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Resurrection of Indian-Ocean humbug damselfish, *Dascyllus abudafur* (Forsskål) from synonymy with its Pacific-Ocean sibling, *Dascyllus aruanus* (L.)

Résurrection de la demoiselle domino de l'océan Indien, Dascyllus abudafur *(Forsskål), de la synonymie avec son espèce-jumelle de l'océan Pacifique,* Dascyllus aruanus *(L.)*

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A B S T R A C T

Previous phylogeographic studies of the humbug damselfish, a widespread Indo-West Pacific coral-reef fish, have revealed a split of two main mitochondrial lineages distributed on either side of the Indo-Pacific barrier. This has been interpreted as the result of vicariance. It has been hypothesized that reproductive barriers might currently limit gene flow between humbug damselfish populations from the Indian Ocean and the Pacific Ocean. In this study, we review the published phylogeographic information to update the distribution of the two main mitochondrial lineages of humbug damselfish. The Indian lineage was distributed from the Red Sea to the eastern extremity of the Sunda Shelf while the Pacific lineage, which diverged from the former by 0.6% net nucleotide divergence and diagnostic substitutions at three nucleotide sites at the cytochrome-*b* locus, was distributed east and north of the Sunda Shelf. The two forms, which are also genetically distinct at nuclear loci, were also characterized by distinct pigmentation patterns. We argue that the two forms represent geminate species. Epithet *aruanus* Linnaeus is maintained for the Pacific-Ocean humbug damselfish while epithet *abudafur* (Forsskål**)** is here resurrected for the Indian-Ocean humbug damselfish. Future studies should focus on the population genetic structure of the transition zone between *Dascyllus abudafur* and *D. aruanus*.

Keywords: taxonomy; phylogeography; cryptic species; coral reef; Indo-West Pacific

R É S U M É

Des études phylogéographiques antérieures sur la demoiselle domino, un poisson récifal à large distribution indo-ouest pacifique, ont montré une partition majeure entre lignées mitochondriales de part et d'autre de la barrière indo-pacifique. Ceci a été interprété comme étant le résultat d'un processus de vicariance. L'hypothèse que des barrières reproductives puissent présentement limiter le flux génique entre populations de la demoiselle domino de l'océan Indien et de l'océan Pacifique a été évoquée. Dans cet article, nous passons en revue les données publiées à ce jour, pour déterminer le plus précisément possible la distribution des deux grandes lignées mitochondriales chez la demoiselle domino. Nous montrons que la lignée mitochondriale indienne est présente de la mer Rouge au plateau continental de la Sonde alors que la lignée pacifique, qui est séparée de la première par 0.6% de divergence nucléotidique nette et par des substitutions diagnostiques à trois sites nucléotidiques au locus du cytochrome *b*, est présente à l'est et au nord du plateau de la Sonde. Les deux formes se distinguent également par leurs génotypes nucléaires, ainsi que par leurs patterns de pigmentation. Nous montrons ici que les deux formes représentent des espèces jumelles. L'épithète *aruanus* Linné est maintenue pour la demoiselle domino de l'océan Pacifique, tandis que l'épithète *abudafur* (Forsskål) est ressuscitée pour la demoiselle domino de l'océan Indien. Les études à venir devront aborder la structure génétique de la zone de transition entre *Dascyllus abudafur* et *D. aruanus*.

Mots-clés : taxonomie ; phylogéographie ; espèce cryptique ; récif corallien ; indo-ouest pacifique

1. Introduction

A proportion of Indo-Pacific reef fish species exhibit some phylogeographic discontinuity coinciding with the Indo-Pacific barrier ([1, 2] and references therein). It has been hypothesized that a majority of these phylogeographically structured species actually consist of pairs of geminate species, each restricted to a single ocean [3]. This hypothesis, if confirmed, would considerably affect our current understanding of what determines species richness in the Coral Triangle [4]. This would imply shifting the current paradigm of the Coral Triangle as biodiversity epicenter [4] towards the hypothesis that the higher species richness in the Coral Triangle artefactually results, at least in part, from the overlap in the geographic distributions of Indian and Pacific species [2, 3, 5].

Under the current taxonomic standards, evidence of morphological differentiation is required to support the recognition of separate species. As noted by J.E. Randall [6] about species differentiation between the Indian and the Pacific Oceans: "Many examples of such specific pairs are known […] but perhaps there are others that may have diverged ecologically, behaviourly, or physiologically, but not enough morphologically for us to readily detect". Molecular population genetics should be considered in the latter, as it offers the tools and concepts to address the problem of cryptic species [7]. Actually, new species discoveries in fishes increasingly stem from molecular genetic studies (e.g. [8-14]).

Here, we examine the taxonomic status of a widespread Indo-West Pacific reef fish, the humbug damselfish, *Dascyllus aruanus*. The humbug damselfish is common in shallow reef habitats throughout the Indo-West Pacific. It has been used as a model-species for a number of ecological, behavioural, and population genetic studies [15-26]. The phylogeographic structure of the humbug damselfish indicates two main metapopulations across the Indo-West Pacific, each one specific to an ocean [26]. No mitochondrial haplotype characteristic of Indian-Ocean individuals has been found in the western Pacific and no Pacific haplotype has been found in the Indian Ocean [25, 26]. The phylogeographic break as determined from mitochondrial haplotypes is located at the eastern edge of the Sunda Shelf [25]. Nuclear markers (microsatellites) have also been used for a similar distribution-wide analysis of geographic structure in humbug damselfish [26], showing a same distinct genetic break between the Indian and Pacific Oceans that could not be explained by mere isolation by distance [26]. These observations have been interpreted as the result of vicariance on either side of the Indo-Pacific barrier [26]. It has been hypothesized that reproductive barriers currently limit gene flow between humbug damselfish populations from the Indian Ocean and the Pacific Ocean [25].

We review the phylogeographic information available on the humbug damselfish and provide the geographical delineation of the two main mitochondrial lineages previously uncovered in this species [3, 25-27]. The combined evidence from both mitochondrial DNA and nuclear DNA, and pigmentation patterns allows us to address the issue of cryptic speciation in the humbug damselfish.

2. Materials and Methods

2.1. Review of published sequence data

Previous studies have produced the mitochondrial sequences of humbug damselfish from a total of 43 sampling sites throughout the Indo-West Pacific [3, 20, 25-27] (Fig. 1A). The published mitochondrial data selected for the present survey thus included the complete ATP6/8 gene sequences [842 base pairs (bp)] of 18 individuals from 10 sampling sites in the Indo-West Pacific [27], the partial cytochrome-oxidase I gene sequences (842 bp) of 11 individuals from two sampling sites in the southwestern Indian Ocean and one sampling site in the central Pacific [3], the partial control-region sequences (approximately 359 bp) of 401 individuals from 18 sampling sites in the Coral Triangle [25], and the partial cytochrome-*b* gene sequences (1058 bp) of 260 individuals from 13 sampling sites throughout the Indo-West Pacific [26] (Supplementary material, Table S1). In each of the foregoing surveys, haplotypes have been determined as belonging to either one of two haplogroups, here coined 'Indian' and 'Pacific'. The humbug damselfish population of Cebu Strait, Philippines was sampled by both J.M. Raynal et al. [25] and S.-Y.V. Liu et al. [26]. The haplotype composition of the Cebu sample in [26] was therefore used to establish haplogroup relationships of the control-region haplotypes produced by [25]. The partial control-region sequence from an individual from Glorieuses Islands, southwestern Indian Ocean (C. Fauvelot, unpublished) clustered with haplotypes of the Central Java Sea, thus establishing that the latter belonged to the 'Indian' mitochondrial type (mitotype) [25]. In addition, we examined the set of partial control-region sequences (387 bp) of 45 individuals from 5 sampling sites in French Polynesia [20]. These sequences, accessible from GENBANK (http://www.ncbi.nlm.nih.gov/; accession nos. KM406787-KM406831) were included in a single FASTA file together with reference control-region sequences of the Indian and Pacific mitotypes, and the whole matrix was subjected to Neighbor-Joining analysis using the MEGA 5 package [28].

Similarly, multi-locus microsatellite genotypes from 13 samples from sites across the Indo-West Pacific (Fig. 1B) were determined to belong to either an 'Indian' or a 'Pacific' form according to their probability score under Bayesian structure analysis [26].

2.2. Additional sampling and sequencing

The cytochrome *b* gene sequences of an additional sample of 6 individuals from Madang, Bismarck Sea were produced according to the methods and protocols detailed in [26]. Another 5 individuals from Serangan, on Bali Island, western shore of Lombok Strait were also sequenced at the cytochrome *b* locus, using primers *HIMB01* (5'- G T G A C T T G A A A A A C C A C C G T T G -3') and *HIMB02* (5'- A A T A G G A A G

T A T C A T T C G G G T T T G A T -3'). Polymerase chain reaction was done in 25 μL volumes, using 1 μL of template DNA, 4 μL 10x PCR buffer (Applied Biosystems, Foster City CA, U.S.A.), 2.5 μL 10 mM dNTPs, 1.25 μL of each primer at 10 mM, 2 μL 25 mM MgCl2 solution, 0.125 μL AmplyTaq Gold™ (Applied Biosystems) in doubly-distilled H2O. The thermocycling profile included an initial denaturation of 94 °C for 15 s, 38 cycles of 94 °C for 30 s, 50 °C for 30 s, and 72 °C for 45 s, with a final extension of 72 °C for 5 min. All 11 new sequences were then determined to haplogroup. For this, we compared them to previously published cytochrome *b* gene sequences [26] by checking their nucleotide composition at diagnostic nucleotide sites under MEGA 5 [28]. Control-region sequences of another batch composed by 1 individual from Zanzibar, 3 individuals from the Mozambique channel, 5 from the Philippines, 1 from Guam, and 2 from Fiji, produced according to the protocols exposed in [20], and using primers *CR-A-DA* (5'-A T G A A T C T T A C A A C T C A A C A C C T G-3') and *CR-E-DA* (5'-T C A A C C A A G T A C A A C C C C T G T-3') specifically designed by one of us (CF) for *D. aruanus* were added, and similarly determined to one of the two main haplogroups.

2.3. Pigmentation patterns

Specimens from throughout the distribution of the species were photographed either live underwater or after being captured. The photographs were subsequently examined for pigmentation patterns. Additional photographs were retrieved from FishBase (http://www.fishbase.org/; [29]), and from the Barcoding of Life database (BOLD) (http://www.barcodinglife.com/). The sampling sites for individuals sampled for pigmentation patterns are reported in Fig. 1C. We focused on the intensity of the pigmentation of the blackish blotch at the basis of caudal fin as this was found to be variable among individuals. Four patterns were determined, from blackish (Fig. 2A; scored 3) to lacking pigmentation (Fig. 2B; scored 0) with two intermediate patterns, where the basis of caudal fin appeared light grey (scored 1) or solid grey (scored 2). The samples examined for pigmentation patterns were from the Red Sea (N = 6 individuals), the Mozambique Channel (N = 24), the Mascarene Islands (N = 6), the Maldives (N = 10), Bali (N = 8), Lombok (N = 18), Komodo (N = 8), the South China Sea (N = 5), the Philippines (N = 11), Raja Ampat (N = 35), the Bismarck Sea (N = 4), Chuuk (N = 1), Yap (N = 1), Lord Howe (N = 1), New Caledonia (N = 32), Fiji (N = 4), and the Society Islands (N = 1) (details in Supplementary material, Table S2 and Figs. S1-S7). Mitochondrial sequences were available for 55% of the Indian-Ocean individuals and 41% of the Pacific-Ocean individuals examined from photographs. "Indian Ocean" here refers to the region of the Indo-Pacific west of Lombok Strait, while "Pacific Ocean" refers to the region north and east of Lombok Strait.

