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Two Distinct Ranid Frog Lineages (Anura: *Hylarana*) from Halmahera, Northern Moluccas, with the Description of a New Species

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Abstract: We revise the systematics of the ranid frogs of the genus *Hylarana* occurring on Halmahera Island, Northern Moluccas of eastern Indonesia based on molecular and morphological data. Our results show that two distinct species each being nested within two distinct clades (hereafter *papua* clade and *celebensis* clade) exist on the island. One corresponds to *H. moluccana* (*celebensis* clade) and the other one to an unnamed species (*papua* clade) that we describe herein. The new species is genetically distinct from all congeners of the *papua* clade by *p* distances ranging from 6.9% to 11.5% on the 16S rRNA gene. Morphologically, the new species can be distinguished from all congeners by having the following combination of characteristics: A large species with adult males reaching a maximum snout-to-vent length (SVL) 67.3 mm; vomerine teeth in two oblique rows with narrow interodontophore distance; snout rounded dorsally; dorsum with few scattered cone-shaped tubercles that are black with white tips; distinct skin folds (ridges) on the dorsal side of the thigh coinciding with dark brown cross bars; a marbled pattern on the ventral side of thigh and yellowish groin. Here we provide a redescription for *H. moluccana* based on its lectotype from Ternate. We also provide new occurrence records for *H. daemeli*, *H. volkerjane*, and *H. arfaki* from the western part of mainland Papua. Furthermore, we demonstrated that *H. celebensis* harbors multiple mtDNA lineages suggestive of cryptic diversity within the *celebensis* clade. The occurrence of two distinct species from Halmahera calls for further research on the biogeographic history of *Hylarana* in Northern Moluccas.

Key words: Halmahera; *Hylarana*; Northern Moluccas; *Papurana*; Ranidae

The frog genus *Papurana* was proposed by Lesson (1831) and Dubois (1992) described it as a subgeneric level of *Hylarana*, with the type species of *Rana papua* Lesson 1829 from "Offak Bay" (now Fak Fak Bay) on the north coast of Waigeo Island (Lesson 1829, 1831; Dubois 1992; Frost 2023). Recent molecular studies recognized *Papurana* as monophyletic and also treated the clade at the genus level (Oliver et al. 2015; Reilly et al. 2022). *Papurana* consists of 17 nominal species that are distributed in Sulawesi, Lesser Sunda Islands, Moluccas, and Australo-Papua (Reilly et al. 2022), of which 14 have been included in recent molecular phylogenies (Reilly et al. 2022; Portik et al. 2023). Therefore, three nominal species within *Papurana* have their phylogenetic positions that remain unknown: *H. moluccana*, *H. novaeguineae*, and *H. grisea*.

The Northern Moluccas consists of several islands (e.g., Halmahera, Ternate, Tidore, Obi, Bacan, and Morotai). Halmahera is a "K"-shaped Island and the largest island in Northern Moluccas. The sole representative of *Papurana* in Northern Moluccas is *H. moluccana*, and it is distributed across the islands of Ternate, Halmahera, and Bacan (Frost 2023). *Hylarana moluccana* was described by Boettger (1895) based on two specimens from Ternate and Halmahera Islands, hence syntypes. One of the syntypes (SMF 6562) was mistakenly identified by Mertens (1967) as a holotype. This species has long been considered a junior synonym of *Rana varians* by Boulenger (1920) and *R. papua* by van Kampen (1923) until the revision of Frost et al. (2006). Furthermore, Mertens (1967) placed it as a subspecies of *R. papua*. In their molecular analyses, Reilly et al. (2022) considered *H. papua* as a valid taxon and retained the status of *H. moluccana* as a valid taxon.

During the examination of specimens in the collection of the Museum Zoologicum Bogoriense (MZB), we found two phenotypes of *Hylarana* from Halmahera Island. One of the phenotypes was provisionally identified as *H. moluccana*, and the second one was found very similar to *H. papua*, which is considered endemic to mainland Papua. Those individuals were initially identified as *H. grisea*, which was originally described from Mt. Went (elevation of 1300 m above sea level [a.s.l.]) in Central Papua. However, we noted the peculiarity of finding Central Papuan species on Halmahera Island. Therefore, here we sought to compare those species (1) *H. moluccana* and (2) *H.* cf. *papua* to confirm their identity.

Molecular and morphological analyses confirmed that the latter represents a distinct unnamed species belonging to the *papua* clade (group), which we describe as a new species herein.

Materials and Methods

For this study, we collected morphological, genetic, and distribution data for *Hylarana* (focusing on members of the *papua* clade), which we compared to publicly available data. We compared the name-bearing type specimens of the species (including synonyms) occurring on Moluccas, directly with

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available specimens and also with the original descriptions of other known congeners. We used the same morphological and morphometric characters used in recent descriptions of the genus *Hylarana* (e.g., Günther 2003; Kraus and Allison 2007).

Tissue Collection and Specimen Examination

We used preserved tissue samples (liver biopsies) stored in 0.6 ml Tris/Saline/EDTA (STE) buffer containing: 10 mM Tris/HCl, 100 mM NaCl. and 1 mM Ethylenediaminetetraacetic acid (EDTA); pH 8.0 (following Matsui et al. 2010), and the corresponding ethanol-preserved specimens were deposited at the herpetological collection at the MZB for DNA analysis. We also examined specimens/photographs (Appendix) from the Naturmuseum Senckenber, Frankfurt am Main, Germany (SMF); University of Papua New Guinea, Port Moresby, Papua New Guinea (UPNG); Museum für Naturkunde, Berlin, Germany (ZMB); Museum acronyms are those of Sabaj (2020). We obtained morphological, morphometric, and meristic data for species comparisons, and distribution data from examined specimens, as well as from published literature.

Morphometric Character Data

Measurements, terminology, and abbreviations follow Kraus and Allison (2007) with modifications. We measured the following characters with a Mitutoyo digital calliper and AmScope SM-1BZ-RL (10–90×; United Scope LLC) dissecting microscope: snout–vent length (SVL, measured from tip of snout to posterior margin of vent); head length (HL, distance between posterior edge of mandible and tip of snout); head width (HW, at widest point, typically at the level of posterior edge of mandible); head height (HH, maximum depth of the head at the level of posterior edge of mandible); internarial distance (IN, distance between anterior edges of external nares); snout length (SL, distance from anterior corner of eye to tip of snout); eye-nostril distance (EN, distance from anterior corner of eyes to center of nares); tympanum–eye length (TE, distance between anterior-most margin of tympanum and posterior corner of eye); eye–eye distance (EE, transverse distance between the anterior corners of the eyes); upper eyelid width (UEW, the greatest width of the upper eyelids); eye diameter (ED, maximum diameter of eye); tympanum diameter (TD, horizontal tympanum diameter); upper arm length (UAL, distance between the axilla and the outer surface of flexed elbow); forearm length (FAL, distance between outer surface of flexed elbow and metacarpal fold); finger length (F, distance from the base of metacarpal fold to the tip of each finger disc); femur length (FL, distance between groin and outer surface of flexed knee); tibia length (TL, distance between outer surface of flexed knee and heel); tarsal length (TaL, distance between heel and metatarsal fold); toe length (T, distance from base of metacarpal fold to tip of each toe disc). Maturity in males was determined by the presence of mature testes or nuptial pads and vocal slits; maturity in females was based on the presence of ovaries.

Morphometric Analysis

Statistically informative tests could not be performed on separate sexes because of the small sample sizes of females from the different phenotypes. However, we performed tests separately for adult males and both sexes together. Juveniles were excluded to avoid allometric bias. Boxplots were generated for SVL, HL, and TL to visualize the range, mean, and median between the different phenotypes. We performed separate Kruskal–Wallis one-way analysis of variance tests on the SVL, HL, and TL to detect any significant difference between *H. moluccana* ($n = 66$; males = 44, females = 22) and *H.* cf. *papua* ($n = 36$; males = 33, females = 3) on Halmahera Island. We used this test because of the small sample size (Zar 2010). Variation in adult size was normalized using the following equation: $\log X_{adj} = \log(X) - \beta[\log(SVL) - \log(SVL_{mean})]$, where X_{adj} = adjusted value; X = measured value; β = unstandardized regression coefficient for each phenotype; and SVL_{mean} = overall average SVL of all phenotypes (Lleonart et al. 2000; Chan and Grismer 2022) prior to multivariate analyses on 18 morphometric characters, SVL, HL, HW, HH, IN, SL, EN, EE, ED, TD, FAL, F4, FL, TL, TaL, T3, T4, and T5.

