant population structuring in *P. ornatus* (Yellapu et al. 2017; Farhadi et al. 2022).

Of 105 sequences collected from GenBank, 63 unique haplotypes (43 for *P. ornatus* and 20 for *P. versicolor*) were retained for phylogenetic analysis (Table S1). The CAS of *P. ornatus* (PO-CAS) differed by two residues from that of *P. versicolor* (PV-CAS). One haplotype in *P. ornatus* (MN810265), which differed from the PO-CAS by one residue, was given one red flag. Two red flags were given to two haplotypes in *P. ornatus* (AF339467, KF827964), which differed from the PO-CAS by four and two residues, respectively. The K2P distance between AF339467 and the other haplotypes of *P. ornatus* ranged from 5.7 % to 7.2 %. A BLAST search for this haplotype nominated COI-gene sequences of *P. ornatus* as the most similar, but homology scores were lower than 95 %. Therefore, this haplotype was determined to be a problematic sequence. The K2P distance between KF827964 and the other haplotypes in *P. ornatus* ranged from 23.5 % to 25.5 %. No highly homologous sequence for KF827964 was retrieved from the BLAST search. This haplotype was determined to be a problematic sequence, possibly contaminated by NUMTs. Ng et al. (2024) suspected AF339467 to be a NUMT. No haplotype with red flag was observed in *P. versicolor*.



Fig. 6. *Panulirus interruptus*. Neighbor-joining phylogenetic tree of 240 unique COI nucleotide sequences (494–720 bp) constructed with the pairwise deletion option using T92+G+I as the best fit model. *Panulirus japonicus* was used as outgroup. The number in exponent to a red flag is the number of amino-acid residue differences from the most common amino-acid sequence of this species (PIt-CAS). Bootstrap values of  $\geq 70\%$  (out of 1,050 replicates) are shown at the nodes. Essentially the same tree topology was obtained with the complete deletion option. See the caption of Fig. 1 for the red flags. Phylogenetic tree with accession numbers of all haplotypes used can be obtained [here](#).



### *Panulirus interruptus* (Fig. 6)

This species is confined to subtropical to temperate regions in the Eastern North Pacific, from Monterey

Bay to Baja California (Holthuis 1991). No apparent genetic structuring has been reported throughout its range (Iacchei et al. 2013).

Of 245 sequences collected from GenBank, 240 unique haplotypes were retained for phylogenetic analysis (Table S1). Of 240 haplotypes, 225 haplotypes shared the same deduced amino-acid sequence (designated as PIt-CAS). The amino-acid sequences of 14 haplotypes differed from PIt-CAS by one residue and also differed from one another. Two red flags were given to one haplotype (AF339460), which had two 12-nucleotide indels and differed from the PIt-CAS by 17 amino-acid residues. The K2P distance between AF339460 and the other haplotypes ranged from 6.5 % to 9.7 %. These values are likely underestimated due to the presence of indels. A BLAST search for this haplotype nominated COI-gene sequences of *P. interruptus* as the best matches, but the homology scores were lower than 95 %. This haplotype was determined to be a problematic sequence, possibly contaminated by NUMTs.

#### ***Panulirus gracilis*, *P. inflatus*, *P. polyphagus*, and *P. stimpsoni* (Fig. 7)**

*Panulirus gracilis* and *P. inflatus* are distributed in the tropical to subtropical region of the Eastern Pacific, whereas *P. polyphagus* and *P. stimpsoni* are distributed in the tropical to subtropical regions of the Indo-West Pacific (Holthuis 1991).

Of 127 sequences collected from GenBank, 74 unique haplotypes (two to 62 per species) were retained for phylogenetic analysis (Table S1).

Two major lineages (L1 and L2) were observed in *P. polyphagus*, as determined by Jeena et al. (2024). The respective CAS of these two lineages were identical and designated as PPo-CAS. The deduced amino-acid sequences of two haplotypes (JN418939, MW514206) differed from the PPo-CAS by one residue and also differed from each other. These two haplotypes were given one red flag. The placement of two other haplotypes (LC536749, MW514205) was external to the two lineages. The deduced amino-acid sequence of LC536749 differed from the PPo-CAS by two residues, and the K2P distance between LC536749 and the other

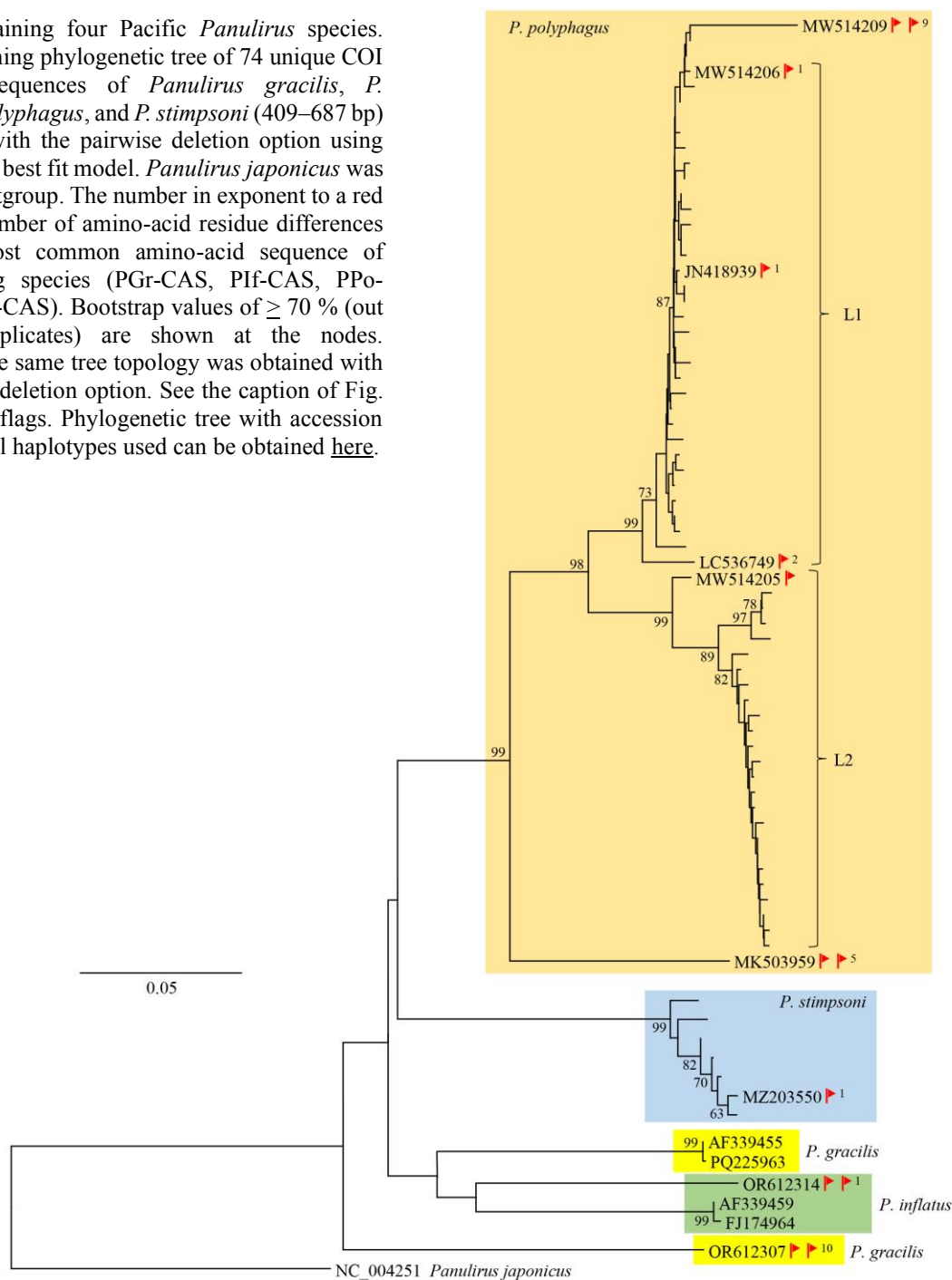
haplotypes in lineage L1 ranged from 2.3 % to 3.1 %. The deduced amino-acid sequence of MW514205 was identical to the PPo-CAS and the K2P distance between MW514205 and the other haplotypes in lineage L2 ranged from 2.2 % to 3.8 %. The amino-acid sequences of two other highly divergent haplotypes (MW514209, MK503959) differed from the PPo-CAS by nine and five residues, respectively. The K2P distance between these two divergent haplotypes and the other haplotypes in the L1 and L2 lineages ranged from 3.3 % to 11.7 %. A BLAST search for MW514209 and MK503959 nominated COI-gene sequences of *P. polyphagus* as the best matches, but the homology scores were lower than 97 % and 91 %, respectively. These two haplotypes were determined to be problematic sequences, possibly contaminated by NUMTs.