2.4. Statistical analyses

The Mantel test implemented in GENETIX 4.02 [30] was used to test the correlation between pairwise θ estimates at the 7 microsatellite loci scored by [26] and their homologues at the cytochrome *b* locus [26]. Net nucleotide distance between the Indian and Pacific mitochondrial haplogroups, based on Kimura-2 parameter nucleotide substitution model, was estimated under MEGA 5 [28]. A χ^2 test of homogeneity was used to test the null hypothesis of no morphological difference between Indian and Pacific forms, as defined by, respectively, geography and mitotype.

3. Results

The frequencies of Indian and Pacific mitochondrial haplogroups per sampling site are given in Supplementary material, Table S1 and reported on Fig. 1A. New cytochrome-*b* and control-region sequences are presented in Fig. 3 and in Supplementary material, Tables S3-S5. The new cytochrome-*b* gene sequences from Lombok Strait (N = 5) comprised one sequence clustering with the previously identified Indian haplogroup, the remainder being of the previously identified Pacific haplogroup. Those from the Madang sample (N = 6) were all found to belong to the Pacific haplogroup. The control-region haplotypes sampled in Zanzibar and Glorieuses Islands all belonged to the Indian-Ocean haplogroup whereas all control-region haplotypes from the Philippines, Guam, Fiji, and French Polynesia belonged to the Pacific-Ocean haplogroup, as confirmed by Neighbor-Joining analysis. The nucleotide distance between the Indian and Pacific mitochondrial haplogroups, estimated from cytochrome *b* gene sequences, was 0.6% (MEGA 5; Kimura-2 parameter net distance [28]).

Two main nuclear forms, Indian and Pacific, were also determined from microsatellite genotypes [26]. The distribution of these two forms is presented Fig. 1B. Genetic differences at nuclear loci were highly correlated with those at the mitochondrial locus (Appendix A).

The average score of pigmentation intensity was 2.51 ± 0.68 in the Indian-Ocean sample (N = 49) and 0.08 ± 0.27 in the Pacific-Ocean sample (N = 103). It was 2.61 ± 0.63 in the Indian mitochondrial form (N = 28) and 0.20 ± 0.40 in the Pacific mitochondrial form (N = 46). The null hypothesis of no differences in pigmentation patterns between the two oceans was rejected (χ^2 test; 3 df; $P < 10^{-28}$), and so was it between the two mitochondrial forms (χ^2 test; 3 df; $P < 10^{-13}$). A majority (62%) of individuals from the Indian Ocean exhibited a blackish blotch at the basis of the caudal fin and no-one was unpigmented. Conversely, a majority (92%) of Pacific-Ocean individuals had an unpigmented caudal fin, the remainder (8%) being slightly pigmented (details in Supplementary material, Table S2). All individuals of the Indian mitotype and for which a photograph was available, except two, possessed a blackish or solid-grey blotch at the basis of the caudal fin. For the two exceptions, from Europa Island, the basis of the caudal fin was light-grey. Conversely, the caudal fin of individuals of the Pacific mitotype was entirely white in most cases, or with a light-grey basis. Individuals sampled in the presumed zone of transition, i.e., Lombok Strait (N = 23) exhibited a variety of

pigmentation patterns, from white to blackish (Supplementary material, Table S2). A specimen exhibiting the typical pigmentation pattern of the Indian-Ocean form is presented Fig. 2A, while a typical specimen of the Pacific form is presented Fig. 2B.

4. Discussion

Under K. de Queiroz's unified species concept [31] species are defined simply as separately evolving metapopulation lineages. K. de Queiroz [31] and other authors [32, 33] have emphasized the fundamental distinction between the definition of species, and species delimitation. The latter concerns properties of species, which include, e.g. morphological distinction, niche separation, reproductive isolation, and monophyly. These properties, one by one or altogether, constitute the lines of evidence researchers in practice may have access to, to hypothesize the existence of a species [31-33].

Several of the expected properties of species characterize the Indian Ocean versus the Pacific Ocean forms of humbug damselfish. The mitochondria of humbug damselfish from the Indian Ocean belong to a single lineage sister to that from the Pacific Ocean: the two forms are reciprocally monophyletic [25-27]. No location with mitotype (Indian vs. Pacific) polymorphism was observed, except at the western shore of Lombok Strait. The transition zone between the two mitochondrial forms lies east of the Central Java Sea and west of Lombok. A phylogeographic partition into two forms was also evident from allelic frequencies at nuclear loci, similar to that at the mitochondrial locus [26]. The location of the nuclear transition zone, which could not be determined with as much precision as the mitochondrial one, lied east of Madagascar and west of the central part of the Coral Triangle. The fact that population genetic differences at nuclear loci were highly correlated with differences at the mitochondrial locus indicates a common history of divergence, between the two forms, for the nuclear and mitochondrial genomes. The distribution of these two genetically separate forms of the humbug damselfish thus appears to be either parapatric, or slightly overlapping, in a narrow transition zone located at the western edge of the Coral Triangle. The two forms were also characterized by distinct pigmentation patterns, a criterion of considerable importance to taxonomists working on reef fishes (e.g., [6, 9, 13]). The two forms thus fulfill the criteria of morphological and genetic distinctness, which in turn allows us to consider them as distinct evolutionary units, hence separate species under the unified species concept [31]. In addition, their apparently parapatric distribution suggests some degree of reproductive isolation [34], which is another property of separate species [31].

In C. Niebuhr's posthume publication of the notes taken by P. Forsskål during his naturalist expedition to the Red Sea, a fish from Jeddah locally named *abu dafur* was mentioned in the following terms: "*Chaetodon abu dafur Arab. est charactere Aruani Linn. Albus, fasciis tribus nigris: spinis Dors. 12. A. 2. Djiddæ habitat, rarior, inter corallia. Spith. Longit.*" [35]. It is possible that P. Forsskål considered this fish to be identical with C. Linnaeus' *Chaetodon aruanus* [36]. *Dascyllus abudafur* (Forsskål) was subsequently synonymized with *D. aruanus* (Linnaeus)

[37, 38]. Epithet *aruanus* has since then been maintained as the single valid one to designate both the Indian and the Pacific forms of the humbug damselfish [29, 39-42]. C. Linnaeus writes that *D. aruanus* "*Habitat in Indiis*" [43]. W.N. Eschmeyer precisely indicates the type-locality to be Aru Islands, Moluccas [42]. Since the Aru Islands are located within the distribution range of the Pacific mitotype, epithet *aruanus* is here kept to designate the Pacific form of the humbug damselfish. No description of the Indian-Ocean form of humbug damselfish is known before P. Forsskål's [42]. Hence, the Indian-Ocean form should be renamed *D. abudafur*.

There are numerous examples of allopatric lineage diversification among marine populations on either side of the Coral Triangle ([1, 2] and references therein). However, ecological factors that have led to lineage diversification, and those that maintain it, are difficult to identify because of the usual difficulty to distinguish contemporary, extrinsic barriers to larval connectivity, from internal barriers to gene flow, i.e. reproductive isolation [44, 45]. Transition zones are privileged situations for addressing such questions, for they offer the possibility to explore the apportion of the various mechanisms, passive vs. active, involved in speciation in marine organisms [46-48]. Investigating the population genetic structure of humbug damselfish populations across the transition zone between *D. abudafur* and *D. aruanus* is thus warranted. Phylogeographic surveys similar to those undertaken in *D. aruanus* ([25, 26]; present study) are needed to assess the taxonomic status of separate genetic forms within a nominal species, in all other cases where a geographic break has been identified.

5. Taxonomy

Dascyllus abudafur (Forsskål 1775), resurrected species

5.1. Previous denominations

Chaetodon abu dafur [35]; *Pomacentrus aruanus* [37]; *Dascyllus aruanus* ([38, 39, 49, 50]; other references in [42]).

5.2. Diagnosis

A *Dascyllus* of white pigmentation with three transversal black bands [35]. Caudal fin bilobate. Possesses a grey to blackish blotch at the basis of caudal fin that distinguishes it from its Pacific-Ocean sister-species, *Dascyllus aruanus* [43], whose tail is generally entirely white. Also distinct from *D. aruanus* by its cytochrome *b* gene sequence: nucleotide sites nos. 450, 459 and 702 of the gene are diagnostic between the two species (Fig. 3). At these three sites, *D. abudafur* has the triplet (A, T, T) whereas *D. aruanus* has (G, C, C).

5.3. Distribution

Indian Ocean, from the Red Sea to the Sunda Shelf.

Disclosure of interest

We have no conflicts of interest concerning this article.

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Fig. 1. Summary of phylogeographic data on humbug damselfish (*Dascyllus abudafur* and *D. aruanus*). *Circle area* proportional to sample size. **A.** Geographic distribution of mitochondrial types from a compilation of published and unpublished sources ([3, 20, 25-27]; present study). *Black*, Indian-Ocean mitotype characterizing *D. abudafur*; *light-grey*, Pacific-Ocean mitotype characterizing *D. aruanus*. **B.** Geographic distribution of nuclear types inferred from microsatellite genotypes [26]. *Black*, genotypes assigned to *D. abudafur*; *grey*, genotypes assigned to *D. aruanus*. **C.** Pie diagrams representing the frequencies of individuals with black or solid-grey (*black*), and light-grey or white (*light grey*) basis of caudal fin.

Fig. 2. Photographs that illustrate the differences in pigmentation patterns between *Dascyllus abudafur* and *D. aruanus*. **A.** *D. abudafur* 64 mm standard length (SL) from Juan de Nova Island, Mozambique Channel; pigmentation score of the blackish blotch at the basis of caudal fin is 3. **B.** *D. aruanus* 45 mm SL from Raja Ampat, West Papua; pigmentation score is 0.

Fig. 3. Variable nucleotide sites in cytochrome *b* gene sequences of humbug damselfish, *Dascyllus abudafur* and *D. aruanus*. Nucleotide sites numbered from the homologous first nucleotide of the gene in *Salmo salar* (GENBANK EU492281) and mutations marked using S.-Y.V. Liu et al.'s [26] sequence of the most common haplotype from the Red Sea, RS02 (GENBANK KF754784), as reference. Haplotypes ordered by location of first occurrence, from West to East (Supplementary material, Table S3). *Highlighted grey*: nucleotide site diagnostic between *D. abudafur* and *D. aruanus*; *dot*: nucleotide identical to reference.