The scaled morphometric characters were treated as the dependent variable and the phenotypes as the predictor variable for multivariate analysis. Multivariate analysis was conducted using principal component analysis (PCA) to reduce the highly correlated multidimensional data matrix into a few uncorrelated variables, that is, principal components (PCs). We used the princomp function in the R statistical software program (v4.0.4; R Core Team 2021). A biplot of the first two PC scores were used to examine the degree of relative influence of each morphometric character on the data set. All statistical analyses were conducted using the R statistical software program (v4.0.4; R Core Team 2021).

Genetic Data

Total genomic DNA was extracted from the tissue samples using the standard phenol-chloroform method (Sambrook et al. 1989). For molecular-genetic analyses, we used fragments of 16S rRNA using primers H3056 and L2606 (Hedges et al. 1993). We used standard PCR protocols with annealing temperature and elongation time of 55°C and 30 s. Detailed protocols, reagents, and polymerase chain reaction (PCR) conditions followed Hamidy et al. (2018) and Munir et al. (2018). We conducted cycle sequencing reactions and DNA sequencing using BigDye® Terminator (1st Base Asia). The PCR products were sequenced in both directions using the same primers used in amplification.

A total of 34 new sequences of 16S (~500 base pairs [bp]) were generated for this work and deposited in GenBank. They were combined with available data from previously published studies (Supplemental Table S1, available online; Chen et al. 2005; Frost et al. 2006; Gawor et al. 2009; Oliver et al. 2015; Chan et al. 2020; Reilly et al. 2022). Except for *H. grisea*, for which there are no available genetic data, all other nominal species of the subgenus *Papurana* were included in the phylogenetic analysis. *Abavorana luctuosa* was used as outgroups (Zainudin et al. 2008). The sequences were aligned in MEGA 7 (Kumar et al. 2015) using the Muscle algorithm (Thompson et al. 2005) with default settings. We obtained a 417-bp-long alignment, comprising 256 conserved sites, 157 variable sites, and 129 parsimony-informative sites.

Phylogenetic trees were constructed using Bayesian inferences (BI) and maximum likelihood (ML). We used MrBayes v3.2.7 (Huelsenbeck and Ronquist 2001) for the

BI tree and IQ-Tree (Trifinopoulos et al. 2016) for the ML tree. BI analyses were undertaken using the general time-reversible (GTR) model with parameter (GTR Gamma) as the best evolution model selected by Kakusan 3 (Tanabe 2007). BI analysis was performed with mcmc ngen = 10,000,000 generations, print freq = 1000, sample freq = 1000, and mcmc diagn freq = 100,000, and a consensus topology was inferred after discarding the first 2500 trees as burn-in. In BI analysis, the Bayesian posterior probabilities (BPP) support value of 0.95 or more was considered as significant (Leaché and Reeder 2002). In the ML analysis, the maximum likelihood bootstrap support (MLBS) using the Ultrafast Bootstrap (UFBoot) support value ≥95% and SH-aLRT support value ≥80% were considered reliable (Guindon et al. 2010). We also computed pairwise comparisons of uncorrected sequence divergence (*p* distance) for the partial sequences of the 16S rRNA gene using MEGA 7 (Kumar et al. 2015).

We estimated species delimitation hypotheses using a method that builds and ranks species partitions from single locus sequence alignments called Assemble Species by Automatic Partitioning (ASAP; Puillandre et al. 2021) and also using a Bayesian Poisson Tree Processes (bPTP) model to infer putative species boundaries on the obtained ML tree (Zhang et al. 2013). The ASAP was performed with 15 partitions. We used the substitution model of Jukes–Cantor (JC69) to compute the distance (Jukes and Cantor 1969).

RESULTS

Morphometric Analyses

Based on the examination of 102 specimens of *Hylarana* from Halmahera Island (Fig. 1), 17 of 18 morphometric ratio comparisons showed significant differences ($P < 0.05$) between *H. moluccana* and *H.* cf. *papua*. Kruskal–Wallis tests of the SVL, head HL, and tibia TL lengths showed that these body size metrics significantly separate the two putative species: Longer head (HL, $\chi^2 = 45.95$, $P < 0.001$), body (SVL, $\chi^2 = 20.23$, $P < 0.001$), and tibia (TL, $\chi^2 = 22.82$, $P < 0.001$) of *H.* cf. *papua* indicated a relatively elongated and larger body size with longer limbs than that of *H. moluccana* on Halmahera Island (Fig. 2A–C).

PCA for males showed distinct overall differences in normalized morphometric character data between the two phenotypes, with distinct and nonoverlapping clusters (Fig. 2D; Table S2). However, when both sexes are considered altogether the two phenotypes slightly overlap along the first two PCs. Principal components 1 and 2 collectively explained over 90% of the variation in the morphometric data matrix when only males are considered as well as when both sexes are analyzed together (Fig. 2E; Table S3). Scaled morphometric SVL loaded negatively with PC2, whereas all other major characters loaded positively with both PCs.

Despite limited sampling of females in *H.* cf. *papua*, a significant sexual dimorphism is seen in body size (SVL; see Fig. 2A–C) among both *H.* cf. *papua* (male SVL = 59.5–67.3 mm, $n = 33$, $\bar{X} \pm SD = 62.9 \pm 1.9$; female SVL = 64.0–81.0 mm, $n = 3$, $\bar{X} \pm SD = 74.7 \pm 1.3$) and *H. moluccana* (male SVL = 47.5–65.0 mm, $n = 44$, $\bar{X} \pm SD = 53.5 \pm 4.0$; female SVL = 53.0–77.0 mm, $n = 22$, $\bar{X} \pm SD = 64.7 \pm 5.8$).

We present diagnostic morphological (morphometric and meristic) data taken for each of the above two phenotypes in Table 1 and compared them to the species forming the Papuan clade of *Hylarana* in Wallacea and Sahul Shelf (see Table 1; Boulenger 1882, 1897, 1920; Boettger 1895; van Kampen 1909; 1923; Roux 1911; Loveridge 1948; Tyler 1963; Menzies 1987; Günther 2003; Kraus and Allison 2007; Donnellan 2010; this study). After a morphological comparison between voucher specimens and respective type specimens, we concluded that one of the phenotypes corresponds to *H. moluccana* and that the other (*H.* cf. *papua*) required an integrative systematic approach.

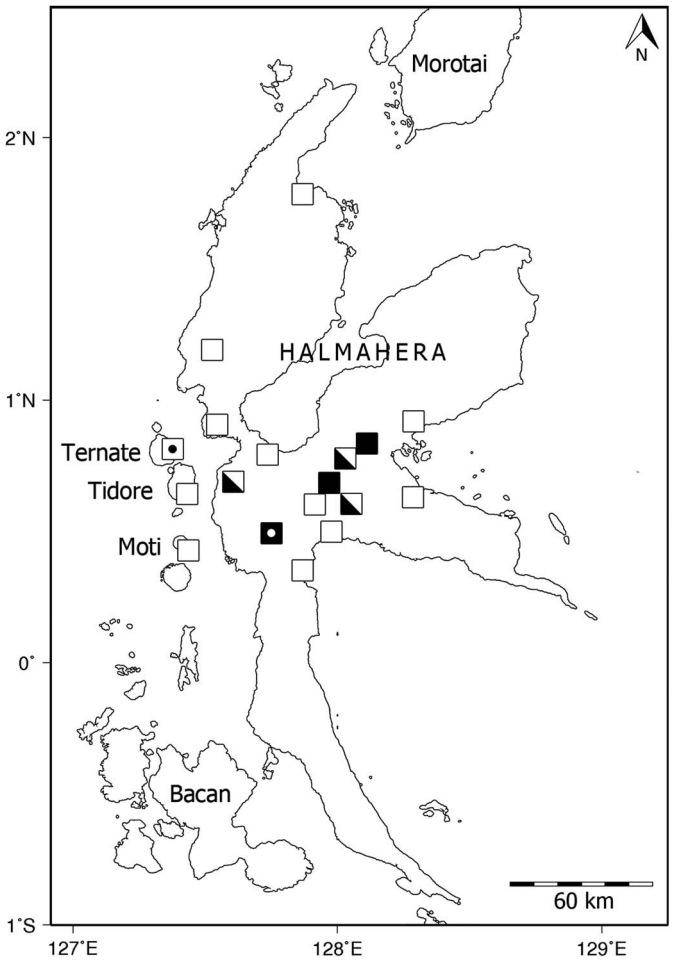

FIG. 1.—Current distribution map showing the collection/observation localities of the two *Hylarana* putative species on Halmahera Island in filled (*H.* cf. *papua*, the new species) and open squares (*H. moluccana*), sympatric localities filled partially. The symbols with dot in the middle represent the type locality of respective species.