No haplotype in *P. stimpsoni* was given two red flags.

Three haplotypes of *P. gracilis* were assigned to two distinct lineages. The deduced amino-acid sequences of two haplotypes (AF339455, PQ225963) were identical and designated as PGr-CAS. The PGr-CAS differed from the amino-acid sequence of OR612307 by 10 residues and two deletions. The K2P distance between OR612307 and the other two haplotypes ranged from 19.7 % to 19.8 %. A BLAST search for OR612307 nominated a haplotype of *P. ornatus* (KF827964) with a relatively high homology score (98.5 %), but this haplotype of *P. ornatus* was determined to possess a problematic sequence (see Fig. 5). The K2P distance between OR612307 and the other *P. ornatus* haplotypes ranged from 20.8 % to 23.9 %. Therefore, we determined OR612307 to be a problematic sequence, possibly contaminated by NUMTs.

Three haplotypes of *P. inflatus* were assigned to two highly divergent lineages. The deduced amino-acid sequences of two haplotypes (AF339459, FJ174964) were identical and designated as PIf-CAS. The deduced amino-acid sequence of OR612314 differed

Fig. 7. Remaining four Pacific *Panulirus* species. Neighbor-joining phylogenetic tree of 74 unique COI nucleotide sequences of *Panulirus gracilis*, *P. inflatus*, *P. polyphagus*, and *P. stimpsoni* (409–687 bp) constructed with the pairwise deletion option using T92+G as the best fit model. *Panulirus japonicus* was used as an outgroup. The number in exponent to a red flag is the number of amino-acid residue differences from the most common amino-acid sequence of corresponding species (PGr-CAS, PIf-CAS, PPo-CAS, and PS-CAS). Bootstrap values of  $\geq 70\%$  (out of 1,050 replicates) are shown at the nodes. Essentially the same tree topology was obtained with the complete deletion option. See the caption of Fig. 1 for the red flags. Phylogenetic tree with accession numbers of all haplotypes used can be obtained [here](#).



from the PIf-CAS by one residue. The K2P distance between OR612314 and the other two haplotypes ranged from 13.6 % to 14.0 %. A BLAST search for OR612314 nominated a haplotype of *P. penicillatus* (MT533488) with a high homology score (99.1 %), but this haplotype (MT533488) of *P. penicillatus* was determined to be a problematic sequence (see Fig. 3). Therefore, we presumed OR612314 as well as

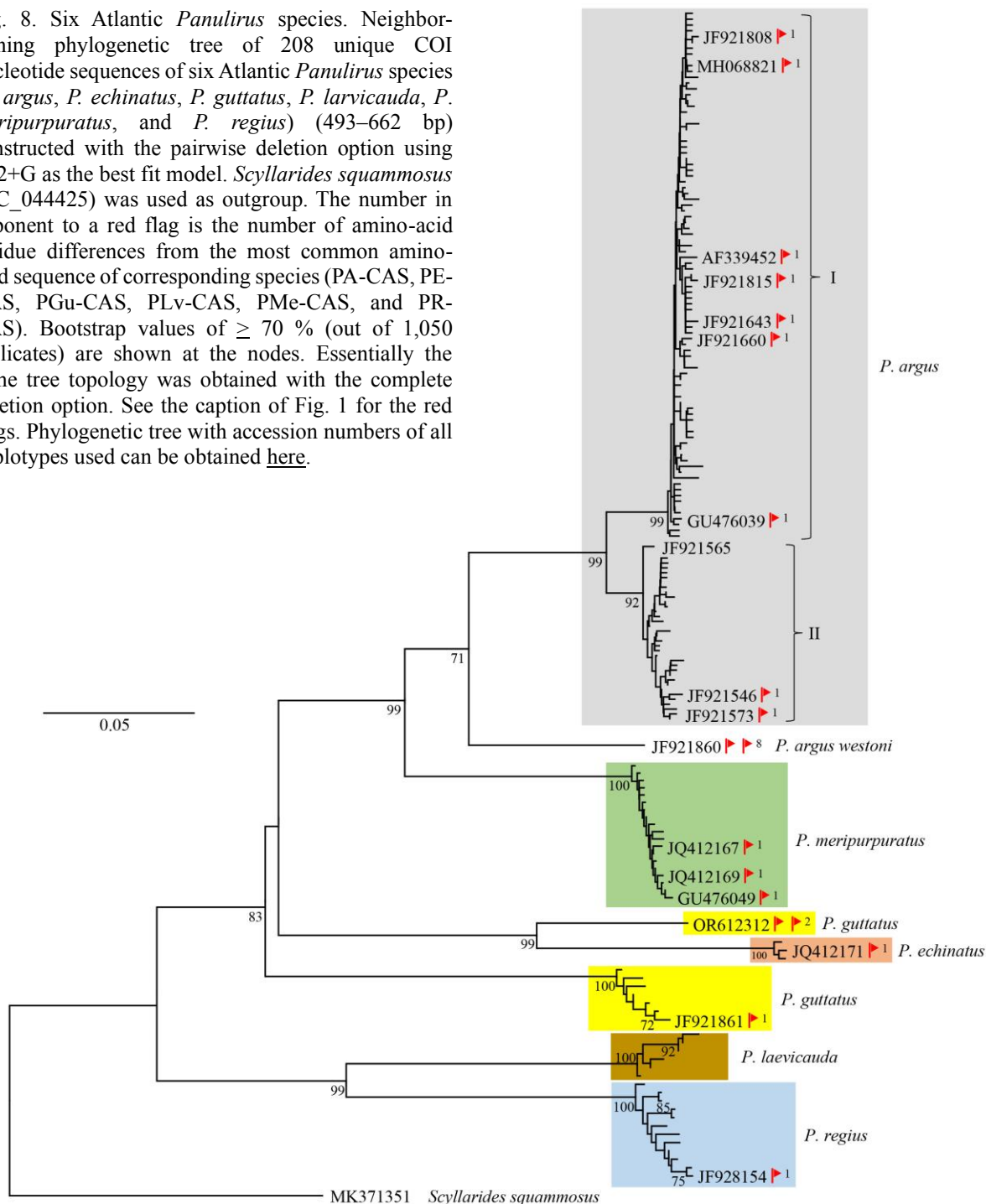
MT533488 of *P. penicillatus* to be possibly contaminated by NUMTs.