Fig. 1

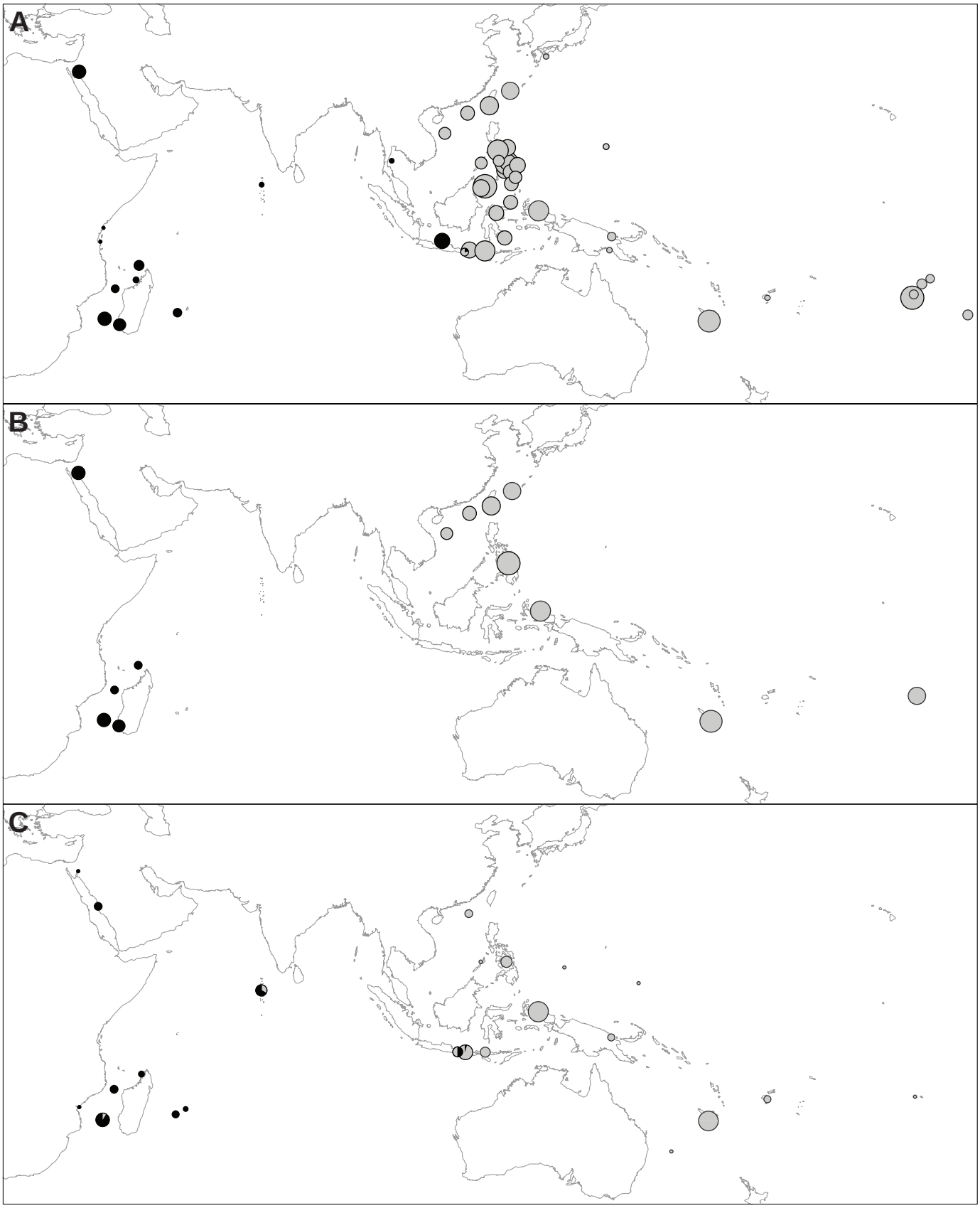

Fig. 2

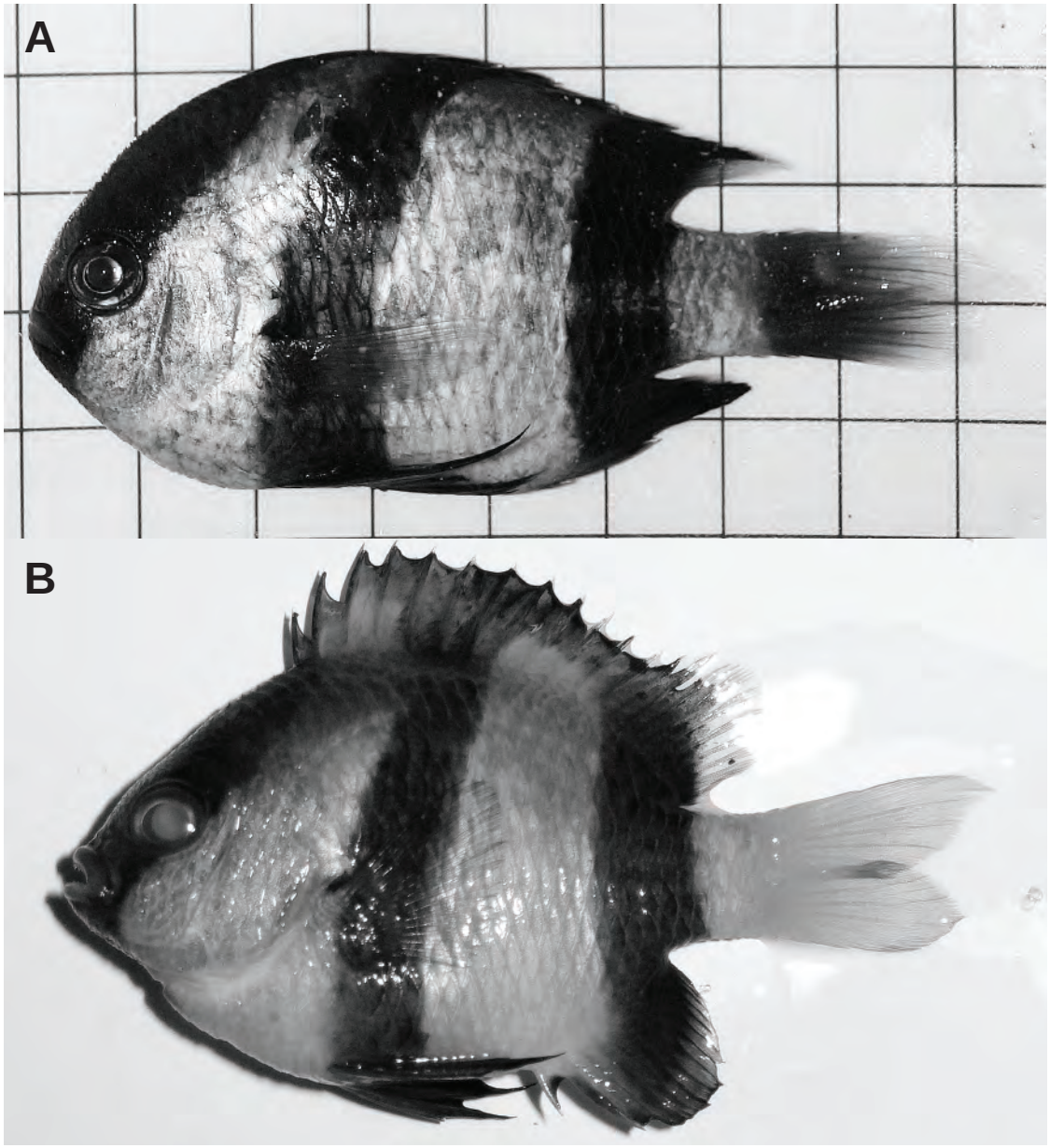

Fig. 3

	GenBank no.	Nucleotide site no.																																																												
		63	108	127	132	133	231	240	252	372	396	432	435	450	459	466	468	471	477	486	489	492	522	543	576	582	585	594	603	639	654	663	681	702	719	735	736	786	789	798	813	840	849	852	870	882	883	895	897	900	915	951	954	966	978	1032	1044	1057	1062	1071	1083	1086	1088
D. abudafur	KF754784	G	C	G	G	A	A	G	T	A	G	C	A	A	T	G	C	C	C	A	A	C	T	C	A	C	T	C	C	G	C	C	G	T	T	C	G	A	G	T	A	C	A	T	G	A	C	T	A	A	T	C	G	A	A	A	C	G	A	C	T	C	C
	KF754787	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
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	KF754783	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
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	KF754778	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	A	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T
	KF754782	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T
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	KF754791	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
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	KF754792	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	G	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T
	KF754794	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	G	.	.	.	.	.	.	.	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	.	T
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	KF754777	.	.	.	.	.	.	A	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	A	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	T
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D. aruanus	KF754774	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
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	KF754763	.	.	.	.	.	.	.	.	G	.	A	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
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	KF754742	.	.	.	.	.	.	.	.	G	.	.	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	A	.	.	.	.	T
	KF754748	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	A	.	.	.	.	.	C	.	.	.	.	.	T	.	.	.	.	.	T
	KF754750	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754759	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	G	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754767	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	T	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
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	KF754746	.	.	A	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	A	.	.	.	.	.	C	.	.	.	.	.	T	.	.	.	.	.	T
	KF754749	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	T	.	.	.	C	.	T
	KF754766	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
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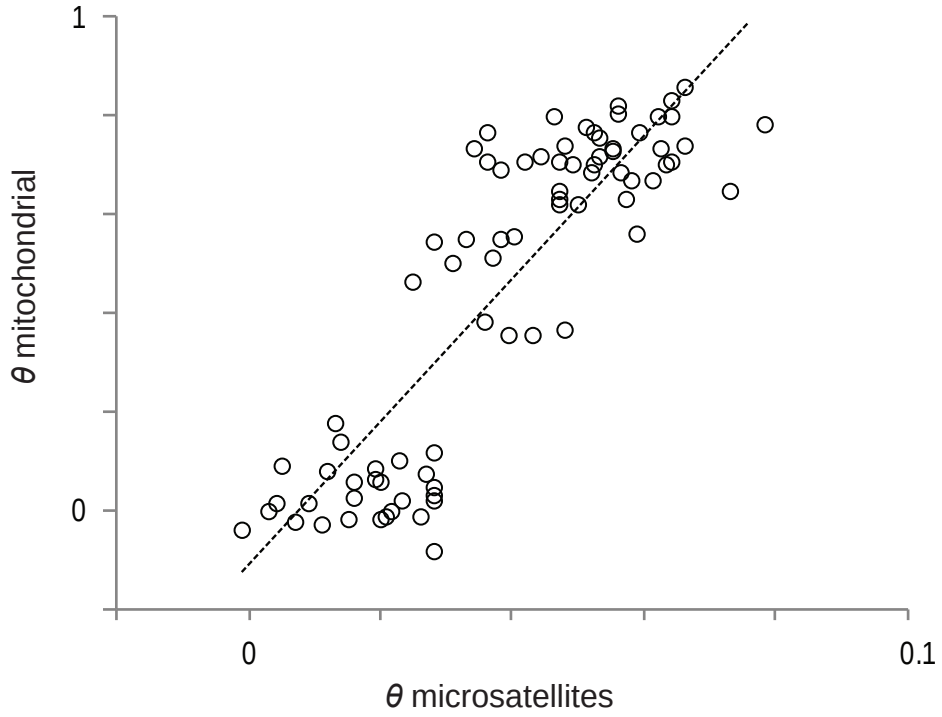
	KF754758	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	A	G	.	.	.	.	.	.	.	G	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754769	.	.	.	.	.	.	.	.	G	A	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754770	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	C	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754771	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754772	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	C	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754740	.	.	.	.	.	.	.	.	G	.	.	.	G	C	.	.	T	.	G	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	A	.	.	.	.	T
	KF754762	.	.	.	.	.	.	.	.	G	.	A	.	G	C	A	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754773	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	A	.	.	.	.	.	.	.	.	.	T
	KF754764	.	.	.	.	.	.	.	.	G	A	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	C	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754765	.	.	.	.	.	.	.	.	G	.	.	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754737	.	.	.	.	.	.	.	.	G	.	.	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	.	.	T	.	.	.	C	.	.	.	.	.	.	A	.	.	.	.	T
	KF754736	.	.	.	A	.	.	.	.	G	.	.	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	A	.	.	.	.	T
	KF754755	.	.	.	.	.	.	.	.	G	.	G	C	G	C	A	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754756	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	C	.	.	.	C	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754738	.	.	.	.	.	.	.	.	G	.	.	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	A	.	.	.	.	T
	KF754739	.	.	.	.	.	.	.	.	G	.	.	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	A	.	.	.	.	.	C	.	.	.	.	.	.	A	.	.	.	.	T
	KF754743	A	.	.	.	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	A	.	.	.	.	.	C	.	.	.	.	.	T	.	.	.	.	.	T
	KF754744	.	.	.	.	.	.	.	.	.	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	A	.	.	.	.	.	C	.	.	.	.	.	T	.	.	.	.	.	T
	KF754745	.	.	.	A	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	A	.	.	.	.	.	C	.	.	.	.	.	T	.	.	.	.	.	T
	KF754735	.	.	.	.	.	.	.	.	G	.	.	.	G	C	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	T	.	.	.	.	.	A	.	.	.	.	T
	KF754752	.	.	.	.	.	.	.	.	G	.	G	.	G	C	.	T	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	A	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754753	.	.	.	.	.	.	.	.	G	.	G	.	G	C	A	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	G	.	.	.	T
	KF754733	.	.	.	.	.	.	A	.	G	.	G	.	G	C	A	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	C	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754761	.	.	.	A	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754734	.	.	.	A	.	.	A	.	G	.	G	.	G	C	A	.	T	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	.	.	.	.	.	C	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T
	KF754760	.	.	.	A	.	.	.	.	G	.	G	.	G	C	.	.	T	.	.	.	.	.	.	.	.	.	.	.	A	.	.	.	C	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	.	.	.	.	.	T