Molecular phylogenetic and genetic variability.—The molecular analyses of 16S rRNA using ML and BI (Fig. 3) showed two clades within the subgenus *Papurana* sensu Reilly et al. (2022). The subgenera *Papurana* and *Hylarana* are paraphyletic. Two poorly supported clades are recovered each formed by species of both subgenera. Therefore, for the clarity of presenting our results, we ignore the subgeneric classification. The two identified clades (or groups) in our phylogenetic tree are hereafter considered as *celebensis* clade (group) and the *papua* clade (group).

The entire clade is formed by species with genetic distances ranging from 1.8–2.2% (*H. papua* and *H. kreffti*) to 14.0–15.1% (*H. elberti* and *H. celebensis*; Table S3). Both

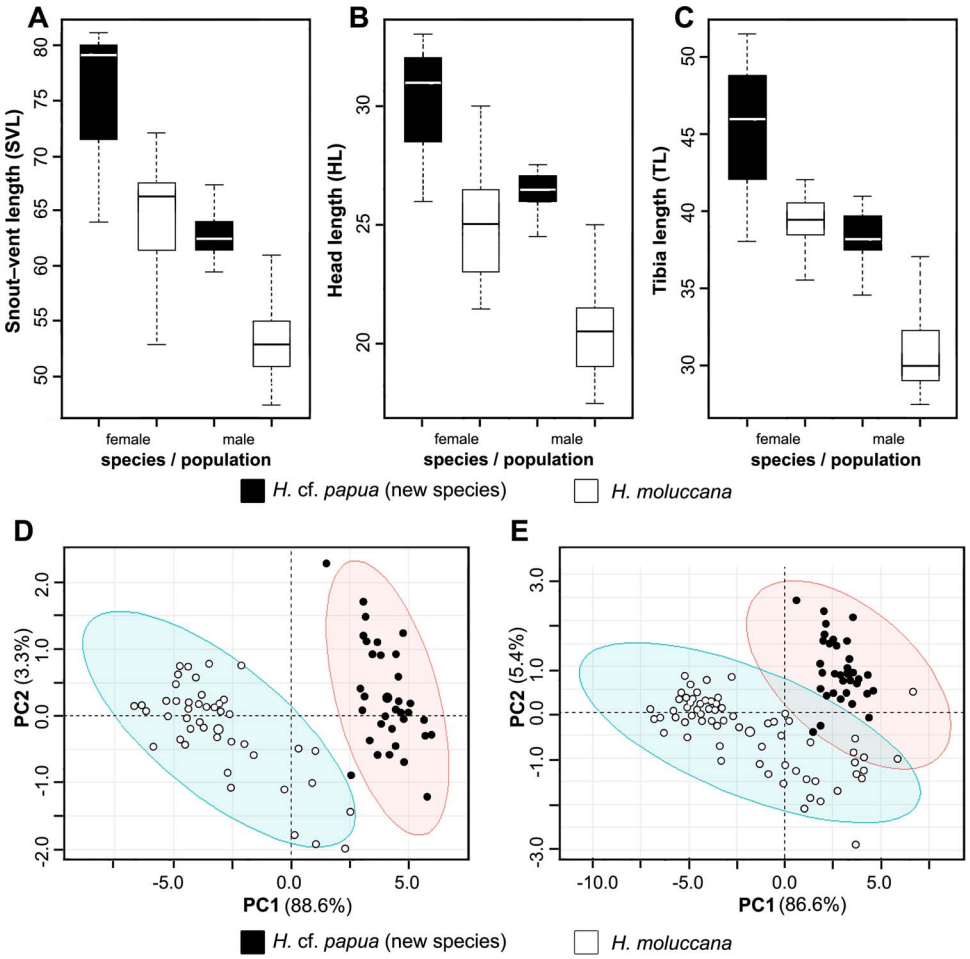

FIG. 2.—Boxplots of (A) snout–vent length, (B) head length, and (C) tibia length indicating differences between *Hylarana* species/populations in Halmahera (note that sexes were separated). PCA: biplot of morphometric variation among, (D) males separately, and (E) when both sexes combined together, between *Hylarana* species/populations in Halmahera. Each point represents an individual specimen, and the relative distance between two points is equivalent to the amount of dissimilarity.

ML and BI provided the same topologies (Fig. 3), but the ASAP and bPTP analyses provided different partitioning with 21–25 candidate species in the *papua* clade, then 3–5 candidate species in *celebensis* clade (Fig. 3).

Our phylogenetic trees also revealed that (1) *H. novaeguinea* from West Papua and (2) *H. celebensis* from Gorontalo (northern Sulawesi) are two distinct evolutionary lineages that are related to (1) *H. papua* + *H. kreffti* in Papua and (2) *H. moluccana* respectively. *Hylarana* cf. *papua* is in fact distantly related to all the species of *Hylarana* of the *papua* clade with a distance of 5.3–8.8%. This phenotype is thus described below as a new species.

TAXONOMY

Hylarana ***nigroverrucosa*** Wiradarma, Amarasinghe, Farajallah, Fouquet, Riyanto, Arida and Hamidy sp. nov.
(Tables 1, 2; Figs. 4A, B, 5, 6, S3)

***Rana grisea*—**Setiadi and Hamidy (2006).

Holotype.—MZB Amph. 12774, an adult male from Central Lolobata (0°29′38.75″N, 127°44′48.45″E; datum = WGS84 in all cases; 450 m a.s.l.), Halmahera, North Maluku, eastern Indonesia, collected by A. Hamidy in 2006.

Paratypes (*n* = 35; 32 males, 3 females).—MZB Amph. 12775–77, 12779–92, adult males, other collection details the same as holotype; MZB Amph. 16027–32, adult males from Gumoang River (0°48′45.55″N, 128°4′25.90″E), West Tofu, East Halmahera, MZB Amph. 16033, adult male from Mt. Boki Mekot (0°36′39.53″N, 128°2′26.19″E), Weda, Central Halmahera, and MZB Amph. 16035–38, adult males from Mt. Tofu (0°48′7.59″N, 128°1′56.70″E), Blewen, Central Halmahera, collected by Mulyadi in 2010; MZB Amph. 16560, 16562–64, adult males from Kaorahai (0°39′45.82″N, 127°58′45.73″E), East Halmahera, collected by A. Riyanto and Mulyadi in 2010; MZB Amph. 12778, adult female, other collection details the same as holotype; MZB Amph. 12794, an adult female from Sofifi (0°41′23.36″N, 127°35′45.93″E), north Oba, Tidore, West Halmahera, collected by A. Hamidy in 2006; MZB Amph. 16561, an adult female from Kaorahai (0°39′45.82″N, 127°58′45.73″E), East Halmahera, collected by A. Riyanto and Mulyadi in 2010.