#### Atlantic species

*Panulirus argus*, *P. echinatus*, *P. guttatus*, *P. laevicauda*, *P. meripurpuratus*, and *P. regius* (Fig. 8)

Of six *Panulirus* species described in the Atlantic, only one species, *P. regius*, is distributed in the coast

Fig. 8. Six Atlantic *Panulirus* species. Neighbor-joining phylogenetic tree of 208 unique COI nucleotide sequences of six Atlantic *Panulirus* species (*P. argus*, *P. echinatus*, *P. guttatus*, *P. larvicauda*, *P. meripurpuratus*, and *P. regius*) (493–662 bp) constructed with the pairwise deletion option using T92+G as the best fit model. *Scyllarides squammosus* (NC\_044425) was used as outgroup. The number in exponent to a red flag is the number of amino-acid residue differences from the most common amino-acid sequence of corresponding species (PA-CAS, PE-CAS, PGu-CAS, PLV-CAS, PME-CAS, and PR-CAS). Bootstrap values of  $\geq 70\%$  (out of 1,050 replicates) are shown at the nodes. Essentially the same tree topology was obtained with the complete deletion option. See the caption of Fig. 1 for the red flags. Phylogenetic tree with accession numbers of all haplotypes used can be obtained [here](#).



of Africa including the Mediterranean Sea (Holthuis 1991).

Of 405 sequences collected from GenBank, 175 unique haplotypes (three to 127 per species) were retained for phylogenetic analysis (Table S1).

Sarver et al. (1998, 2000) found substantial genetic differentiation between Caribbean and Brazilian

samples of *P. argus* and proposed two subspecies (*P. argus argus* for the Caribbean form and *P. argus westoni* for the Brazilian form). This finding was further corroborated by Diniz et al. (2005) and Naro-Maciel et al. (2011), and finally Giraldes and Smyth (2016) described the Brazilian form as *P. meripurpuratus*. Two lineages (I and II) were observed

in *P. argus*, corresponding to the previous genetic studies (Silberman et al. 1994; Diniz et al. 2005; Naro-Maciel et al. 2011; Tourinho et al. 2012). Despite substantial nucleotide sequence divergence, the CAS of *P. argus* and *P. meripurpuratus* (designated as PA-CAS and PMe-CAS, respectively) were identical. The deduced amino-acid sequences of nine haplotypes in *P. argus* differed from the PA-CAS by one residue, of which eight differed from one another. Deduced amino-acid sequences of three haplotypes in *P. meripurpuratus* differed from the PMe-CAS by one residue and also differed from one another. Naro-Maciel et al. (2011) determined a COI-gene sequence of *P. argus westoni* provided by J. D. Silberman, but this sequence (JF921860) considerably differed from both *P. argus* (average K2P distance:  $11.6 \pm 1.5$  %) and *P. meripurpuratus* (average K2P distance:  $12.0 \pm 1.73$  %). Because the deduced amino-acid sequence of JF921860 differed from PA-CAS by eight residues, it was determined that JF921860 was a problematic sequence possibly contaminated by NUMTs.

Of eight haplotypes labelled *P. guttatus*, seven were closely clustered together. Six of these seven haplotypes shared the same amino-acid sequence, which was designated as PGu-CAS. The deduced amino-acid sequence of a highly divergent haplotype (OR612312) differed from the PGu-CAS by two residues. The K2P distance between the OR612312 and the other haplotypes of *P. guttatus* ranged from 22.7 % to 24.5 %. All sequences nominated for OR612312 by a BLAST search were of *P. penicillatus*, in which sequences showing over 99 % homology score were COI-gene sequences of *P. penicillatus* from the Eastern Pacific (lineage II in Fig. 3). This result indicates that OR612312 is *P. penicillatus* and not *P. guttatus*.

No haplotype with two red flags was observed in the other species.

#### Amino-acid sequence divergence between species

The CAS from all species and subspecies were

aligned (Fig. 9), and the amino-acid residue differences between them are presented in Table 1. Species were assigned to four morphological groups (I to IV). Previous molecular phylogenetic analyses using nucleotide sequences of mtDNA have supported two main groups (I+II and III+IV), but the relationships among species within each main group were not well resolved (Ptacek et al. 2001; Chow et al. 2006; Davis et al. 2015). The number of different residues between CAS ranged from 0 to four (0.0 % to 1.8 % in p-distance) in the I+II group and from 0 to 11 (0.0 % to 5.2 % in p-distance) in the III+IV group, while that between the I+II and III+IV groups ranged from seven to 20 (3.5 % to 10.0 % in p-distance). The phylogenetic tree based on the p-distance (Fig. 10) was consistent with the results of the previous molecular studies but showed a considerably divergent status for the CAS of *P. gracilis* (PGr-CAS). The number of residue differences between the PGr-CAS and the I+II group ranged from 15 to 20 (9.0 % to 10.0 % in p-distance), and those between the PGr-CAS and the III+IV group ranged from nine to 11 (4.7 % to 5.2 % in p-distance). Furthermore, the deletion of two residues was observed only in the PGr-CAS (Fig. 9). Indels are very rare in the COI gene in most animal groups (Hebert et al. 2003b). The number of residue differences between the *Panulirus* CAS, except for PGr-CAS, and the amino-acid sequence of the other family member *Scyllarides squammosus*, which was used as outgroup, ranged from seven to 14 (3.0 to 7.2 % in p-distance). These differences were comparable to, or even smaller than those between the PGr-CAS and the other species of the genus *Panulirus*. These observations strongly suggest that the two haplotypes (AF339455, PQ225963) of *P. gracilis* are problematic, possibly contaminated by NUMTs. Excluding the PGr-CAS, the number of residue differences between species in the III+IV group ranged from 0 to three (0.0 % to 1.4 % in p-distance) and that between the I+II and III+IV groups ranged from seven to 11 (3.5 % to 6.6 % in p-distance).