Appendix A.

Correlation of pairwise θ estimates at the mitochondrial locus against homologous θ estimates at 7 microsatellite loci in humbug damselfish (from Supplementary Tables S2 and S3 of [26]). Mantel's test of the correlation between the two matrices: $Z = 1.770$; $r = 0.869$; $P < 0.001$.

Appendix B.

Supplementary material.


1
0
θ mitochondrial
0
0.1
θ microsatellites

SUPPLEMENTARY MATERIAL

Resurrection of Indian-Ocean humbug damselfish, *Dascyllus abudafur* (Forsskål) from synonymy with its Pacific-Ocean sibling, *Dascyllus aruanus* (L.)

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Tables S1-S5 and Figs. S1-S7 here appended.

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Table S1 List of humbug damselfish (*Dascyllus abudafur* and *D. aruanus*) samples used in this article, with frequency of mitochondrial haplotype (either "Indian" or "Pacific"). Samples listed according to geographic location, from West to East. *ATP6/8* ATP synthetase 6, 8 gene; *CO1* cytochrome-oxidase 1 gene; *CR* control region; *cytb* cytochrome b gene; *N*, sample size

Region, Location	Site	*N*	Marker	Haplotype frequency		Reference
				Indian	Pacific	
Indian Ocean						
Red Sea	Gulf of Aqaba	15	*cytb*	100%	-	Liu et al. (2014)
Zanzibar	-	1	*CR*	100%	-	present work
Kenya	-	1	*ATP6/8*	100%	-	McCafferty et al (2002)
Mozambique Channel	Europa I.	15	*cytb*	100%	-	Liu et al. (2014)
Mozambique Channel	Juan de Nova I.	5	*cytb*	100%	-	Liu et al. (2014)
Madagascar	Toliara	12	*cytb*	100%	-	Liu et al. (2014)
Mozambique Channel	Nosy Be	3	*CO1*	100%	-	Hubert et al. (2012)
Mozambique Channel	Glorieuses Is.	5	*cytb*	100%	-	Liu et al. (2014)
Mozambique Channel	Glorieuses Is.	3	*CR*	100%	-	present work
Mascarene Islands	Reunion I.	2	*ATP6/8*	100%	-	McCafferty et al. (2002)
Mascarene Islands	Reunion I.	4	*CO1*	100%	-	Hubert et al. (2012)
Maldives archipelago	-	2	*ATP6/8*	100%	-	McCafferty et al. (2002)
Coral Triangle						
Thailand	-	2	*ATP6/8*	100%	-	McCafferty et al. (2002)
Java Sea, Indonesia	Karimunjawa Is.	19	*CR*	100%	-	Raynal et al. (2014)
Lombok Strait, Indonesia	Serangan	5	*cytb*	20%	80%	present work
South China Sea	Paracels Is.	12	*cytb*	-	100%	Liu et al. (2014)
Lombok Strait, Indonesia	Gili Is.	21	*CR*	-	100%	Raynal et al. (2014)
South China Sea	Dongsha I.	16	*cytb*	-	100%	Liu et al. (2014)
Sulawesi Sea	Semporna, Sabah	23	*CR*	-	100%	Raynal et al. (2014)
Sulu Sea	Palawan I.	12	*CR*	-	100%	Raynal et al. (2014)
Sulawesi Sea	Tawi-Tawi I.	46	*CR*	-	100%	Raynal et al. (2014)
Flores Sea, Indonesia	Komodo I.	33	*CR*	-	100%	Raynal et al. (2014)
Taiwan	Houbihu	27	*cytb*	-	100%	Liu et al. (2014)
Tomimi Bay	Togian Is.	18	*CR*	-	100%	Raynal et al. (2014)
Sibuyan Sea, Philippines	Romblon	36	*CR*	-	100%	Raynal et al. (2014)
Sulu Sea	Guimeras	10	*CR*	-	100%	Raynal et al. (2014)
Bohol Sea, Philippines	Siquijor	20	*CR*	-	100%	Raynal et al. (2014)
Banda Sea, Indonesia	Wakatobi	17	*CR*	-	100%	Raynal et al. (2014)
Cebu Strait, Philippines	Caohagan I., Lapu-Lapu	32	*cytb*	-	100%	Liu et al. (2014)
Cebu Strait, Philippines	Olango	42	*CR*	-	100%	Raynal et al. (2014)
Philippine Sea	Sorsogon	21	*CR*	-	100%	Raynal et al. (2014)
Bohol Sea, Philippines	Camiguin	18	*CR*	-	100%	Raynal et al. (2014)
Philippines	-	1	*ATP6/8*	-	100%	McCafferty et al. (2002)
Philippines	-	5	*CR*	-	100%	present work
Sulawesi Sea	Manado	16	*CR*	-	100%	Raynal et al. (2014)
Sulawesi Sea	General Santos	16	*CR*	-	100%	Raynal et al. (2014)
Davao Gulf, Philippines	Davao	13	*CR*	-	100%	Raynal et al. (2014)
Philippine Sea	Dinagat	20	*CR*	-	100%	Raynal et al. (2014)
Raja Ampat, West Papua	Pulau Fam	33	*cytb*	-	100%	Liu et al. (2014)
Pacific Ocean						
Ryukyu archipelago	Sesoko I.	25	*cytb*	-	100%	Liu et al. (2014)
Japan	-	2	*ATP6/8*	-	100%	McCafferty et al. (2002)
Guam	-	2	*ATP6/8*	-	100%	McCafferty et al. (2002)
Guam	-	1	*CR*	-	100%	present work
New Guinea	-	2	*ATP6/8*	-	100%	McCafferty et al. (2002)
Bismarck Sea, Papua New Guinea	Madang	6	*cytb*	-	100%	present work
New Caledonia	-	2	*ATP6/8*	-	100%	McCafferty et al. (2002)
New Caledonia	SW Lagoon	38	*cytb*	-	100%	Liu et al. (2014)
Fiji	-	2	*CR*	-	100%	present work
Society Islands	Moorea I.	2	*ATP6/8*	-	100%	McCafferty et al. (2002)
Society Islands	Moorea I.	4	*CO1*	-	100%	Hubert et al. (2012)
Society Islands	Moorea I.	25	*cytb*	-	100%	Liu et al. (2014)
Society Islands	Moorea I.	10	*CR*	-	100%	Fauvelot et al. (2003)
Society Islands	Tetiaroa I.	8	*CR*	-	100%	Fauvelot et al. (2003)
Tuamotu Archipelago	Rangiroa Atoll	10	*CR*	-	100%	Fauvelot et al. (2003)
Tuamotu Archipelago	Takapoto Atoll	7	*CR*	-	100%	Fauvelot et al. (2003)
Gambier Archipelago	Marutea Atoll	10	*CR*	-	100%	Fauvelot et al. (2003)

Table S2 *Dascyllus abudafur* and *D. aruanus*. Differences in pigmentation of caudal fin, from photographs of specimens in public repositories, internet sites, and on Figs. S1-S7 here appended. Intensity of pigmentation of the basis of caudal fin was encoded from 3 (blackish) to 0 (no pigmentation). *BOLD* Barcode of Life Database (http://www.barcodinglife.com/), accessed 18 March 2014; *FishBase* (http://www.fishbase.org/), accessed 18 March 2014; *shaded* individuals that also have been sequenced at the mitochondrial locus

Individual no.	Origin	Source	Intensity of pigmentation			
			0	1	2	3
Daaru-u0	Red Sea	Randall 1997b				x
Daaru-u4	Jordan, Red Sea	FishBase			x	
-	Jeddah, Red Sea	http://www.oceanfootage.com/			x	
-	Jeddah, Red Sea	http://www.oceanfootage.com/				x
-	Jeddah, Red Sea	http://www.oceanfootage.com/				x
-	Jeddah, Red Sea	http://www.oceanfootage.com/				x
Daaru-u1	Mozambique, Mozamb. Channel	FishBase				x
Europa 1	Europa I., Mozamb. Channel	Fig. S1				x
Europa 2	Europa I., Mozamb. Channel	Fig. S1				x
Europa 3	Europa I., Mozamb. Channel	Fig. S1				x
Europa 4	Europa I., Mozamb. Channel	Fig. S1				x
Europa 5	Europa I., Mozamb. Channel	Fig. S1				x
Europa 6	Europa I., Mozamb. Channel	Fig. S1			x	
Europa 7	Europa I., Mozamb. Channel	Fig. S1			x	
Europa 8	Europa I., Mozamb. Channel	Fig. S1			x	
Europa 9	Europa I., Mozamb. Channel	Fig. S1			x	
Europa 10	Europa I., Mozamb. Channel	Fig. S1		x		
Europa 11	Europa I., Mozamb. Channel	Fig. S1		x		
Europa 12	Europa I., Mozamb. Channel	Fig. S1			x	
Europa 13	Europa I., Mozamb. Channel	Fig. S1				x
Europa 14	Europa I., Mozamb. Channel	Fig. S1				x
Europa 15	Europa I., Mozamb. Channel	Fig. S1				x
JdN 1	Juan de Nova I., Mozamb. Channel	Fig. S2				x
JdN 2	Juan de Nova I., Mozamb. Channel	Fig. S2				x
JdN 3	Juan de Nova I., Mozamb. Channel	Fig. S2				x
JdN 4	Juan de Nova I., Mozamb. Channel	Fig. S2				x
JdN 5	Juan de Nova I., Mozamb. Channel	Fig. S2				x
NBE 187	Nosy Be, Mozamb. Channel	BOLD				x
NBE 188	Nosy Be, Mozamb. Channel	BOLD				x
NBE 189	Nosy Be, Mozamb. Channel	BOLD				x
REU 0004-01	Reunion, Mascarene Is.	BOLD				x
REU 0004-02	Reunion, Mascarene Is.	BOLD				x
REU 2764	Reunion, Mascarene Is.	BOLD			x	
REU 2765	Reunion, Mascarene Is.	BOLD			x	
Daaru-u5	Mauritius, Mascarene Is.	FishBase			x	
Daaru-u6	Mauritius, Mascarene Is.	FishBase			x	
-	Maldives	http://www.travelswithhok.com				x
-	Maldives	http://www.travelswithhok.com				x
-	Maldives	http://www.travelswithhok.com				x
-	Maldives	http://www.travelswithhok.com				x
-	Maldives	http://www.travelswithhok.com				x
-	Maldives	http://www.travelswithhok.com			x	
-	Maldives	http://www.travelswithhok.com			x	
-	Maldives	http://www.travelswithhok.com		x		
-	Maldives	http://www.travelswithhok.com		x		
-	Maldives	http://www.travelswithhok.com		x		
Bali 1	Singaraja, Bali	Fig. S2			x	
Bali 2	Singaraja, Bali	Fig. S2				x
Bali 3	Singaraja, Bali	Fig. S2				x
IBRC 11.01.08	Serangan, western Lombok Strait	Fig. S6		x		
IBRC 11.01.14	Serangan, western Lombok Strait	Fig. S6				x
IBRC 11.01.22	Serangan, western Lombok Strait	Fig. S6		x		
IBRC 11.01.27	Serangan, western Lombok Strait	Fig. S6		x		
IBRC 11.01.28	Serangan, western Lombok Strait	Fig. S6	x			
Lombok 1	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 2	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 3	Gili Gede, eastern Lombok Strait	Fig. S7	x			