Diagnosis.—*Hylarana nigroverrucosa* sp. nov. is distinguished from other congeners of (subgenus *Papurana*) by having the following combination of characters: A medium-sized (SVL = 67.3–81.0 mm); vomerine teeth in two oblique rows which protrude posteriorly; males with an external paired lateral vocal sac and humeral gland; an acute snout in

TABLE 1.—Selected characters to diagnose *H. nigroverrucosa* and the other species of the subgenus *Papurana* (sensu Reilly et al. 2022): (1) maximum SVL (mm) of males; (2) TL/SVL%; (3) EN/IN%; (4) TD/ED%; (5) HL/HW%; (6) snout shape at lateral view, acute/angular (ac), rounded (ro), truncate (tc); (7) canthus rostralis at lateral view, rounded (ro) or sharp (sh); (8) loreal, shallowly concave (sc), moderately concave (mc), deeply concave; (9) dark loreal stripe absent (a) or present (p); (10) dark postocular mask absent (a) or present (p); (11) black pustules on dorsum in adults, absent (a) or present (p); (12) dorsum texture, smooth (sm) or shagreened/finely granular (sg); (13) substances on dorsum, asperities (as), tubercles and rounded ridges (tr), warty (wr) or plain (pl); (14) skin folds/ridges across the thigh absent (a) or present (p); (15) dorsolateral fold, absent/indistinct (a) or if present: continuous (c) or discontinued/broken (d), and narrow (n) or wide (w); (16) humeral gland on male absent (a) or present (p); (17) paired black spots on chest absent (a) or present (p); (18) mottled/marbled pattern on throat absent (a) or present (p); (19) vocal sac on male absent (a) or present (p); — = not evaluated.

Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
H. arfaki	92.8	60–61	108–114	52–54	102–103	ro	ro	dc	a	a	a	sg	tr	a	a	p	a	p	p
H. aurata	77.3	62–65	100–115	45–59	108–136	ro	sh	mc	p	p	p	sm	pl	a	c/n	p	p	p	a
H. celebensis	51.0	55	118–143	75	106	ro	sh	dc	p	p	a	sg	wr	a	c/w	a	a	p	a
H. daemeli	64.2	49–61	92–121	67–76	98–115	ro	sh	sc	a	a	a	sm/sg	wr	a	d/n	p	p	p	a
H. elberti	48.0	66	96	64	100	ro	sh	sc	p	p	a	sm	pl	a	c/n	p	p	a	a
H. florensis	66.0	56–57	100	63–75	100–117	ro	sh	mc	p	p	a	sm	as	a	c/n	p	a	a	a
H. garritor	71.9	48–61	93–116	71	121	ac	sh	sc	a	a	a	sm	wr	a	c/n	a	a	p	p
H. grisea	75.0	60–66	78–85	47–56	100–106	tc	sh	mc	p	p	a	sm	pl	a	c/w	p	a	a	p
H. jimiensis	93.0	56–57	96–97	34–36	89–90	tc	ro	dc	a	a	a	sg	tr	a	a	p	a	p	p
H. kreffti	60.0	53–55	88–93	56–57	113–118	ro	sh	mc	p	p	a	sm	pl	a	c/w	p	a	p	p
H. milneana	55.9	55–70	100–121	84	110–117	ro	sh	sc	p	p	a	sm	pl	p	c/w	—	p	a	a
H. moluccana	65.0	52–66	87–111	88–100	113–136	ro	sh	mc	p	p	a	sm/sg	pl	a	c/n	a	a	p	a
H. nigroverrucosa sp. nov.	67.3	58–64	91–107	80–87	114–130	ac	sh	dc	p	a	p	sm/sg	pl	p	c/n	p	a	p	p
H. novaeguineae	36.0	50–51	82–86	65–68	119–128	ro	sh	dc	p	p	a	sg	wr	a	c/w	p	p	—	—
H. papua	66.0	44–58	84–110	61–76	123–128	ro	ro	mc	p	p	a	sm/sg	wr	a	c/n	p	p	p	p
H. supragrisea	84.4	53–63	86–133	54	100–120	tc	sh	dc	p	p	a	sm	pl	a	c/n	p	p	p	p
H. volkerjane	76.2	61–70	105–133	57–71	116–128	ro	sh	dc	p	p	a	sm	pl	p	c/n	p	p	a	a
H. waliesa	76.9	47–57	89–108	66	102–127	ro	sh	mc	a	a	a	sg	as	a	c/w	—	—	p	p

lateral profile; canthus rostralis sharp; smaller sized tympanum, TD/ED = 80–87.5%; dorsum with black warty-pustules on both sexes; a distinct dorsolateral skin folds (ridge) across the dorsal thigh; a dark loreal stripe, but absence of postocular mask; a dorsolateral fold continuous on the body; supernumerary tubercles at the bases of the fingers; the last phalanx of the fourth toe free of webbing.

Comparison.—*Hylarana nigroverrucosa* sp. nov. is closely allied to *H. arfaki* and *H. jimiensis*, but differs from them in its much larger sized tympanum (TD/ED = 52%–54% in *H arfaki* and 34%–36% mm in *H. jimiensis*), having a loreal stripe (absent), dorsal pustules (absent), the dorsolateral skin folds across the thigh (absent), the last phalanx of the fourth toe free of webbing (fully webbed), the snout acute (rounded in *arfaki* and truncate in *H. jimiensis*) in lateral profile, and canthus rostralis sharp (rounded). The new species is also compared with other congeners of *papua* group distributed in the Sahul Shelf; see Table 1.

From *H. novaeguineae* (characters in parentheses), *H. nigroverrucosa* sp. nov. differs in its much larger size (males to 36 mm, females to 43 mm), having skin folds across the thigh (absent), fourth toe free of webbing on the last phalanx only (last two phalanges), snout acute (rounded) in lateral profile, and absence of postocular mask (present). Also, from *H. aurata* and *H. volkerjane*, the new species differs in having dorsal pustules (absent in *H. volkerjane*), vocal sacs (absent) in males, skin folds across the thigh (absent in *H. aurata*), the snout acute (rounded) in the lateral profile, and absence of postocular mask (present). From *H. daemeli* and *H. milneana*, the new species differs in having a loreal stripe (absent in *H. daemeli*), vocal sacs in males (absent), the snout acute (rounded) in lateral profile, dorsolateral fold continuous on the body (broken in *H. daemeli*), and absence of postocular mask (present in *H. milneana*). From *H. garritor*, *H. papua*, and *H. waliesa*, the new species differs in having dorsal pustules (absent), humeral gland (absent in *H. garritor*), a loreal stripe (absent in *H. garritor* and *H. waliesa*), skin folds across the thigh (absent), the snout acute (rounded in *H. papua*, and *H. waliesa*) in lateral profile, canthus rostralis sharp (rounded in *H. papua*), and absence of postocular mask (present in *H. papua*). From *H. grisea* and *H. supragrisea*, the new species differs in having dorsal pustules (absent), supernumerary tubercles at the bases of the fingers (absent in *H. grisea*), skin folds across the thigh (absent), the snout acute (truncate) in lateral profile, and absence of postocular mask (present). From *H. kreffti* (characters in parentheses), *H. nigroverrucosa* sp. nov. differs in having dorsal pustules (absent), vomerine teeth in two oblique rows (in oblique groups or short series), the snout acute (rounded) in the lateral profile, fourth toe free of webbing on the last phalanx only (last two phalanges), and absence of postocular mask (present).

Furthermore, the new species is compared with other congeners of *papua* group distributed in the Lesser Sunda Islands and Wallacea (*celebensis* group); see Table 1. From *H. elberti* and *H. florensis* from the Lesser Sunda, the new species differs in having dorsal pustules (absent), vomerine teeth in two oblique rows (in oblique groups or short series), vocal sacs in males (absent), skin folds across the thigh (absent), the snout acute (rounded) in the lateral profile, fourth toe free of webbing on the last phalanx only (last two phalanges), and absence of postocular mask (present).

The new species is also genetically divergent from its closest congener, *H. jimiensis* distributed on western and central parts of mainland Papua with a *p* distance of 5.3%–5.5% in the 16S rRNA gene.