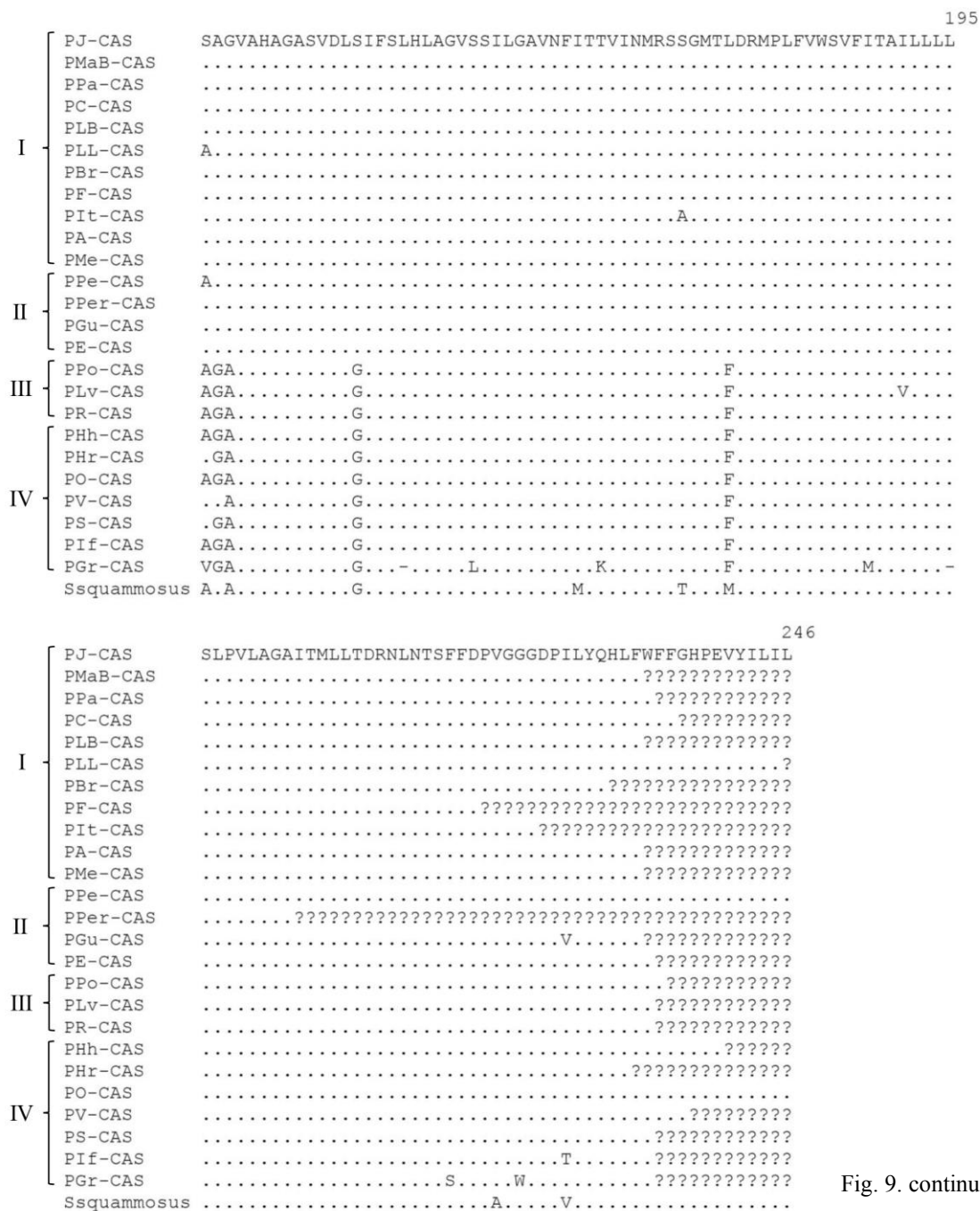


Fig. 9. continued.

reconstruction, nucleotide sequence analysis, and deduced amino-acid sequence analysis is useful for identifying questionable COI-gene sequences in *Panulirus* spiny lobsters. However, it should be noted that this approach cannot distinguish between young NUMTs, sequencing errors, and heteroplasmy.

As the cytochrome c oxidase is involved in the cell respiration process and the COI barcode region encodes a core region of this enzyme, any mutation

affecting its function or its structure may be lethal (Pentinsaari et al. 2016). Since evolutionary plasticity is known to be greater in loop regions than in core regions of proteins (Panchenko et al. 2005), it is necessary to further investigate nonsynonymous nucleotide substitutions in relation to region. Nevertheless, it is possible that COI subunits with different amino-acid sequences may not be able to coexist within a species, suggesting that nonsynony-

Table 1. Number of amino-acid residue differences (below diagonal) and percent p-distance (upper diagonal) between the most common amino-acid sequences (CAS) of *Panulirus* species and subspecies. Deduced amino-acid sequence of *Scyllarides squammosus* (NC\_044425) was used as out group.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	
I 1. PJ-CAS		0.0	0.0	0.0	0.0	0.4	0.0	0.0	0.9	0.0	0.0	0.5	0.0	0.5	0.9	4.4	5.0	4.7	4.4	4.8	4.5	3.5	4.1	5.1	9.0	6.0	
I 2. PMaB-CAS	0		0.0	0.0	0.0	0.4	0.0	0.0	0.9	0.0	0.0	0.5	0.0	0.5	0.9	4.5	5.0	4.7	4.5	4.8	4.5	3.6	4.1	5.2	9.0	6.3	
I 3. PPa-CAS	0	0		0.0	0.0	0.5	0.0	0.0	1.0	0.0	0.0	0.5	0.0	0.5	0.9	4.7	5.1	4.7	4.7	4.8	4.7	3.7	4.2	5.1	9.0	6.5	
I 4. PC-CAS	0	0	0		0.0	0.4	0.0	0.0	0.9	0.0	0.0	0.5	0.0	0.5	0.9	4.4	5.0	4.7	4.4	4.8	4.5	3.5	4.1	5.1	9.0	6.2	
I 5. PLB-CAS	0	0	0	0		0.5	0.0	0.0	1.0	0.0	0.0	0.5	0.0	0.5	0.9	4.6	5.0	4.7	4.6	4.8	4.6	3.7	4.1	5.2	9.0	6.4	
I 6. PLL-CAS	1	1	1	1	1		0.5	0.5	1.4	0.5	0.5	0.0	0.7	0.9	1.3	4.0	4.5	4.2	3.9	5.3	3.9	3.9	4.5	4.7	9.0	5.5	
I 7. PBr-CAS	0	0	0	0	0	1		0.0	0.9	0.0	0.0	0.5	0.0	0.5	0.9	4.5	5.1	4.8	4.5	4.8	4.6	3.6	4.2	5.2	9.1	6.1	
I 8. PF-CAS	0	0	0	0	0	1	0		1.0	0.0	0.0	0.5	0.0	0.0	1.0	5.1	5.6	5.1	5.1	5.1	5.1	4.1	4.6	5.1	9.3	6.2	
I 9. PIt-CAS	2	2	2	2	2	3	2	2		1.0	1.0	1.5	1.3	1.0	1.8	4.6	5.2	4.9	4.6	5.0	4.7	3.7	4.3	4.9	9.4	5.4	
I 10. PA-CAS	0	0	0	0	0	1	0	0	2		0.0	0.5	0.0	0.5	0.9	4.6	5.0	4.7	4.6	4.8	4.6	3.7	4.1	5.2	9.0	6.4	
I 11. PMe-CAS	0	0	0	0	0	1	0	0	2	0		0.5	0.0	0.5	0.9	4.6	5.0	4.7	4.6	4.8	4.6	3.7	4.1	5.2	9.0	6.4	
II 12. PPe-CAS	1	1	1	1	1	0	1	1	3	1	1		0.7	1.0	1.4	4.3	4.8	4.3	4.2	5.3	4.1	4.2	4.8	4.8	9.1	5.9	
II 13. PPer-CAS	0	0	0	0	0	1	0	0	2	0	0	1		0.0	0.7	5.9	6.6	5.9	5.9	5.3	5.9	4.6	5.3	5.9	10.0	7.2	
II 14. PGu-CAS	1	1	1	1	1	2	1	0	2	1	1	2	0		1.4	5.0	5.5	5.2	5.0	5.3	5.0	4.1	4.6	5.2	9.5	5.9	
II 15. PE-CAS	2	2	2	2	2	3	2	2	4	2	3	1	3		4.4	5.0	4.7	4.4	4.3	4.5	4.5	3.5	4.1	5.1	9.0	6.0	
III 16. PPO-CAS	10	10	10	10	10	9	10	10	10	10	10	9	9	11	10		0.5	0.0	0.0	0.5	0.0	0.9	0.5	0.5	4.7	3.1	
III 17. PLv-CAS	11	11	11	11	11	10	11	11	11	11	11	10	10	12	11	1		0.5	0.5	1.1	0.5	1.4	0.9	0.9	5.2	3.6	
III 18. PR-CAS	10	10	10	10	10	9	10	10	10	10	10	9	9	11	10	0	1		0.0	0.5	0.0	0.9	0.5	0.5	4.7	3.3	
IV 19. PHh-CAS	10	10	10	10	10	9	10	10	10	10	10	9	9	11	10	0	1	0		0.5	0.0	0.9	0.5	0.5	4.7	3.0	
IV 20. PHr-CAS	9	9	9	9	9	10	9	9	9	9	9	10	8	10	8	1	2	1	1		0.5	0.5	0.0	1.1	4.8	4.3	
IV 21. PO-CAS	10	10	10	10	10	9	10	10	10	10	10	9	9	11	10	0	1	0	0	1		0.9	0.5	0.5	4.7	3.0	
IV 22. PV-CAS	8	8	8	8	8	9	8	8	8	8	8	9	7	9	8	2	3	2	2	1	2		0.5	1.4	5.2	3.0	
IV 23. PS-CAS	9	9	9	9	9	10	9	9	9	9	9	10	8	10	9	1	2	1	1	0	1	1		0.9	4.7	3.6	
IV 24. PIf-CAS	11	11	11	11	11	10	11	10	10	11	11	10	9	11	11	1	2	1	1	2	1	3	2		5.2	3.3	
IV 25. PGr-CAS	19	19	19	19	19	19	19	18	19	19	19	19	15	20	19	10	11	10	10	9	10	11	10	11		8.0	
26. Ssqua	14	14	14	14	14	13	14	12	12	14	14	13	11	13	14	7	8	7	7	8	7	7	8	7	8	7	17