Table S2 (continued)

Individual no.	Origin	Source	Intensity of pigmentation			
			0	1	2	3
Lombok 4	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 5	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 6	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 7	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 8	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 9	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 10	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 11	Gili Gede, eastern Lombok Strait	Fig. S7			x	
Lombok 12	Gili Gede, eastern Lombok Strait	Fig. S7	x			
Lombok 13	Gili Layar, eastern Lombok Strait	Fig. S7	x			
Lombok 14	Gili Layar, eastern Lombok Strait	Fig. S7	x			
Lombok 15	Gili Layar, eastern Lombok Strait	Fig. S7	x			
Lombok 16	Gili Layar, eastern Lombok Strait	Fig. S7	x			
Lombok 17	Gili Layar, eastern Lombok Strait	Fig. S7	x			
Lombok 18	Gili Layar, eastern Lombok Strait	Fig. S7	x			
-	Komodo I., Flores Sea	http://www.sfups.org/	x			
-	Komodo I., Flores Sea	http://www.sfups.org/	x			
-	Komodo I., Flores Sea	http://www.sfups.org/	x			
-	Komodo I., Flores Sea	http://www.sfups.org/	x			
-	Komodo I., Flores Sea	http://www.sfups.org/	x			
-	Komodo I., Flores Sea	http://www.sfups.org/	x			
-	Komodo I., Flores Sea	http://www.sfups.org/	x			
-	Komodo I., Flores Sea	http://www.sfups.org/	x			
042913-06	Dongsha., South China Sea	Fig. S2	x			
WJC2243	Dongsha., South China Sea	Fig. S2	x			
DS 1	Dongsha., South China Sea	Fig S4	x			
DS 2	Dongsha., South China Sea	Fig S4	x			
DS 3	Dongsha., South China Sea	Fig S4	x			
HLC-10828	Philippines	BOLD	x			
HLC-10829	Philippines	BOLD	x			
HLC-10830	Philippines	BOLD		x		
HLC-10831	Philippines	BOLD		x		
Daaru-u2	Palawan, Philippines	FishBase	x			
Daaru-u3	Philippines	Randall (1997a)		x		
LL 1	Lapu-Lapu I., Philippines	Fig S4	x			
LL 2	Lapu-Lapu I., Philippines	Fig S4	x			
LL 3	Lapu-Lapu I., Philippines	Fig S4	x			
LL 4	Lapu-Lapu I., Philippines	Fig S4	x			
LL 5	Lapu-Lapu I., Philippines	Fig S4	x			
-	Pulau Pef, Raja Ampat, West Papua	http://faroutphotos.com	x			
Sayonek 1	Pulau Sayonek, Raja Ampat	Fig. 2B	x			
Fam 1	Pulau Fam, Raja Ampat, West Papua	Fig. S3		x		
Fam 2	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 3	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 4	Pulau Fam, Raja Ampat, West Papua	Fig. S3		x		
Fam 5	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 6	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 7	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 8	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 9	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 10	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 11	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 12	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 13	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 14	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 15	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 16	Pulau Fam, Raja Ampat, West Papua	Fig. S3		x		
Fam 17	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 18	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 19	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 20	Pulau Fam, Raja Ampat, West Papua	Fig. S3		x		
Fam 21	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 22	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 23	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			

Table S2 (continued)

Individual no.	Origin	Source	Intensity of pigmentation			
			0	1	2	3
Fam 24	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 25	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 26	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 27	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 28	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 29	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 30	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 31	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 32	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
Fam 33	Pulau Fam, Raja Ampat, West Papua	Fig. S3	x			
PR161-1	Madang., Bismarck Sea	Fig S2	x			
PR161-2	Madang., Bismarck Sea	Fig S2	x			
PR161-3	Madang., Bismarck Sea	Fig S2	x			
PR161-4	Madang., Bismarck Sea	Fig S2	x			
-	Chuuk	Allen and Erdmann (2012)	x			
-	Yap	http://reefguide.org/	x			
-	Lord Howe I.	Randall (2005)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
-	New Caledonia	Laboute and Grandperrin (2000)	x			
Poé 1	Poé, New Caledonia	Fig. S4	x			
Poé 2	Poé, New Caledonia	Fig. S4	x			
Poé 3	Poé, New Caledonia	Fig. S4	x			
Poé 4	Poé, New Caledonia	Fig. S4	x			
Poé 5	Poé, New Caledonia	Fig. S4	x			
Poé 6	Poé, New Caledonia	Fig. S4	x			
Poé 7	Poé, New Caledonia	Fig. S4	x			
Poé 8	Poé, New Caledonia	Fig. S4	x			
Poé 9	Poé, New Caledonia	Fig. S4	x			
Poé 10	Poé, New Caledonia	Fig. S4	x			
SWLagoon 1	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 2	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 3	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 4	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 5	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 6	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 7	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 8	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 9	SW lagoon, New Caledonia	Fig. S5	x			
SWLagoon 10	SW lagoon, New Caledonia	Fig. S5	x			
Muaivuso 1	Viti Levu, Fiji	Fig. S4		x		
Muaivuso 2	Viti Levu, Fiji	Fig. S4	x			
Muaivuso 3	Viti Levu, Fiji	Fig. S4	x			
Muaivuso 4	Viti Levu, Fiji	Fig. S4	x			
MBIO 556_1	Moorea, Society Is.	BOLD	x			

Table S3 Humbug damselfish, *Dascyllus abudafur* and *D. aruanus*. Cytochrome *b* gene haplotype frequency per sample. *Abbreviations for samples* as in Liu et al. (2014); *other abbreviation*: *MA* Madang, Bismarck Sea; *N* sample size; *n* frequency of haplotype in total sample. Samples listed according to geographic location, from West to East

GenBank no.	Sample														*n*
	RS	EU	JN	MD	GL	PI	DS	TW	LL	SK	RA	MA	NC	SI	
KF754784	6														6
KF754787	4														4
KF754776	1														1
KF754780	1														1
KF754783	1														1
KF754785	1														1
KF754786	1														1
KF754778		3	1	1											5
KF754782		2	2	3											7
KF754788		2		2											4
KF754791		2		1	1										4
KF754793		2		1											3
KF754781		1	1	1	1										4
KF754792		1													1
KF754794		1	1												2
KF754795		1		1											2
KF754779				1											1
KF754789				1											1
KF754775					1										1
KF754777					1										1
KF754790					1										1
KF754774						6	6	15	13	10	10		1		61
KF754757						2									2
KF754763						2	1	1							4
KF754741						1									1
KF754742						1	1	3	5	6	13	3	8		40
KF754748							4	1	3	5	1	1	11		26
KF754750							1	4	2		2	2			11
KF754759							1		1	1					3
KF754767							1								1
KF754768							1								1
KF754746								1							1
KF754749								1							1
KF754766								1			1				2
KF754747									1						1
KF754751									1						1
KF754754									1						1
KF754758									1						1
KF754769									1				1	2	4
KF754770									1						1
KF754771									1						1
KF754772									1						1
KF754740										1					1
KF754762										1					1
KF754773										1					1
KF754764											2				2
KF754765											1				1
KF754737											1				1
KF754736											1				1
KF754755											1				1
KF754756													6		6
KF754738													4		4
KF754739													1		1
KF754743													1		1
KF754744													1		1
KF754745													1		1
KF754735													1		1
KF754752													1		1
KF754753													1		1
KF754733														19	19
KF754761														2	2
KF754734														1	1
KF754760														1	1
N	15	15	5	12	5	12	16	27	32	25	33	6	38	25	266

Table S4 Partial cytochrome-*b* gene sequences (in FASTA format) of 5 humbug damselfish individuals from Serangan, western Lombok Strait