Description of holotype.—An adult male. Head moderately wide (HW/SVL = 33.1%), as wide as body, and long (HL/SVL = 40.9%, HL/HW = 123.8%); round canthus rostralis, concave loreal region; nostrils slightly vertically compressed, oblique, closer to tip of snout than to eyes, with small white papilla at posteroventral corner of

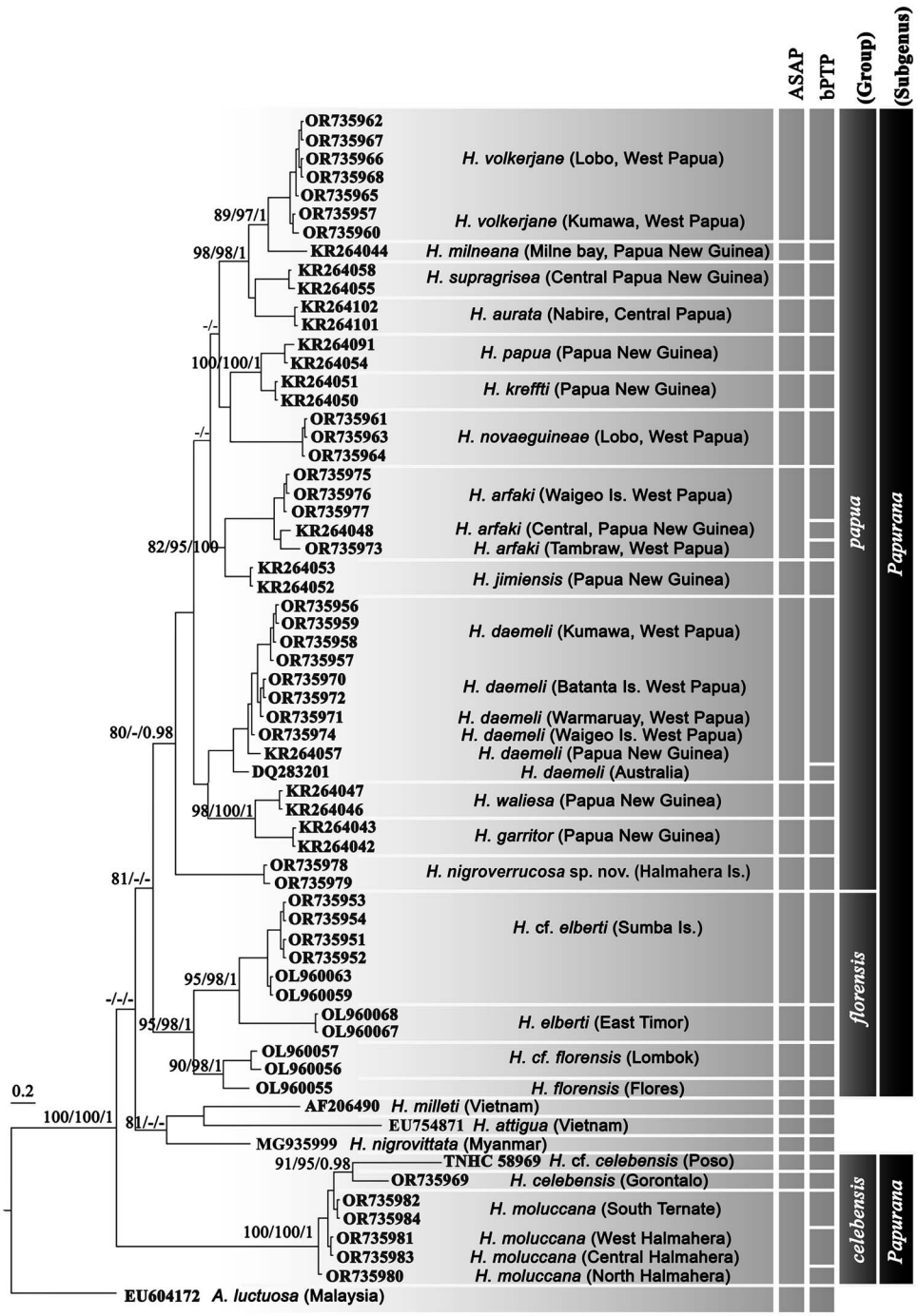

FIG. 3.—Phylogenetic affinities of the members of the subgenus *Papurana* using a ML and BI analysis of the 16S rRNA region; The support node in with considered as reliable in slash series: SH-aLRT ≥ 80%/UFBoots ≥ 95/BI ≥ 0.95.

each; internarial distance almost equal to distance from external naris to eye (EN/IN = 100%, IN/SVL = 9.4%, EN/SVL = 9.4%); snout short (SL/SVL = 16.5%), broadly rounded when viewed from above, top half truncate when viewed from side but lower half bluntly pointed; eyes large (ED/SVL = 11.0%), eyelid slightly wider than interorbital distance; tympanum distinct, horizontal diameter smaller than width of eye (TD/ED = 85.7%), separated from eye by distance less than half tympanum width, vocal slits located near to angle of jaw.

Dorsal surfaces smooth or finely granular, with minute asperities present above groin, shanks, and eyelid, dorsum with few scattered, cone-like, white-tipped, black warty-pustules; lateral surfaces glandular; dorsal surfaces of thighs smooth, thin ridge-like skin folds along the dark cross bars on the thigh. Thick dorsolateral ridge extending from posterior corner of each eyelid to beyond region of the sacral diapophysis but becoming obscure before reaching groin. Ventral surfaces smooth except for posterior third of thighs, which are coarsely granular.

TABLE 2.—Selected morphometric characters (in mm) of *H. nigroverrucosa* sp. nov. holotype and paratype series (see species account for accession data).

	Male ($n = 33$)			Female ($n = 3$)	
		Paratypes		Paratypes	
Character	Holotype MZB 12774	Mean ± SD	Range	Mean ± SD	Range
SVL	63.5	62.9 ± 1.9	59.5–67.3	74.7 ± 9.3	64.0–81.0
IN	6.0	5.9 ± 0.1	5.5–6.0	6.7 ± 0.6	6.0–7.0
EN	6.0	5.8 ± 0.2	5.5–6.0	7.0 ± 0.9	6.0–7.5
SL	10.5	10.2 ± 0.6	9.0–11.0	11.5 ± 1.7	9.5–12.5
ED	7.0	7.0 ± 0.3	6.0–8.0	7.2 ± 1.0	6.0–8.0
EE	5.5	5.4 ± 0.3	4.5–6.0	6.2 ± 1.0	5.0–7.0
UEW	7.0	6.7 ± 0.4	6.0–7.5	7.0 ± 0.9	6.0–7.5
TD	6.0	6.0 ± 0.3	5.0–7.0	6.2 ± 1.0	5.0–7.0
TE	2.0	2.0 ± 0.2	1.4–2.5	2.7 ± 0.6	2.0–3.0
HL	26.0	26.3 ± 0.9	24.5–27.5	30.0 ± 3.6	26.0–33.0
HW	21.0	21.1 ± 0.7	19.5–22.5	24.7 ± 3.8	20.5–28.0
HH	9.0	10.0 ± 0.6	9.0–11.0	10.9 ± 1.2	9.5–11.7
UAL	12.0	12.2 ± 1.0	10.0–14.0	15.5 ± 2.6	12.5–17.0
FAL	11.0	12.0 ± 0.7	10.5–13.0	14.5 ± 2.6	11.5–16.0
Finger I length (F1)	13.0	13.4 ± 0.7	12.0–15.0	16.7 ± 2.4	14.0–18.6
Finger II length (F2)	12.5	12.7 ± 0.7	11.5–14.0	15.0 ± 1.3	13.5–15.7
Finger III length (F3)	18.6	18.5 ± 2.0	17.0–28.5	21.6 ± 3.2	18.0–24.1
Finger IV length (F4)	13.6	15.3 ± 0.7	13.6–16.5	18.2 ± 2.5	15.5–20.3
FL	28.5	28.5 ± 1.4	26.0–32.5	34.9 ± 4.3	30.5–39.0
TL	37.5	38.4 ±1.6	34.5–41.0	45.2 ± 6.8	38.0–51.5
TaL	20.0	20.0 ± 0.9	18.0–22.0	23.7 ± 3.7	20.0–27.5
Toe I length (T1)	10.8	11.4 ± 0.9	9.5–13.5	12.9 ± 1.4	12.0–14.5
Toe II length (T2)	15.1	16.3 ± 1.1	13.0–18.5	19.0 ± 1.8	17.5–21.0
Toe III length (T3)	21.4	23.2 ± 1.3	19.1–26.5	27.2 ± 3.7	23.0–29.7
Toe IV length (T4)	31.0	32.6 ± 1.9	26.5–35.5	37.6 ± 5.6	31.5–42.4
Toe V length (T5)	22.5	24.2 ± 1.5	20.5–27.0	28.4 ± 3.7	24.1–31.0

Fingers unwebbed, relative lengths 3 > 1 > 4 > 2, tips flattened and expanded; F3 and F4 bearing lateral grooves and F1 and F2 with traces of grooves on each side; subarticular tubercles present at bases of fingers and white; subarticular and metacarpal tubercles prominent; outer metacarpal tubercle divided in two; nuptial pads are not developed well. Toes webbed to the base of each disc except on T4, which is webbed to base of penultimate phalanx, relative lengths 4 > 3 > 5 > 2 > 1; toe webbing formula I 0 – 1 $\frac{4}{5}$ II 0 – 2 III 0 – 2 IV 2 – 0 V; tips pointed and bearing discs with circummarginal grooves; subarticular and metatarsal tubercles prominent. Hind legs moderately long (TL/SVL = 59.0%).