I to IV: morphological groups determined by George and Main (1967). PJ: *Panulirus japonicus*, PMaB: *P. marginatus*-B, PPa: *P. pascuensis*, PC: *P. cygnus*, PLB: *P. longipes bispinosus*, PLL: *P. longipes longipes*, PBr: *P. brunneiflagellum*, PF: *P. femoristriga*, PIt: *P. interruptus*, PA: *P. argus*, PMe: *P. meripurpuratus*, PPe: *P. penicillatus*, PPer: *P. penicillatus* Red Sea population, PGu: *P. guttatus*, PE: *P. echinatus*, PPO: *P. polyphagus*, PLv: *P. laevicauda*, PR: *P. regius*, PHh: *P. homarus homarus*, PHr: *P. homarus rubellus*, PO: *P. ornatus*, PV: *P. versicolor*, PS: *P. stimpsoni*, PIf: *P. inflatus*, PGr: *P. gracilis*, Ssqua: *Scyllarides squammosus*.

mous nucleotide substitutions observed between individuals of the same species are possibly to be caused by either sequencing errors or NUMT contamination.

In *Panulirus* lobsters, the among-species maximum number of amino-acid residue differences in the Folmer region was four (1.8 % in p-distance) in the I+II group, and three (1.4 % in p-distance) in the III+IV group. This supports the proposed threshold at which haplotypes differing by three or more amino-acid residues from the CAS are given two red flags. This also indicates that the haplotypes for which the number of amino-acid residue differences with the CAS is much larger are not of closely-related cryptic species but possibly contaminated by NUMTs. Haplotypes given two red flags and therefore determined as problematic sequences are listed in

Table 2. These problematic sequences should not be used for phylogenetic analysis or for species identification. Haplotypes based on specimens with incorrect species identification should be corrected or deleted by the submitter. Haplotypes with one red flag are not listed here, but these should be used with caution.

Buhay (2009) observed that a number of COI-gene sequences of decapod crustaceans deposited in GenBank contained stop codons or indels. She suggested these to be due to either sequence editing errors or NUMTs. Furthermore, based on the occurrence of stop codons, Woodling et al. (2019) detected potential NUMTs in up to 35 % of the phyllosoma larvae of spiny and slipper lobsters. Problematic sequences presumed NUMTs or to be NUMT-contaminated sequences were also extensively

observed in the present study. Thus, achelate lobsters may be particularly prone to NUMT occurrence. The relatively large genome size of large achelate lobsters (3.0 to 5.5 picograms) (Jimines et al. 2010; Veldsman et al. 2021; Baeza and Pirro 2024) comparable to, or even larger than the human one (3.2 picograms) (Piovesan et al. 2022) might provide an explanation to this phenomenon.

The unnoticed inclusion of NUMTs or of sequences

contaminated with NUMTs may inflate intraspecific genetic diversity and distort phylogenies as observed in, e. g., Hettiarachchi et al. (2022). As the adverse consequences of artefactual sequence data and biased evolutionary analyses cannot be considered negligible, authors, reviewers, and editors should be aware of the risk of NUMT occurrence in nucleotide sequence datasets.



Fig. 10. Neighbor-joining phylogenetic tree based on p-distance with the pairwise deletion option between the most common amino-acid sequences (CAS) (152 to 234 residues) of *Panulirus* species and subspecies shown in Fig. 9. *Scyllarides squammosus* was used as outgroup. Accession numbers are shown in the parenthesis. Bootstrap values of  $\geq 50\%$  (out of 1,050 replicates) are shown at the nodes. I–IV: morphological groups determined by George and Main (1967). Similar tree topologies were obtained using number of different amino-acid residues and with the complete deletion option.

Table 2. List of problematic COI haplotypes of spiny lobsters of the genus *Panulirus* deposited in GenBank.

	Highly problematic	Problematic	misidentification of species
<i>P. argus</i>	JF921860		
<i>P. gracilis</i>	AF339455, PQ225963, OR612307*		
<i>P. guttatus</i>			OR612312* (= <i>P. penicillatus</i> )
<i>P. homarus homarus</i>	AF339457, KX275311, MZ203548	KX275312, KX275376	
<i>P. homarus megasculpta</i>	KY860543		
<i>P. homarus rubellus</i>	MH622291	KY860551	
<i>P. inflatus</i>	OR612314*		
<i>P. interruptus</i>	AF339460		
<i>P. japonicus</i>	LC571565–LC571570, LC571572– LC571577, LC654683–LC654687, LC782283–LC782290, MZ203547, OK037050	AF339461	
<i>P. longipes bispinosus</i>	AB237599, AB237600	AB237598	
<i>P. longipes longipes</i>	AB237602, MZ203549		
<i>P. marginatus</i>	AF339465, PQ232563		
<i>P. ornatus</i>	AF339467, KF827964		
<i>P. penicillatus</i>	KT954772, MT533488*		
<i>P. polyphagus</i>	MK503959*, MW514209		

\*whole mtDNA sequence.

### Acknowledgments

We are grateful to three anonymous reviewers for their valuable comments and suggestions.

### References

- Abdulah, M. F., Chow, S., Sakai, M. Cheng, J-H., Imai, H. (2014a). Genetic diversity and population structure of pronghorn spiny lobster *Panulirus penicillatus* in the Pacific region. *Pac. Sci.* 68: 197–211.
- Abdulah, M. F., Alimunddin, Muththalib, M., Salama, A. J., Imai, H. (2014b). Genetic isolation among the northwestern, southwestern and central-eastern Indian Ocean populations of the pronghorn spiny lobster *Panulirus penicillatus*. *Int. J. Mol. Sci.* 15: 9242–9254.
- Andriyono, S., Alam, M. J., Pramono, H., Abdillah, A. A., Kim, H. W. (2019). Next-generation sequencing yields the complete mitochondrial genome of mud spiny lobster, *Panulirus polyphagus* (Crustacea: Decapoda) from Madura water. *IOP Conf. Ser.: Earth Environ. Sci.* 348: 012020.
- Baeza, J. A., Fuentes, M. S. (2013). Exploring phylogenetic informativeness and nuclear copies of mitochondrial DNA (numts) in three commonly used mitochondrial genes: mitochondrial phylogeny of peppermint, cleaner, and semi-terrestrial shrimps (Caridea: Lysmata, Exhippolysmata, and Merguia). *Zoo. J. Linn. Soc.* 168: 699–722.
- Baeza, J. A. (2018). The complete mitochondrial genome of the Caribbean spiny lobster *Panulirus argus*. *Sci. Rep.* 8: 17690.
- Baeza, J. A., Baker, A., Childress, M., Pirro, S. (2024). Nuclear and mitochondrial genome datasets for spiny lobsters genus *Panulirus* (Decapoda: Achelata: Palinuridae). *Data Brief* 55: 110588.
- Baeza, J. A., Pirro, S. (2024). Genomics resources for the Rapa Nui (Eastern Island) spiny lobster *Panulirus pascuensis* (Crustacea: Decapoda: Achelata). *Rev. Chil. Hist. Nat.* 97: 9.
- Bensasson, D., Zhang, D-X., Hartl, D. L., Hewitt, G. M. (2001). Mitochondrial pseudogenes: evolution's misplaced witnesses. *TREE* 16: 314–321.
- Benson, D. A., Cavanaugh, M., Clark, K., Karsch-Mizrachi, I., Lipman, D. J., Ostell, J., Sayers, E.W. (2017). GenBank. *Nucl. Acids Res.* 45: D37–D42.
- Berry, P. F. (1974). A revision of the *Panulirus homarus*-group of spiny lobsters (Decapoda, Palinuridae). *Crustaceana* 27: 32–42.
- Bracken-Grissom, H. D., Ah Yong, S. T., Wilkinson, R. D., Feldmann, R. M., Schweitzer, C. E., Breinholt, J. W., Bendall, M., Palero, F., Chan, T.Y., Felder, D. L., Robles, R., Chu, K. H., Tsang, L. M., Kim, D., Martin, J. W., Crandall, K. A. (2014). The emergence of lobsters: phylogenetic relationships, morphological evolution and divergence time comparisons of an ancient group