Sequence
>IBRC 110114_Serangan ATGGCCAGCTTCCGCAAAACTCACCCACTGCTTAAAGCAGCAAATGATGCAGTAATCGAC CTGCCTACGCCATCCAACATCTCTGTCTGATGGAACTTTGGCTCCCTCCTAGGACTCTGCT TAATCGCCCAGATCCTCACAGGCCTCTTTCTCGCCATACATTATACATCAGATATCGCC AC AGCTTTCTCCTCCGTAACCCACATCTGCCGAGATGTCAACTACGGATGACTTATCCGGAAT ATACACGCTAACGGCGCATCCTTCTTCTTTATCTGCATCTACCTCCACATCGGACGA GGAC TCTACTACGGCTCATTCCTTCACAAAGAGACATGAAACGTAGGAGTAATCCTCCTCCTCTT AGTAATAATAACAGCTTTCGTAGGCTACGTGCTTCCTTGAGGCCAAATATCCCTC TGAGG TGCCACCGTAATCACAAACCTCCTATCAGCCTTTCCCTACGTCGGTGATTCCCTAGTCCAAT GAATCTGAGGGGGCTTCTCAGTAGACAATGCCACTCTCACCCGATTCTTTGCC TTCCACTT CCTTTTTCCCTTTGTAATTGTAGCCATAACCCTCGTTCACCTTATCTTCCTCCACGAAACTG GATCAAACAACCCAACTGGCCTAAACTCGGATGCAGACAAAATCTCCTTCCACCCTTATTT CTCCTACAAGGACATCCTCGGCTTTGCAGTTCTGCTACTAGCACTAATTTCACTAGCACTA TTCGCCCCCAATCTTCTCGGAGACCCCGACAACTTC---------- ---------- ---------- ---------- ---------- >IBRC 110127_Serangan ATGGCCAGCTTCCGCAAAACTCACCCACTGCTTAAAGCAGCAAATGATGCAGTAATCGAC CTGCCTACGCCATCCAACATCTCTGTCTGATGGAACTTTGGCTCCCTCCTAGGACTCTGC T TAATCGCCCAGATCCTCACAGGCCTCTTTCTCGCCATACATTATACATCAGATATCGCCACA GCTTTCTCCTCCGTAACCCACATCTGCCGAGATGTCAACTACGGATGACTTATCCGG AATA TACACGCTAACGGCGCATCCTTCTTCTTTATCTGCATCTACCTCCACATCGGACGA GGACT CTACTACGGCTCATTCCTTCACAAAGAGACATGAAACGTAGGAGTAATCCTCCTC CTCTTA GTAATGATAACAGCTTTCGTAGGCTACGTGCTTCCTTGAGGCCAAATATCCCTCTGAGGT GCCACCGTAATCACAAACCTCCTGTCAGCCTTCCCCTACGTCGGTGATTCCCTA GTCCAAT GAATCTGAGGGGGCTTCTCAGTAGACAATGCCACTCTCACCCGATTCTTTGCCTTCCACTT CCTTTTTCCCTTTGTAATTGTAGCCATAACCCTCGTTCACCTTATCTTCCTC CACGAAACTG GATCAAACAACCCAACTGGCCTAAACTCGGATGCAGACAAAATCTCCTTCCACCCTTATTT CTCCTACAAGGACATCCTCGGCTTTGCAGTCCTGCTACTAGCACTAATT TCACTAGCACTA TTTGCCCCCCAATCTTCTCGGAGACCCCGACAACTTCATCCCCGCTAAC CCCATGGTGACAC CACCTCAC---------- ---------- ---------- ---------- ---------- >IBRC 110122_ Serangan ATGGCCAGCTTCCGCAAAACTCACCCACTGCTTAAAGCAGCAAATGATGCAGTAATCGACC TGCCTACGCCATCCAACATCTCTGTCTGATGGAACTTTGGCTCCCTCCTAGGACTCTGCTT AATCGCCCAGATCCTCACAGGCCTCTTTCTCGCCATACATTATACATCAGATATCGCCACA GCTTTCTCCTCCGTAACCCACATCTGCCGAGATGTCAACTACGGATGACTTATCCGGAATA TACACGCTAACGGCGCATCCTTCTTCTTTATCTGCATCTACCTCCACATCGGACGAGGACT CTACTACGGCTCATTCCTTCACAAAGAGACATGAAACGTAGGAGTAATCCTCCTCCTCTTA GTAATGATAACAGCTTTCGTAGGCTACGTGCTTCCTTGAGGCCAAATATCCCTCTGAGGT GCCACGGTAATCACAAACCTCCTGTCAGCCTTCCCCTACGTCGGTGATTCCCTAGTCCAAT GAATCTGAGGGGGCTTCTCAGTAGACAATGCCACTCTCACCCGATTCTTTGCCTTCCACTT CCTTTTTCCCTTTGTAATTGTAGCCATAACCCTCGTTCACCTTATCTTCCTCCACGAAACTG GATCAAACAACCCAACTGGCCTAAACTCGGATGCAGACAAAATCTCCTTCCACCCTTATTT CTCCTACAAGGACATCCTCGGCTTTGCAGTCCTGCTACTAGCACTAATTTCACTAGCACTA TTCGCCCCCAATCTTCTCGGAGACCCCGACAACTTCATCCCCGCTAACCCCATGGTGACAC CACCTCAC---------- ---------- ---------- ---------- ---------- >IBRC 110128_ Serangan ATGGCCAGCTTCCGCAAAACTCACCCACTGCTTAAAGCAGCAAATGATGCAGTAATCGACC TGCCTACGCCATCCAACATCTCTGTCTGATGGAACTTTGGCTCCCTCCTAGGACTCTGCTT AATCGCCCAGATCCTCACAGGCCTCTTTCTCGCCATACATTATACATCAGATATCGCCACA GCTTTCTCCTCCGTAACCCACATCTGCCGAGATGTCAACTACGGATGACTTATCCGGAATA TACACGCTAACGGCGCATCCTTCTTCTTTATCTGCATCTACCTCCACATCGGACGAGGACT CTACTACGGCTCATTCCTTCACAAAGAGACATGAAACGTAGGAGTAATCCTCCTCCTCTTA GTAATGATAACAGCTTTCGTAGGCTACGTGCTTCCTTGAGGCCAAATATCCCTCTGAGGT GCCACGGTAATCACAAACCTCCTGTCAGCCTTCCCCTACGTCGGTGATTCCCTAGTCCAAT GAATCTGAGGGGGCTTCTCAGTAGACAATGCCACTCTCACCCGATTCTTTGCCTTCCACTT CCTTTTTCCCTTTGTAATTGTAGCCATAACCCTCGTTCACCTTATCTTCCTCCACGAAACTG GATCAAACAACCCAACTGGCCTAAACTCGGATGCAGACAAAATCTCCTTCCACCCTTATTT CTCCTACAAGGACATCCTCGGCTTTGCAGTCCTGCTACTAGCACTAATTTCACTAGCACTA TTCGCCCCCAATCTTCTCGGAGACCCCGACAACTTCATCCCCGCTAACCCCATGGTGACAC CACCTCAC---------- ---------- ---------- ---------- ---------- >IBRC 110108_ Serangan ATGGCCAGCTTCCGCAAAACTCACCCACTGCTTAAAGCAGCAAATGATGCAGTAATCGACC TGCCTACGCCATCCAACATCTCTGTCTGATGGAACTTTGGCTCCCTCCTAGGACTCTGCTT AATCGCCCAGATCCTCACAGGCCTCTTTCTCGCCATACATTATACATCAGATATCGCCACA GCTTTCTCCTCCGTAACCCACATCTGCCGAGATGTCAACTACGGATGACTTATCCGGAATA TACACGCTAACGGCGCATCCTTCTTCTTTATCTGCATCTACCTCCACATCGGACGAGGACT CTACTACGGCTCATTCCTTCACAAAGAGACATGAAACGTAGGAGTAATCCTCCTCCTCTTA GTAATGATAACAGCTTTCGTAGGCTACGTGCTTCCTTGAGGCCAAATATCCCTCTGAGGT GCCACGGTAATCACAAACCTCCTGTCAGCCTTCCCCTACGTCGGTGATTCCCTAGTCCAAT GAATCTGAGGGGGCTTCTCAGTAGACAATGCCACTCTCACCCGATTCTTTGCCTTCCACTT

CCTTTTTCCCTTTGTAATTGTAGCCATAACCCTCGTTCACCTTATCTTCCTCCACGAAACTG
GATCAAACAACCCAACTGGCCTAAACTCGGATGCAGACAAAATCTCCTTCCACCCTTATTT
CTCCTACAAGGACATCCTCGGCTTTGCAGTCCTGCTACTAGCACTAATTTCACTAGCACTA
TTCGCCCCCAATCTTCTCGGAGACCCCGACAACTTCATCCCCGCTAACCCCATGGTGACAC
CACCTCAC----------

Table S5 Partial control-region sequences (in FASTA format) of humbug damselfish, *Dascyllus abudafur* and *D. aruanus*

Species, Sequence
Dascyllus abudafur
>DZA2 (Zanzibar)
GATATAACTAACATTAATTCTTGGTGCTTTAAATAGTTTATAGTATTTAAAATGTGATTTA
AAAATGTGAATCTTACAACTCAACACCTGTTT-GTAGCATTATCCCATAAGCTTGCACCGCT
ACGTGCATATATGTACATTCACCATAAATCTATATTAACCATATCATAGAATATTCAAGTAC
ATGATTTCAAACTTTAACTAATATTAAAGTCAAGGATATACTTGTTTTGAAACATTCACCT
GAATAAAGGTACATAAACCTCTACATGTGTCAAAAGCTAAGCGGGTACAGGTAAAAATTT
AAGATTCAACTGTTACATACTCATGGGTCCAGATATACCAAGTATTCAGTACCTCTGCAAG
TCATATGTTTAATACAGTAAGA
>G lor1 (Glorieuses Islands)
GACATAACTAACATTAATTCTTAGTGCTATAAATAGTTTATAGTATTTAAAATGTGATTTA
AAAATGTGAATCTTACAACTCAACACCTGTTT-GTAGCATTATCCCATAAGCTTGCACCGCT
ACGTGCATATATGTACATTCACCATAAATCTATATTAACCATATCATAGATATTTAAGTACA
TGATTTCAAGCTTTAACTAATACCAAAGTCAAAGGACATACTTGTTTTGAAACATTCACCT
GAATAAAAGCACATAAACCTCTACATGTATCAAAAAGTAAGCGGGTACAATTAAAGATTTA
AGATTTAACCGTTACATACTCATGGGTTCAGATATACCAAGTATTCAGTACCTCTGCAAGT
CATATGTTTAATACAGTAAGAGCCCAGCATCAGTTGATTCCTGAATGTCCACAGTTCTTGA
TGGTCAAGGACAGAATAA
>G lor2 (Glorieuses Islands)
GACATAACTAACATTAATTCTTAGTGCTATAAATAGTTTATAGTATTTAAAATGTGATTTA
AAAATGTGAATCTTACAACTCAACACCTGTTT-GTAGCATTATCCCATAAGCTTGCACCGCT
ACGTACATATATGTACATTCACCATAAATCTATATTAACCATATCATAGATATTCAAGTACA
TGATTTCAAACTTTAACTAATACTAAAGTCAA-GGATATACTTGTTTTGAAACATTCACCTG
AATAAAAGTACATAAACCTCTACATGTGTCAAAAAGTAAGCGGGTACAAGTAAAGATTTA
AGATTCAACTGTTACATACTCATGAGTTCAGATATACCAAGTATTCAGTACCTCTGCAAGT
CATATGTTTAATACAGTAAGAGCCCAGCATCAGTTGATTCCTAAATGTCCATAGTTCTTGA
TGGTCAAGGACAGAATAA
>Glor3 (Glorieuses Islands)
GACATAACTAACATTAATTCTTAGTGCTATAAATAGTTTATAGTATTTAAAATGTGATTTA
AAAATATGAATCTTACAACTCAACACCTGTTT-GTAGCATTATTCCATAAGCTTGCACCGCT
ACGTGCATATATGTACATTCACCATAAATCTATATTAACCATATCATGGGTACTCAAGTAC
ATGATTTCAAACTTTGACTAATATTAAAGTCAA-GGATATAATTGTTTTAAGGCATTTACCT
GAATAAAAGTACATAAACCTCTGCATGTGTCAAAAGGTAAGCGGGTACAAACGAAGATTT
AAGATTCAGTTGTTACATACTCATGGGTCCAGATATACCAAGTACTCAGTACCTCTGCAAG
TCACATGTTTAATACAGTAAGAGCCCAGCATCAGTTGATTTCTAAATGTCCACGGTTCTTG
ATGGTCAAGGACAGAATAA
Dascyllus aruanus
>Phil5 (Philippines)
----------TACAACTCAACACCTG
TTTTGTAGCATTATTCCATAAGCTTGCACTTCTACGTGCATATATGTATATTCACCATGAAT
CTATATTAACCATATCATGGATATTCAAGTACATGATTTCAAGCTTTAACTAATATTAAAAT
CAA-GGATATACTTGTTTTAAAGCATTCACCTAAATAAAAGTACATGAACCCCTACATGTGT
CAAAAGGTAAGCGGGCACAAGTAAAAATTTAAGACTCAACCGTTACATACTCATGGGTCT
AGATATACCAAGTATTCAGTACCTCTGCAAGTCACATGTTTAATACAGTAAGAGCCCAGCA
TCAGTTGATTTCTAAA----------
>Phil11 (Philippines)
----------TACAACTCAACAC-TG
TTTTGTAGCATTATTCCATAAGCTTGCACTTCTACGTGCATATATGTATATTCACCATAAAT
CTATATTAACCATATCATGGATATTCAGGTACATGACTTCAAGTCTTAACCAATCTTAAAAT
CAA-GGATATGCTTGTTCCAAAACATTCACCTAAATAGAAGTACATAAACCCCCACATGTG
TCGAAAGGTAAACGAGTACAAGTAAAAATTTAAGATTTAACCGTTACATACTCATGAGTTC
AGATATACCAAGTACTCAGCACCTCTGCAAGCCATATGTTTAATACAGTAAGAGCCCAGCA
TCAGTTGATTTCTAAA----------
>Phil12 (Philippines)
----------TACAACTCAACACCTG
TTTTGTAGCATTATTCCATAAGTTTGCACTTCTACGTGCATATATGTATATTCACCATGAAT
CTATATTAACCATATCATGGATATTCGAGTACATGATTTCAAGCTTTAACTAATCTTAAAAT
CAA-GGATATACTTGTTTTAAAGCATTCACCTAAATAAAAGTACATAAACCCCTACATGTGT
CAAAGGGTAAGCAGGCACAAGTAAAAATTTAAAACTCAACCGTTACATACTCATGAGTCCA
GATATACCAAGTATTCAATACCTCTGCAAGTCACATGTTTAATACAGTAAGAGCCCAGCAT
CAGTTGATTTCTAAA----------
>Phil13 (Philippines)
----------TACAACTCAACACCTG
TTTTGTAGCATTATTCCATAAGCTTGCACTTCTACGTGCATATATGTATATTCACCATGAAT
CTATATTAACCATATCATGGATATTCGAGTACATGACTTCAAGCTTTAACTAATCTTAAAAT
CAA-GGATATACTTGTTTTAAAGCATTCACCTAAATAAAAGTACATAAACCCCTACATGTGT
CAAAAGGTAAGCAGGCACAAGTAAAAATTTAAGACTCAACCGTTACATACTCATGTGTCCA
GATATACCAAGTATTCAGTACCTCTGCAAGTCACATGTTTAATACAGTAAGAGCCCAGCAT
CAGTTGATTTCTAAA----------
>Phil20 (Philippines)
----------TACAACTCAACACCTG
TTTTGTAGCATTATTCCATAAGCTTGCACTTCTACGTGCATATATGTATATTCACCATGAAT
CTATATTAACCATATCATGGGTATTCGAGTACATGATTTCAAGCTTTAACTAATCTTAAAA
TCAA-GGATATACTTGTTTTAAAACATTCACCTAAATAAGAGTACATAAACCCTCACATGTA
TCAAAAGGTAAGCAGGCACAAGTAAGAATTTAAGACTCAACCGTTACATACTCATGAGTC
AAGATATACCAAGTATTCAGTACCTCTGCAAGTCACATGTTTAATACAGTAAGAGCCCAGC
ATCAGTTGATTTCTAAA----------
>Guam17 (Guam)
----------TACAACTCAACACCTG
TTTTGTAGCATTATTCCATAAGCTTGCACTTCTACGTGCATATATGTATATTCACCATGAAT
CTATATTAACCATATCATGGATATTCGAGTACATGATTTCAAGCTTTAATTAATCTTAAAAT