Coloration of holotype in preservative.—Dorsum pale chestnut brown, sides with low-contrast pattern of brown clouded over white, gray, or faint yellow; upper lip white, contrasting with surrounding color, extending posteriorly to forearm insertion; dorsolateral ridge slightly darker than remainder of dorsum; dark brown postocular mask present but not clearly demarcated posteriorly from ground color; dark loreal stripe distinct on the upper loreal region, entire face between white lip stripe and canthus pale; side of face same color as dorsal head; tympanum dark chestnut brown.

Ventral surfaces of chin, throat, chest, and abdomen white mottled with light grayish brown; ventral surfaces of vocal sacs lack white and, thus, contrast in appearance with adjacent throat. Thighs and shanks each with three or four dark brown stripes on a lighter grayish brown ground; groin yellow; dark bars on thighs obscure and narrower than intervening pale grayish brown ground color; rear of thighs clouded with dark brown on light brown or straw ground color; center of ventral surfaces of thighs clouded with light grey blotches; and venter white or with faint dark yellow cast, evenly suffused with dark punctations or grey clouding. Soles, palms, and webbing brown.

Variation.—Adult males vary from 59.5 mm to 67.3 mm and adult females from 64.0 mm to 81.0 mm; males have darker body color than females; only males have external vocal sacs; females were collected with mature oocytes. Upper lips might be dusky with a few white blotches, rarely fully white stripe. Our sample comes from different regions of Halmahera Island, and in some populations loreal stripe and postocular mask might be paler, or rarely absent, probably an infraspecific variation; the marbled pattern on the ventral side of thigh (MPVT) spreads over 50% of thigh width, but some males have spread over half width of the thigh.

Etymology.—The specific epithet "*nigroverrucosa*" is a Latin compound adjective (*nigro* + *verrucosa*) in the nominative singular given in feminine, which refers to "black warty" on the dorsum, a distinct distinguish character of the species. English name: Black-warty frog; local (Bahasa Indonesia) name: Katak bintil-hitam.

Natural history.—This species is found in ponds and rivers that flow across primary montane forests and forest fringes. All specimens of the new species were collected less than 1.5 m away from the river bank. By the gut examination of some specimens, the diet is insects. Only males are known to make a calling sound (Setiadi and Hamidy 2006). There is no information about diet and call for this species.

Distribution.—According to Setiadi and Hamidy (2006), *H. nigroverrucosa* sp. nov. (then *R. grisea* sensu lato) has been recorded in East, West, and South Halmahera (Fig. 1). We observed *H. moluccana* in sympatry with the new

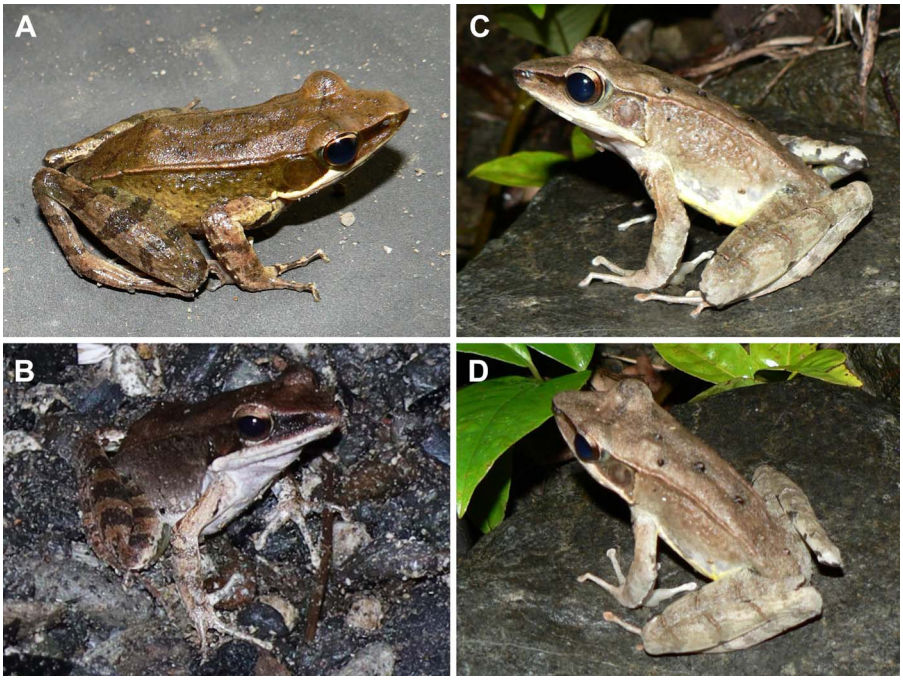

FIG. 4.—*Hylarana moluccana* from (A) Ternate (MZB Amph. 12649) and (B) Halmahera (MZB Amph. 33553); and *H. nigroverrucosa* sp. nov. from Halmahera (C, D) paratype (MZB Amph. 12794). Photos by M.I. Setiadi and M.F. Fauzan.

species at the elevations 50–800 m a.s.l. in Halmahera. This species is likely distributed in surrounding islands like Morotai as well (Setiadi and Hamidy 2006).

Hylarana moluccana (Boettger, 1895)
(Tables 1, 3; Figs. 4B,C, S2–S4, available online)

Rana moluccana—Boettger (1895).
Hylorana* (sic) *molluccana—Deckert (1938).
Rana papua moluccana—Mertens (1967).
Rana* (*Hylarana*) *moluccana—Dubois (1987).
Rana* (*Papurana*) *moluccana—Dubois (1992).
Rana papua—Setiadi and Hamidy (2006); Setiadi et al. (2009).
Sylvirana moluccana—Frost et al. (2006).
Hylarana moluccana—Che et al. (2007); Dubois et al. (2021).
Papurana moluccana—Fei et al. (2010); Oliver et al. (2015); Chan et al. (2020).
Hylarana* (*Papurana*) *moluccana—Reilly et al. (2022).

Lectotype.—SMF 6562 (designated by Mertens 1967, by inference of "holotype"), an adult female from Ternate, Molukken (North Maluku, eastern Indonesia) collected by W. Kükenthal in 1894.

Specimens examined (n = 66; 44 males, 22 females).—See Appendix.

Diagnosis.—*Hylarana moluccana* is distinguished from other congeners of (subgenus *Papurana*) by having the following combination of characters: A medium-sized species of *Hylarana* with adults reaching a maximum SVL = 65.0 mm (males) and 77.0 mm (females), vomerine teeth in two oblique rows that protrude posteriorly beyond the choana line and wide intervomer distance (IVTD); indistinct external paired lateral vocal sac on males; humeral gland are absent; a marbled pattern on the ventral side of thigh (MPVT) spreads less than 50% of thigh width; a somewhat short and rounded snout EN/IN 87.5%–130%; white upper lips; the eye wide as the tympanum, TD/ED 88%–100%; dorsum smooth or somewhat rough with fine granules; dorsolateral fold distinct from supratympanic ridge and continue to sacral; long hind limbs, TL/SVL 51.9%–66.1%; the heel reaches far beyond the tip of the snout when flexed; toe webbing formula I 1 – 2 II 1 – 2 $\frac{1}{2}$ III 0 – 2 $\frac{1}{2}$ IV 2 – 0 V; the species is also genetically divergent from its closest congener, *H. celebensis* distributed on Sulawesi with p distance of 2.4%–5.3% in the 16S rRNA gene.