- (Decapoda: Achelata, Astacidea, Glypheidea, Polychelida). *Syst. Biol.* 63: 457–479.
- Buhay, J. E. (2009). “COI-like” sequences are becoming problematic in molecular systematic and DNA barcoding studies. *J. Crust. Biol.* 29: 96–110.
- Castresana, J., Lübben, M., Saraste, M., Higgins, D. G. (1994). Evolution of cytochrome oxidase, an enzyme older than atmospheric oxygen. *EMBO J.* 13: 2516–2525.
- Chang, T.-Y., Ng, P. K. L. (2001). On the nomenclature of the commercially important spiny lobster *Panulirus longipes femoristriga* (Von Martens, 1872), *P. bispinosus* Borradaile, 1899, and *P. albiflagellum* Chan & Chu, 1996 (Decapoda, Palinuridae). *Crustaceana* 74: 123–127.
- Chan, T.-Y., Yang, C.-H., Wakabayashi, K. (2019). Amended larval recruitment model for the Japanese spiny lobster *Panulirus japonicus* based on new larval records and population genetic data in Taiwan. *J. Oceanogr.* 75: 273–282.
- Chow, S., Suzuki, N., Imai, H., Yoshimura, T. (2006a). Molecular species identification of spiny lobster phyllosoma larvae of the genus *Panulirus* from the Northwestern Pacific. *Mar. Biotech.* 8: 260–267.
- Chow, S., Yamada, H., Suzuki, N. (2006b). Identification of mid- to final stage phyllosoma larvae of the genus *Panulirus* White, 1847 collected in the Ryukyu Archipelago. *Crustaceana* 79: 745–764.
- Chow, S., Jeffs, A., Miyake, Y., Konishi, K., Okazaki, M., Suzuki, N., Abdullah, M. F., Imai, H., Wakabayashi, T., Sakai, M. (2011). Genetic isolation between the Western and Eastern Pacific populations of pronghorn spiny lobster *Panulirus penicillatus*. *PLoS ONE* 6: e29280.
- Chow, S., Yanagimoto, T., Takeyama, H. (2021). Detection of heteroplasmy and nuclear mitochondrial pseudogenes in the Japanese spiny lobster *Panulirus japonicus*. *Sci. Rep.* 11: 21780.
- Chow, S., Yasuike, M., Yanagimoto, T. (2024a). Re-evaluation of nuclear mitochondrial pseudogenes (NUMTs) and heteroplasmy in the Japanese spiny lobster *Panulirus japonicus*. *Crust. Res.* 53: 27–36.
- Chow, S., Sato, T., Yanagimoto, T. (2024b). Nuclear mitochondrial pseudogenes (NUMTs) detected in the Aesop slipper lobster *Scyllarides haanii* (Crustacea: Decapoda: Scyllaridae). *Aquat. Anim.* 2024: AA2024-24. (In Japanese with English abstract).
- Chow, S., Borsa, P. (2024). A strange mitogenome sequence in spiny lobster. *Aquat. Anim.* 2024: AA2024-32.
- DecaNet eds. (2025). DecaNet. *Panulirus* White, 1847. Accessed through: World Register of Marine Species at: <https://www.marinespecies.org/aphia.php?p=taxdetails&id=107060> on 2025-08-08.
- Diniz, F. M., Maclean, N., Ogawa, M., Cintra, I. H. A., Bentzen, P. (2005). The hypervariable domain of the mitochondrial control region in Atlantic spiny lobsters and its potential as a marker for investigating phylogeographic structuring. *Mar. Biotech.* 7: 462–473.
- Farhadi, A., Pichlmüller, F., Yellapu, B., Lavery, S., Jeffs, A. (2022). Genome-wide SNPs reveal fine-scale genetic structure in ornate spiny lobster *Panulirus ornatus* throughout Indo-West Pacific Ocean. *ICES J. Mar. Sci.* 79: 1931–1941.
- Folmer, O., Black, M., Hoeh, W., Lutz, R., Vrijenhoek, R. (1994). DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Mol. Mar. Biol. Biotechnol.* 3: 294–299.
- Froufe, E., Cabezas, P., Alexandrino, P., Pérez-Losada, M. (2011). Comparative phylogeography of three Achelata lobster species from Macaronesia (North East Atlantic). In: C. Held, S. Koenemann, C. D. Schubart (Eds.) *Phylogeography and Population Genetics in Crustacea*. CRC Press, p. 1–19.
- Fukuda, M., Wakasugi, S., Tsuzuki, T., Nomiya, H., Shimada, K. (1985). Mitochondrial DNA-like sequences in the human nuclear genome. *J. Mol. Evol.* 186: 257–266.
- George, R. W., Holthuis, L. B. (1965). A revision of the Indo-West Pacific spiny lobsters of the *Panulirus japonicus* group. *Zool. Verh. Leiden* 72: 1–36.
- George, R. W., Main, A. R. (1967). The evolution of spiny lobsters (Palinuridae): A study of evolution in the marine environment. *Evolution* 21: 803–820.
- George, R. W. (2006). Tethys sea fragmentation and speciation of *Panulirus* spiny lobsters. *Crustaceana* 78: 1281–1309.
- Giraldes, B. W., Smyth, D. M. (2016). Recognizing *Panulirus meripurpuratus* sp. nov. (Decapoda: Palinuridae) in Brazil—Systematic and biogeographic overview of *Panulirus* species in the Atlantic Ocean. *Zootaxa* 4017 (3): 353–366.
- Gíslason, Ó. S., Svavarsson, J., Halldórsson, H. P., Pálsson, S. (2013). Nuclear mitochondrial DNA (NUMT) in the Atlantic rock crab *Cancer irroratus* Say, 1817 (Decapoda, Cancridae). *Crustaceana* 86: 537–552.
- Handayani, B. K. T., Farajallah, A., Wardiatno, Y. (2019). The suitable COI marker for lobster of genus *Panulirus*. *Pak. J. Sci. Res. Ser. B. Biol. Sci.* 62: 111–115.
- Hazkani-Covo, E., Zeller, R. M., Martin, W. (2010). Molecular poltergeists: mitochondrial DNA copies (numts) in sequenced nuclear genomes.