CAA-GGATATACTTGTTTTAAAGCATTCACCTAAATAANAGTACATAAACCCCTACTTGTGC
CAAAAGGTAAGCGGGTACAAGTAAGAATTTAAGACTTAACCGTTACATACTCATGAGTCC
AGATATACCAAGCACTCAGCACCTCTGCAAGTCACATGTTTAATACAGTAAGAGCCCAGCA
TCAGTTGATTTCTAAA----------
>Fiji14 (Fiji Islands)
----------TACAACTCAACACCTG
TTTTGTAGCATTATTCCATAAGATTGCACTTCTACGTGCATATATGTATATTCACCATGAAT
CTATATTAACCATATCATGGATATTCGAGTACATGATTTCAAGCTTCAACTAATTCTAAAAT
TAA-GGGCATACTTGTTTTAAAGCATTCACCTAAATAAAAGTACATAAACCCCTACATGTGT
CAAAAGGTAAGCAAGGACAAGTAAAAATTTAAGACTCAACCGTTACATACTCACGAGTCC
AGATATACCAAGTATTCAGTACCTCTGCAAGTCACATGTTTAATACAGTAAGAGCCCAGCA
TCAGTTGATTTCTAAA----------
>Fiji18 (Fiji Islands)
----------TACAACTCAACACCTG
TTTTGTAGCATTATTTCATAAGCTTGCACTTCTACGTGCATATATGTATATTCACCATGAAT
CTATATTAACCATATCATGGATATTCGAGTACATGATTTCAAGCTTTAACTAATCTTAAAAT
CAA-GGACATACTTGTTTCAAAGCATTTATCTAGATAAAAGTACATAAACCCCCACATGTG
TCAAAAGGTAAGCAGGCACAAGTAAAAATTTAAGATTCAACCGTTACATACTCACAAGTCC
AGATATACCAAGTATTCAATACCTCTGCAAGTCACATGTTTAATACAGTAAGAGCCCAGCA
TCAGTTGATTTCTAAA----------

Fig. S1 *Dascyllus abudafur*. Individuals no Europa 1-15 from Europa Island, Mozambique Channel, 06 April 2011 (WJC).

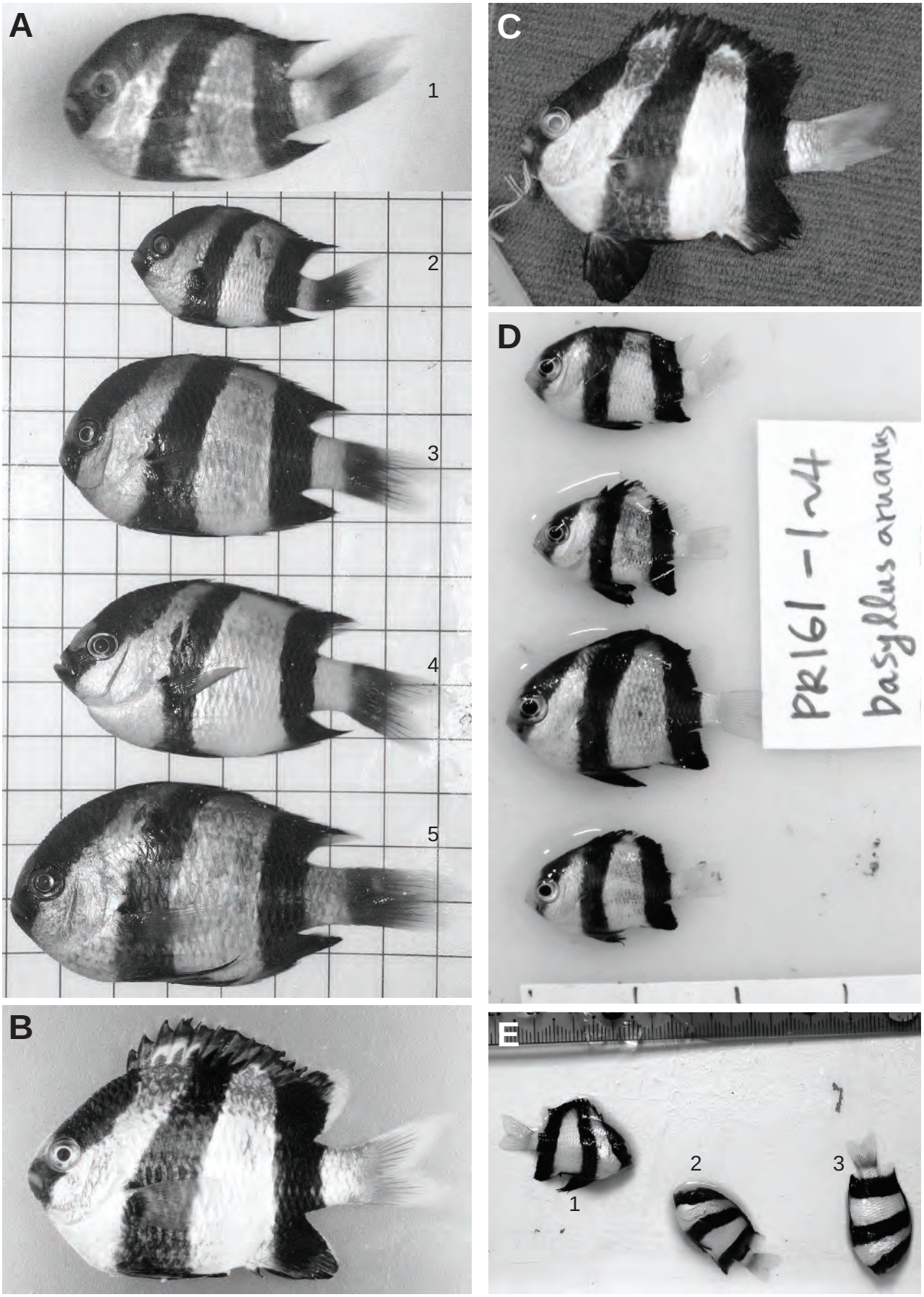

Fig. S2 Humbug damselfish. **A** *Dascyllus abudafur*, individuals nos. JdN 1-5 from Juan de Nova Island, Mozambique Channel, 11-12 April 2011 (PB, WJC). **B** *Dascyllus aruanus*, individual no. 042913-06 from Dongsha, South China Sea, 29 April 2013 (WJC). **C** *Dascyllus aruanus*, individual no. WJC2243 from Dongsha, South China Sea, 09 May 2013 (WJC). **D** *Dascyllus aruanus*, individuals nos. PR161-1 to 4 from Madang, Bismarck Sea, Papua New Guinea, December 2012 (WJC). **E** *Dascyllus abudafur*, individuals nos. Bali 1-3 from Singaraja, Bali Island, April 2014 (PB, AS).

Fig. S3 *Dascyllus aruanus*. Individuals nos. Fam 1-33 from Pulau Fam, Raja Ampat, West Papua, 02 December 2007 (PB).