Description of the species.—Adult males = 47.5–65.0 mm and adult females = 53.0–77.0 mm. Head moderately wide (HW/SVL = 29%–34%), as wide as body, and long (HL/SVL = 34.7–43.1, HL/HW = 113.2%–135.9%); sharp/angular canthus rostralis at lateral view and straight at dorsal view; deeply concave loreal region; nostrils slightly vertically compressed, oblique, closer to tip of snout than to eyes, with small white papilla at posteroventral corner of each; internarial distance almost equal to distance from external naris to eye (EN/IN = 87.5%–111%, IN/SVL = 7.6%–9.5%, EN/SVL = 7.6%–10.4%); snout short (SL/SVL = 12.7%–17.1%), broadly rounded when viewed from above, top half truncate when viewed from side but lower half bluntly pointed; eyes large (ED/SVL = 8.1%–11.2%); Diameter upper eyelid to eye in vary with more than half or more (ULE/ED = 81.8%–111%); tympanum distinct, horizontal diameter smaller than width of eye (TD/ED = 89%–100%), separated from eye by distance less than half tympanum width.

Dorsal surfaces are finely granular, with minute asperities present above groin, on shanks, and on eyelid, in females are more smooth; lateral surfaces with a few glandular; thin dorsolateral fold extending from posterior corner of each eyelid

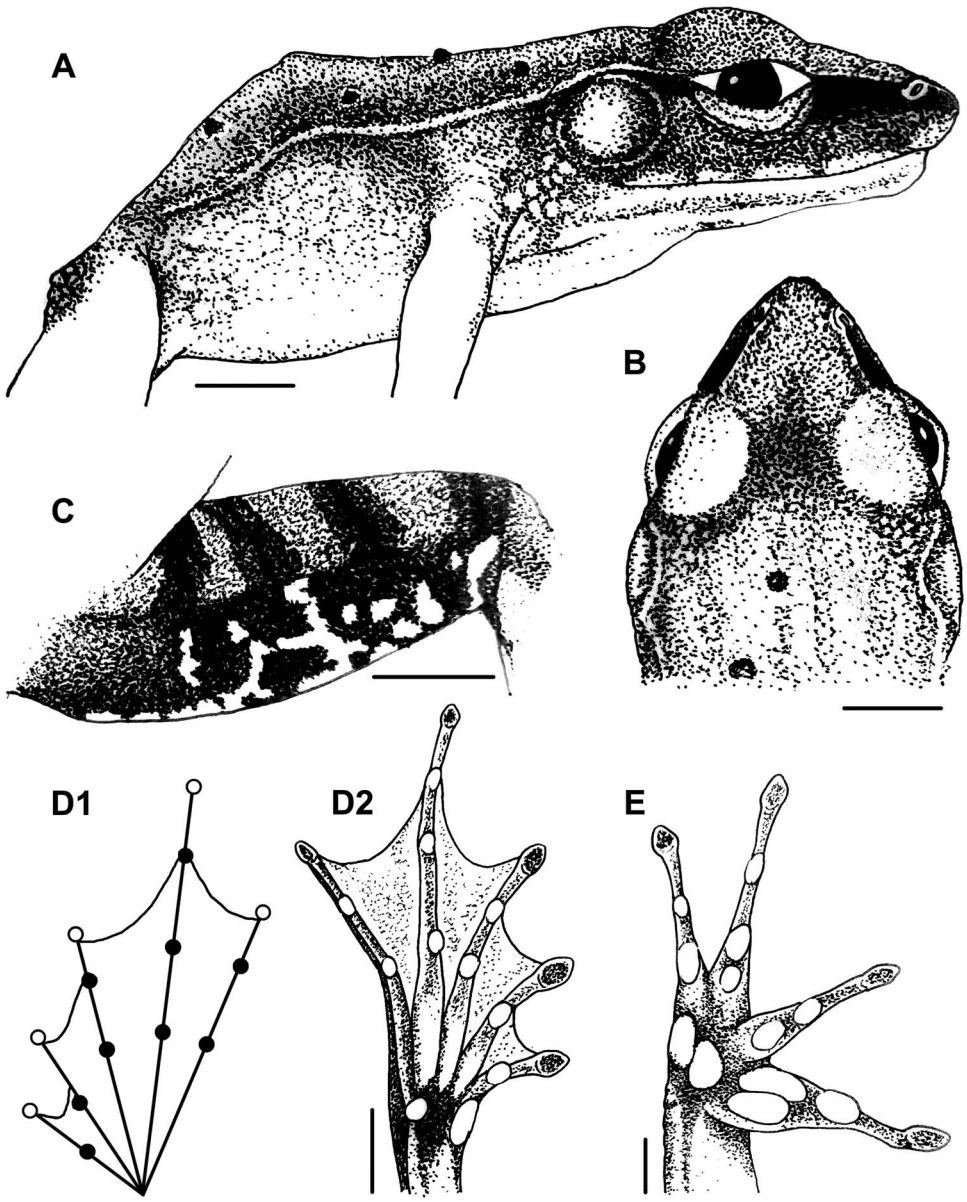

FIG. 5.—*Hylarana nigroverrucosa* sp. nov. holotype (MZB Amph. 12774). (A) Lateral body, (B) dorsum of head, (C) rear of thigh, (D1) schematic illustration of webbing on foot, (D2) foot, and (E) palm. Illustrations by AATA (scale: 5 mm).

to sacral; dorsal surfaces of thighs smooth, no ridge along the dark cross bars on the thigh. Ventral surfaces smooth.

Fingers unwebbed, relative lengths 3 > 4 > 1 > 2, tips flattened and expanded; F3 and F4 bearing lateral grooves and F1 and F2 with traces of grooves on each side; subarticular tubercles present at bases of fingers and white; subarticular and metacarpal tubercles prominent; outer metacarpal tubercle divided in two. Toes webbed to the base of each disc except on T3 and T4, which is webbed half to base of penultimate phalanx, relative lengths of toes 4 > 3 > 5 > 2 > 1; tips pointed and bearing discs with circummarginal grooves; subarticular and metatarsal tubercles prominent. Hind legs moderately long (TL/SVL = 52%–66%).

Coloration of the species in life.—Dorsum color pale chestnut brown, sides with low-contrast pattern of brown clouded over white, gray, or faint yellow; upper lip white, contrasting with surrounding color, extending posteriorly to forearm insertion; dorsolateral fold slightly darker than remainder of dorsum; dark brown postocular mask present; dark loreal stripe distinct on the upper loreal region, entire face between white lip stripe and canthus pale; side of face same color as dorsal head; tympanum dark chestnut brown.

Ventral surface color of chin, throat, chest, and abdomen white mottled with light grayish brown; vocal sacs indistinct in appearance with adjacent throat. Thighs and shanks each with three or four dark brown stripes on a lighter grayish brown ground; dark bars on thighs obscure and narrower than intervening pale grayish brown ground color; rear of thighs clouded with dark brown on light brown or straw ground color; center of ventral surfaces of thighs clouded with light gray blotches; and venter white or with faint dark