- PLoS Genet. 6: e1000834.
- Hebert, P. D. N., Ratnasingham, S., deWaard, J. R. (2003a). Barcoding animal life: cytochrome *c* oxidase subunit 1 divergences among closely related species. *Proc. R. Soc. Lond. B (Suppl.)* 270: S96–S99.
- Hebert, P. D. N., Cywinska, A., Ball, S. L., deWaard, J. R. (2003b). Biological identifications through DNA barcodes. *Proc. Biol. Sci.* 270: 313–321.
- Hebert, P. D. N., Bock, D. G., Prosser, S. W. J. (2023). Interrogating 1000 insect genomes for NUMTs: A risk assessment for estimates of species richness. *PLoS ONE* 18: e0286620.
- Hettiarachchi, S. A., Hyeon, J., Mahardini, A., Kim, H., Byun, J., Kim, H., Jeong, J., Yeo, J., Kim, S., Kim, S., Heo, Y., Sathyadith, J., Kang, D., Hur, S. (2022). DNA barcoding and morphological identification of spiny lobsters in South Korean waters: a new record of *Panulirus longipes* and *Panulirus homarus homarus*. *PeerJ* 10: e12744.
- Hieu, N. A., Nhon, N. N. T., Binh, D. T., Sang, D. V., Hanh, P. T. (2019). Classification of spiny lobsters in Vietnam using DNA barcoding. *J. Agricult. Rural Develop.* 12: 101–109.
- Holthuis, L. B. (1991). *Marine Lobsters of the World: an annotated and illustrated catalogue of species of interest to fisheries known to date*. FAO Species Catalogue No. 13. Rome: FAO. pp 1–292.
- Iacchei, M., Ben-Horin, T., Selkoe, K. A., Bird, C. E., Garcia-Rodríguez, F. J., Toonen, R. J. (2013). Combined analyses of kinship and  $F_{ST}$  suggest potential drivers of chaotic genetic patchiness in high gene-flow populations. *Mol. Ecol.* 22: 3476–3494.
- Iacchei, M., O'Malley, J. M., Toonen, R. J. (2014). After the gold rush: population structure of spiny lobsters in Hawaii following a fishery closure and the implications for contemporary spatial management. *Bull. Mar. Sci.* 90: 331–357.
- Iacchei, M., Gaither, M. R., Bowen, B. W., Toonen, R. J. (2016). Testing dispersal limits in the sea: range-wide phylogeography of the pronghorn spiny lobster *Panulirus penicillatus*. *J. Biogeogr.* 43: 1032–1044.
- Inoue, N., Watanabe, H., Kojima, S., Sekiguchi, H. (2007). Population structure of Japanese spiny lobster *Panulirus japonicus* inferred by nucleotide sequence analysis of mitochondrial COI gene. *Fish. Sci.* 73: 550–556.
- Ito, Y., Nishida, S. (2020). Estimation of the origin of *Panulirus brunneiflagellum* caught in Miyazaki Prefecture Japan. *Nature Environ. Miyazaki* 5: 36–41. (In Japanese).
- Jeena, N. S., Gopalakrishnan, A., Radhakrishnan, E. V., Kizhakudan, J. K., Basheer, V. S., Asokan, P. K., Jena, J. K. (2016). Molecular phylogeny of commercially important lobster species from Indian coast inferred from mitochondrial and nuclear DNA sequences. *Mitochondrial DNA Part A* 27: 2700–2709.
- Jeena, N. S., Rahuman, S., Sebastian, W., Kumar, R., Sajeela, K. A., Kizhakudan, J. K., Menon, K. K., Roul, S. K., Gopalakrishnan, A., Radhakrishnan, E. V. (2024). Mitogenomic recognition of incognito lineages in the mud spiny lobster *Panulirus polyphagus* (Herbst, 1793): A tale of unique genetic structuring and diversification. *Int. J. Biol. Macromol.* 277: 134327.
- Jimenez, A. G., Kinsey, S. T., Dillaman, R. M., Kapraun, D. F. (2010). Nuclear DNA content variation associated with muscle fiber hypertrophic growth in decapod crustaceans. *Genome* 53: 161–171.
- Konishi, K., Yanagimoto, T., Chow, S. (2019). Mid- to late stage phyllosoma larvae of *Panulirus brunneiflagellum* Sekiguchi & George, 2005 collected south of the Ogasawara Islands. *Aquat. Anim.* 2019: AA2019-4.
- Lavery, S. D., Farhadi, A., Farahmand, H., Chan, T-Y., Azhdehakoshpour, A., Thakur, V., Jeffs, A. G. (2014). Evolutionary divergence of geographic subspecies within the scalloped spiny lobster *Panulirus homarus* (Linnaeus 1758). *PLoS ONE* 9: e97247.
- Leray, M., Knowlton, N. (2015). DNA barcoding and metabarcoding of standardized samples reveal patterns of marine benthic diversity. *Proc. Natl. Acad. Sci. U.S.A.* 112: 2076–2081.
- Liu, H., Xia, G. (2021). The complete mitochondrial genome of pronghorn spiny lobster *Panulirus penicillatus* (Olivier, 1791). *Mitochondrial DNA Part B Resour.* 6: 148–150.
- Liu, Y., Cui, Z. (2011). Complete mitochondrial genome of the Chinese spiny lobster *Panulirus stimpsoni* (Crustacea: Decapoda): genome characterization and phylogenetic considerations. *Mol. Biol. Rep.* 38: 403–410.
- Lopez, J. V., Yuhki, N., Masuda, R., Modi, W., O'Brien, S. J. (1994). *Numt*, a recent transfer and tandem amplification of mitochondrial DNA to the nuclear genome of the domestic cat. *J. Mol. Evol.* 39: 174–190.
- Mantelatto, F. L., Terossi, M., Negri, M., Buranelli, R. C., Robles, R., Magalhães, T., Tamburus, A. F., Rossi, N., Miyazaki, M. J. (2018). DNA sequence database as a tool to identify decapod crustaceans on the São Paulo coastline. *Mitochondrial DNA Part A* 29: 805–815.
- Meiklejohn, K. A., Damaso, N., Robertson, J. M. (2019). Assessment of BOLD and GenBank – Their accuracy and reliability for the identification of biological materials. *PLoS One* 14: e0217084.
- Naro-Maciel, E., Reid, B., Holmes, K. E., Brumbaugh, D. R., Martin, M., DeSalle, R. (2011).