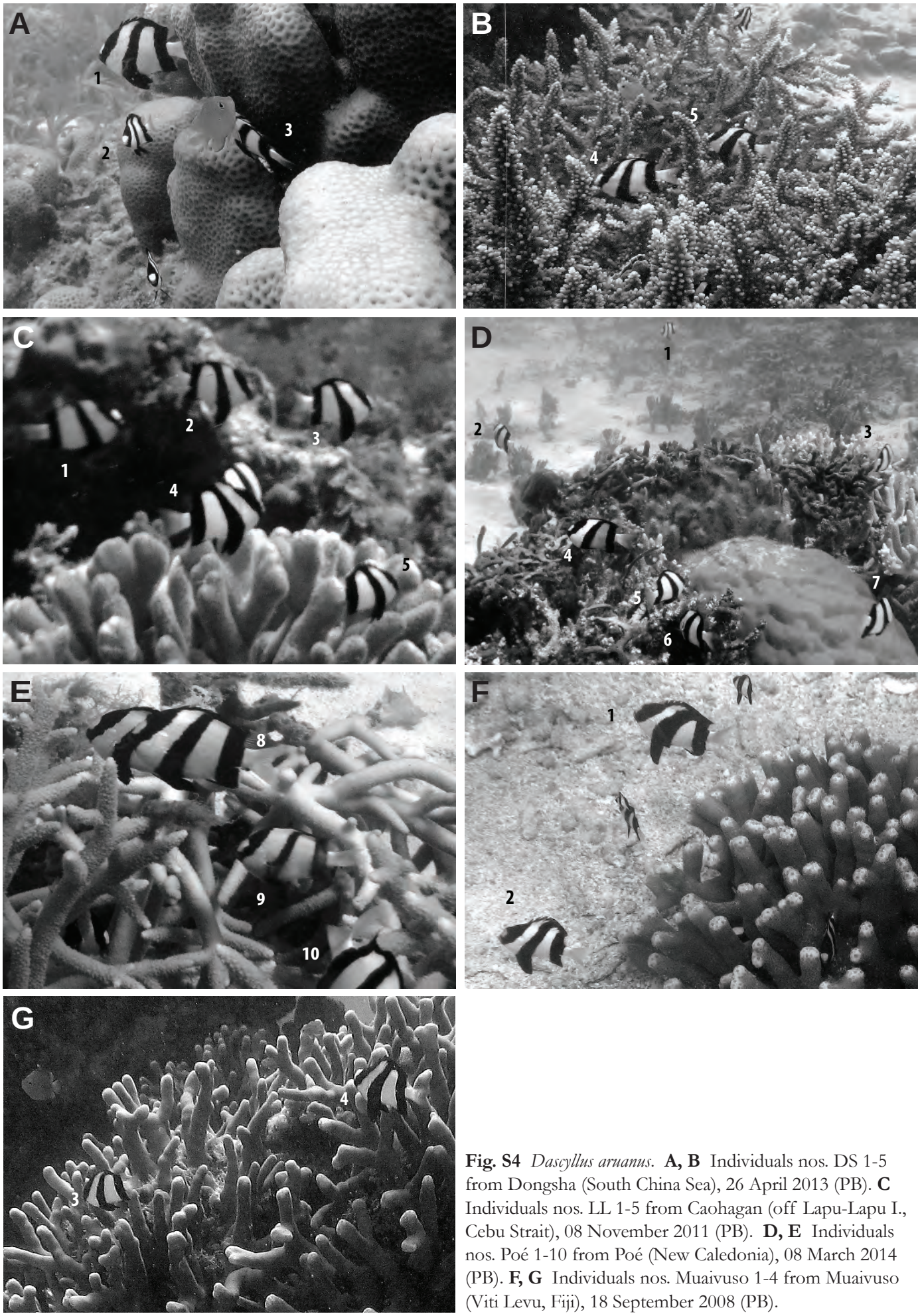

Fig. S4 *Dascyllus aruanus*. **A, B** Individuals nos. DS 1-5 from Dongsha (South China Sea), 26 April 2013 (PB). **C** Individuals nos. LL 1-5 from Caohagan (off Lapu-Lapu I., Cebu Strait), 08 November 2011 (PB). **D, E** Individuals nos. Poé 1-10 from Poé (New Caledonia), 08 March 2014 (PB). **F, G** Individuals nos. Muaivuso 1-4 from Muaivuso (Viti Levu, Fiji), 18 September 2008 (PB).

Fig. S5 *Dascyllus aruanus*. Individuals nos. SWLagoon 1-10 from the southwestern lagon of New Caledonia. **A** 14 March 2008 (PB). **B** 07 April 2008 (PB).

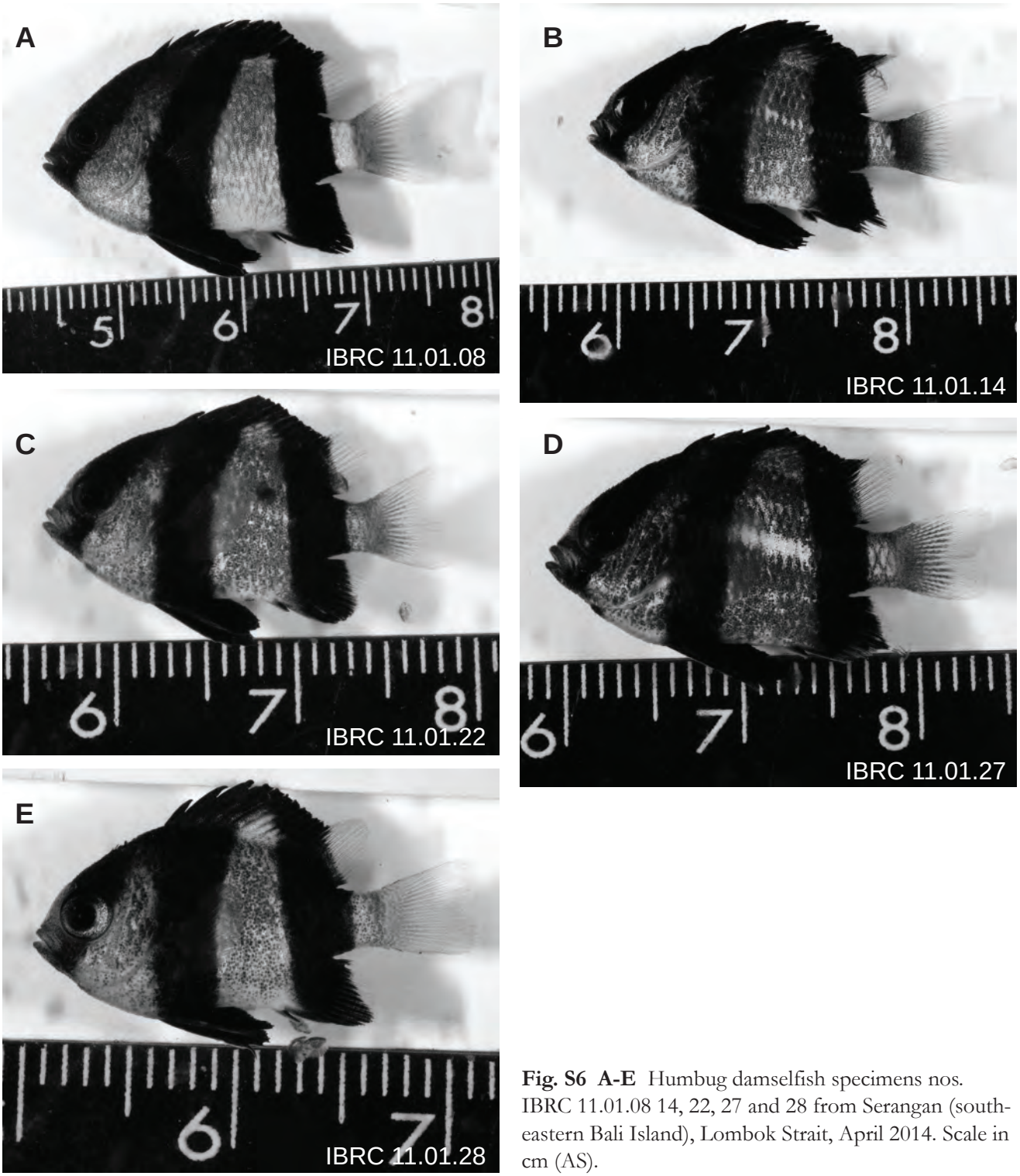

Fig. S6 A-E Humbug damselfish specimens nos. IBRC 11.01.08 14, 22, 27 and 28 from Serangan (south-eastern Bali Island), Lombok Strait, April 2014. Scale in cm (AS).

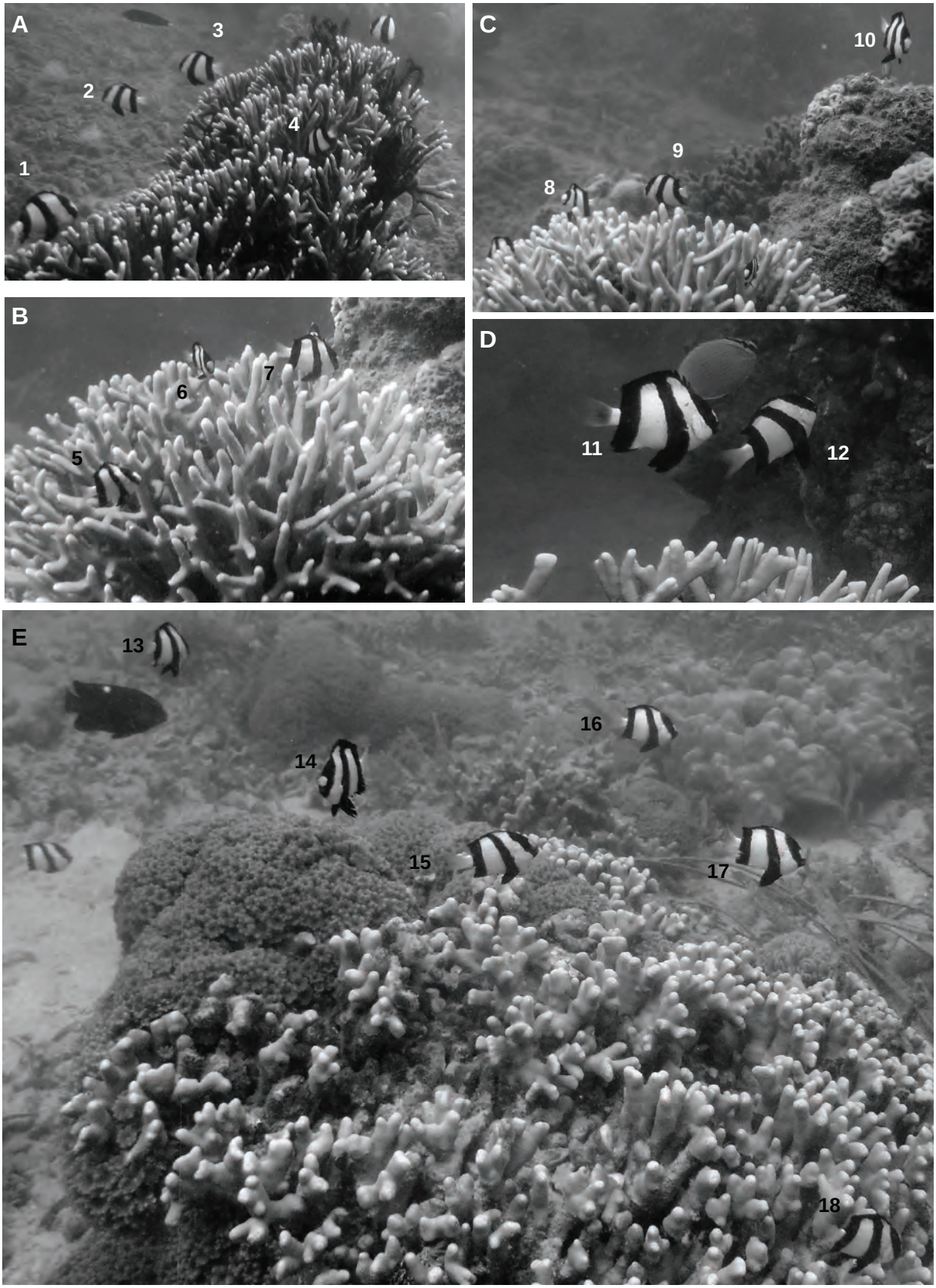

Fig. S7 Humbug damselfish, *Dascyllus abudafur* and *D. aruanus*. Individuals nos. Lombok 1-18 examined for pigmentation patterns (PB). **A-D** Individuals sighted on the fringing reef at the southern tip of Gili Gede (08°46'S 115°56'E), 06 August 2014. **E** Individuals sighted on the reef off southeastern Gili Layar (08°44'S 115°55'E), 07 August 2014.