- Mitochondrial DNA sequence variation in spiny lobsters: population expansion, panmixia, and divergence. *Mar. Biol.* 158: 2027–2041.
- Ng, W-L., Chen, C. A., Mustafa, S., Leaw, C. P., Teng, S. T., Fatimah, S. N., Zakaria, B., Tuzan, A. D., Chan, T-Y. (2022). A new record of the spiny lobster, *Panulirus femoristriga* (von Martens, 1872) from the coastal waters of Malaysia, with revision of global distribution. *Biodivers. Data J.* 10: e77973.
- Ng, W-L., Chen, C. A., Leaw, C. P., Teng, S. T., Nurhasan, R. B., Chan, T-Y. (2024). *Panulirus simpsoni* and checklist of *Panulirus* lobsters in Malaysian waters: morphological and molecular insights. *J. Mar. Biol. Assoc. U. K.* 104: 1–12.
- Pamilo, P., Viljakainen, L., Vihavainen, A. (2007). Exceptionally high density of NUMTs in the honeybee genome. *Mol. Biol. Evol.* 24: 1340–1346.
- Palero et al. (2009). Phylogenetic relationships between spiny, slipper and coral lobsters (Crustacea, Decapoda, Achelata). *Mol. Phylogenet. Evol.* 50: 152–162.
- Panchenko, A. R., Wolf, Y. I., Panchenko, L. A., Madej, T. (2005). Evolutionary plasticity of protein families: Coupling between sequence and structure variation. *Proteins* 61: 535–544.
- Pappalardo, P., Collins, A. G., Lohan, K. M. P., Hanson, K. M., Truskey, S. B., Jaekle, W., Ames, C. L., Goodheart, J. A., Bush, S. L., Biancani, L. M., Strong, E. E., Vecchione, M., Harasewych, M. G., Reedm K., Lin, C., Hartil, E. C., Whelpley, J., Blumberg, J., Matterson, K., Redmond, N. E., Becker, A., Boyle, M. J., Osborn, K. J. (2021). The role of taxonomic expertise in interpretation of metabarcoding studies. *ICES J. Mar. Sci.* 78: 3397–3410.
- Parr, R. L., Maki, J., Reguly, B., Dakubo, G. D., Aguirre, A., Wittcock, R., Robinson, K., Jakupciak, J. P., Thayer, R. E. (2006). The pseudo-mitochondrial genome influences mistakes in heteroplasmy interpretation. *BMC Genomics* 7: 185.
- Pentinsaari, M., Salmela, H., Mutanen, M., Roslin, T. (2016). Molecular evolution of a widely-adopted taxonomic marker (COI) across the animal tree of life. *Sci. Rep.* 6: 35275.
- Pentinsaari, M., Ratnasingham, S., Miller, S. E., Hebert, P. D. N. (2020). BOLD and GenBank revisited – Do identification errors arise in the lab or in the sequence libraries? *PLoS One* 15: e0231814.
- Permana, G. N., Slamet, B., Permana, B. A., Dewi, A. K., Mahardika, G. N. (2019). Population genetic structure of spiny lobsters, *Panulirus homarus* and *Panulirus ornatus*, in the Indian Ocean, Coral Triangle, and South China Sea. *Indon. Aquacult. J.* 14: 7–14.
- Piovesan, A., Pelleri, M. C., Antonaros, F., Strippoli, P., Caracausi, M., Vitale, L. (2019). On the length, weight and GC content of the human genome. *BMC Res. Notes* 12: 106.
- Pranata, B., Fadjar, M., Iranawati, F., Toha, A. H., Jeni (2018). Phylogeny of the spiny lobster *Panulirus versicolor* in Cenderawasih Bay, Papua, Indonesia. *AAACL Bioflux* 11: 1015–1024.
- Pranata, B., Toha, A. H., Kolibongso, D. (2020). Genetic of *Panulirus versicolor* lobster in Cendrawasih Bay Papua and Lombok waters west Nusa Tenggara. *J. Enggano* 5: 249–257.
- Ptacek, M. B., Sarver, S. K., Childress, M. J., Herrnkind, W. F. (2001). Molecular phylogeny of the spiny lobster genus *Panulirus* (Decapoda: Palinuridae). *Mar. Freshw. Res.* 52: 1037–1047.
- Qian, G. H., Zhao, Q., Wang, A., Zhu, L., Zhou, K., Sun, H. (2011). Two new decapod (Crustacea, Malacostraca) complete mitochondrial genomes: bearings on the phylogenetic relationships within the Decapoda. *Zool. J. Linn. Soc.* 162: 471–481.
- Ratnasingham, S., Hebert, P. D. N. (2007). BOLD: The Barcode of Life Data System ([www.barcodinglife.org](http://www.barcodinglife.org)). *Mol. Ecol. Notes* 7: 355–364.
- Ravago, R. G., Juinio-Meñez, A. (2003). Phylogenetic position of the striped-legged forms of *Panulirus longipes* (A. Milne-Edwards, 1868) (Decapoda, Palinuridae) inferred from mitochondrial DNA sequences. *Crustaceana* 75: 1047–1059.
- Reddy, M. M., Macdonald, A. H. H., Groeneveld, J. C., Schleyer, M. H. (2014). Phylogeography of the scalloped spiny-lobster *Panulirus homarus rubellus* in the southwest Indian Ocean. *J. Crust. Biol.* 34: 773–781.
- Sarver, S. K., Silberman, J. D., Walsh, P. J. (1998). Mitochondrial DNA sequence evidence supporting the recognition of two subspecies or species of the Florida spiny lobster *Panulirus argus*. *J. Crust. Biol.* 18: 177–186.
- Sarver, S. K., Freshwater, D. W., Walsh, P. J. (2000). The occurrence of the provisional Brazilian subspecies of spiny lobster (*Panulirus argus westoni*) in Florida waters. *Fish. Bull.* 98: 870–873.
- Sekiguchi, H., George, R. W. (2005). Description of *Panulirus brunneiflagellum* new species with notes on its biology, evolution, and fisheries. *NZ J. Mar. Freshw. Res.* 39: 563–570.
- Schultz, J. A., Hebert, P. D. N. (2022). Do pseudogenes pose a problem for metabarcoding marine animal communities? *Mol. Ecol. Resour.* 22: 2897–2914.
- Senevirathna, J. D. M., Munasinghe, D. H. N. (2013). Identification of taxonomic status of spiny lobster species in Sri Lanka using DNA barcoding and its implications on fisheries and

- conservation programs. *Trop. Agricult. Res.* 25: 96–108.
- Shen, H., Braband, A., Scholtz, G. (2013). Mitogenomic analysis of decapod crustacean phylogeny corroborates traditional views on their relationships. *Mol. Phylogenet. Evol.* 66: 776–789.
- Silberman, J. D., Sarver, S. K., Walsh, P. J. (1994). Mitochondrial DNA variation and population structure in the spiny lobster *Panulirus argus*. *Mar. Biol.* 120: 601–608.
- Song, H., Buhay, J. E., Whiting, M. F., Crandall, K. A. (2008). Many species in one: DNA barcoding overestimates the number of species when nuclear mitochondrial pseudogenes are coamplified. *Proc. Natl. Acad. Sci. USA* 105: 13486–13492.
- Sorenson, M. D., Fleischer, R. C. (1996). Multiple independent transpositions of mitochondrial DNA control region sequences to the nucleus. *Proc. Natl. Acad. Sci. USA* 93: 15239–15243.
- Stein, F., Gailing, O. (2025). Identification of BOLD engine deficiencies and suggestions for improvement based on a curated *Tachina* (Diptera) record set. *PLoS One* 20: e0331216.
- Tamura, K., Stecher, G., Peterson, D., Filipowski, A., Kumar, S. (2013). MEGA6, molecular evolutionary genetics analysis version 6.0. *Mol. Biol. Evol.* 30: 2725–2729.
- Tourinho, J. L., Solé-Cava, A. M., Lazoski, C. (2012). Cryptic species within the commercially most important lobster in the tropical Atlantic, the spiny lobster *Panulirus argus*. *Mar. Biol.* 159: 1897–1906.
- Tsoi et al. (2011). Phylogenetic and biogeographic analysis of the spear lobsters *Linuparus* (Palinuridae, Decapoda), with the description of a new species. *Zool. Anz.* 250: 302–315.
- Veldsman, W. P., Ma, K. Y., Hui, J. H. L., Chan, T. F., Baeza, J. A., Qin, J., Chu, K. H. (2021). Comparative genomics of the coconut crab and other decapod crustaceans: Exploring the molecular basis of terrestrial adaptation. *BMC Genomics* 22: 313.
- Venera-Pontón, D. E., Driskell, A. C., De Grave, S., Felder, D. L., Scioli, J. A., Collin, R. (2020). Documenting decapod biodiversity in the Caribbean from DNA barcodes generated during field training in taxonomy. *Biodivers. Data J.* 8: e47333.
- Williams, S. T., Knowlton, N. (2001). Mitochondrial pseudogenes are pervasive and often insidious in the snapping shrimp genus *Alpheus*. *Mol. Biol. Evol.* 18: 1484–1493.
- Woodings, L. N., Murphy, N. P., Jeffs, A., Suthers, I. M., Liggins, G. W., Strugnell, J. M. (2019). Distribution of Palinuridae and Scyllaridae phyllosoma larvae within the East Australian Current: a climate change hot spot. *Mar. Freshw. Res.* 70: 1020–1033.
- Yamauchi, M. M., Miya, M. U., Nishida, M. (2002). Complete mitochondrial DNA sequence of the Japanese spiny lobster, *Panulirus japonicus* (Crustacea: Decapoda). *Gene* 295: 89–96.
- Yang, C-H., Bracken-Grissom, H., Kim, D., Crandall, K. A., Chan, T-Y. (2012). Phylogenetic relationships, character evolution, and taxonomic implications within the slipper lobsters (Crustacea: Decapoda: Scyllaridae). *Mol. Phylogenet. Evol.* 62: 237–250.
- Yellapu, B., Jeffs, A., Battaglene, S., Lavery, S. (2017). Population subdivision in the tropical spiny lobster *Panulirus ornatus* throughout its Indo-West Pacific distribution. *ICES J. Mar. Sci.* 74: 759–768.

Received: 16 November 2025 | Accepted: 6 January 2025 | Published: 9 January 2026