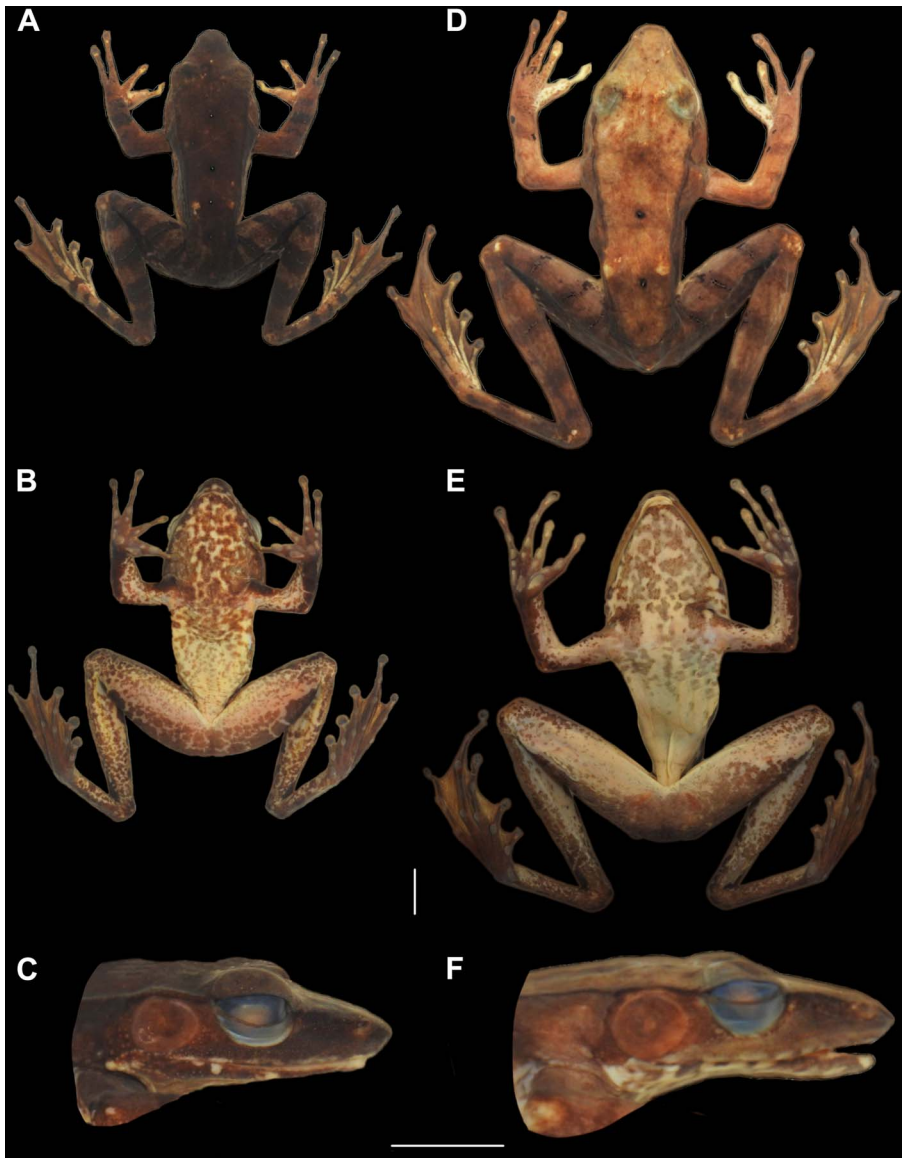

Fig. 6.—*Hylarana nigroverrucosa* sp. nov. (A–C) Holotype male (MZB Amph. 12774), and (D–F) female paratype (MZB Amph. 12778; scale: 10 mm).

yellow cast, evenly suffused with dark punctations or gray clouding. Soles, palms, and webbing brown.

Variation.—Adult males are darker than females; upper lips white, with a few scattered grayish brown only on males; the mottled pattern on males darker than on females, sometimes mottled in the form of dark spread; supernumerary tubercles at the bases of fingers the same as or somewhat little smaller than the above subarticular tubercle especially on third finger (F3).

Natural history.—This species is found in ponds and rivers that flow across primary montane forests and forest fringes. All specimens of the new species were collected less than 1.5 m away from the river bank (Setiadi and Hamidy 2006). There is no information about diet and call for this species.

Distribution.—According to Setiadi and Hamidy (2006), *H. moluccana* (then *R. papua* sensu lato) has been recorded in East, West, and South Halmahera. We observed *H. moluccana* is sympatric with *H. nigroverrucosa* sp. nov. at elevations 0–800 m a.s.l. in Halmahera. This species is also distributed in surrounding islets like Bacan, Tidore, Ternate, Moti, Obi, etc. (Fig. 1).

Discussion

Halmahera is the largest island (17,780 km^2; Fig. 1) in the northern Moluccas. It harbors many endemic species, notably seven species of amphibians: *H. moluccana*, *Nyctimystes infrafrenatus*, *Oreophryne moluccensis*, *Oreophryne frontifasciata*, *Callulops boettgeri*, *Ranoidea rueppelli*, and *Cophixalus montanus*, the last four species being endemic to Halmahera Island itself (Frost 2023). The discovery of *H. nigroverrucosa* sp. nov. increases the number of endemic frogs in Halmahera to five species.

A systematic revision of true frogs of the genus *Rana* (Linnaeus 1758) was carried out by Dubois (1992), who divided the genus into six genera, some being divided into subgenera including *Chalcorana*, *Humerana*, *Papurana*, *Sylvirana*, and

TABLE 3.—Selected morphometric characters (in millimeters) of *H. moluccana* voucher specimens examined (see Appendix for accession data).

	Male ($n = 44$)		Female ($n = 22$)	
	Mean ± SD	Range	Mean ± SD	Range
SVL	53.5 ± 4.0	47.5–65.0	64.7 ± 5.8	53.0–77.0
IN	4.6 ± 0.2	4.0–5.0	5.4 ± 0.4	4.5–6.0
EN	4.6 ± 0.5	3.5–5.5	5.8 ± 0.5	5.0–6.5
SL	7.6 ± 0.7	6.5–9.5	9.4 ± 0.9	7.5–11.0
ED	5.0 ± 0.4	4.2–6.0	5.7 ± 0.5	5.0–7.0
EE	4.5 ± 0.3	4.0–5.5	5.2 ± 0.5	4.5–6.5
UEW	4.7 ± 0.4	4.0–5.5	5.7 ± 0.5	5.0–6.5
TD	4.7 ± 0.4	4.0–5.5	5.3 ± 0.5	4.5–6.5
TE	1.6 ± 0.2	1.0–2.0	2.0 ± 0.3	1.5–3.0
HL	20.3 ± 1.7	17.5–25.0	24.9 ± 2.3	21.0–30.0
HW	16.5 ± 1.6	14.0–21.0	19.9 ± 2.3	16.0–25.0
HH	7.4 ± 0.8	6.5–10.0	9.1 ± 1.2	7.0–12.5
UAL	10.0 ± 0.9	8.4–12.0	12.8 ± 1.3	10.0–15.0
FAL	10.0 ± 0.9	8.4–12.0	12.8 ± 1.3	10.0–15.0
Finger I length (F1)	12.0 ± 0.9	10.0–14.0	14.8 ± 1.3	12.0–17.0
Finger II length (F2)	11.1 ± 0.8	9.5–13.0	13.6 ± 1.2	11.5–16.0
Finger III length (F3)	15.1 ± 1.3	13.0–18.0	18.1 ± 1.5	15.0–20.5
Finger IV length (F4)	12.8 ± 1.1	11.0–15.5	15.5 ± 1.3	12.5–18.0
FL	23.1 ± 2.0	19.0–30.0	28.6 ± 2.3	22.0–31.0
TL	30.9 ± 2.7	27.5–38.0	38.6 ± 3.2	30.5–42.0
TaL	16.6 ± 1.4	15.0–20.0	20.3 ± 2.3	12.5–22.5
Toe I length (T1)	9.8 ± 1.1	8.0–12.5	12.1 ± 0.9	10.0–13.5
Toe II length (T2)	14.0 ± 1.5	11.7–18.0	17.3 ± 1.5	14.5–19.5
Toe III length (T3)	19.5 ± 1.9	17.0–24.5	24.1 ± 2.1	19.5–27.5
Toe IV length (T4)	27.4 ± 2.4	24.3–34.0	33.8 ± 3.8	28.0–38.0
Toe V length (T5)	20.3 ± 2.0	17.5–25.0	24.0 ± 5.5	20.5–28.5

Amnirana. Dubois (1992) placed the Southeast Asian species with *Sylvirana*, which was considered as the most appropriate placement for this species at that time (Chandramouli et al. 2020). Frost et al. (2006) followed Dubois (1992) and placed *R. moluccana* under *Sylvirana* while raising the subgenus to a generic level. However, Che et al. (2007) included *Sylvirana* members again within *Hylarana*. Based on phylogenetic evidence, Oliver et al. (2015) later reassigned *H. moluccana* to *Papurana*. The subsequent authors followed this rearrangement (e.g., Chan et al. 2020; Chandramouli et al. 2020) until Dubois et al. (2021) lumped all these genera into *Hylarana* again.

Chan et al. (2020) assigned Indo–Myanmar and East Asian taxa *H. milleti* and *H. attigua* into genus *Papurana*. Recently, Reilly et al. (2022) included the Papuan and Lesser Sundanese assemblages (17 species) under the subgenus *Papurana*, but excluding *H. milleti* (Indo–Myanmar and east Asian), *H. attigua* (east Asian), *H. nigrovittata* (Asian), and *H. malabarica* (south Asian), but came up with a poorly supported phylogenetic tree, probably because of the short 16S rRNA mitochondrial gene fragments used in the analysis. Widely used, short 16S rRNA mitochondrial gene fragments yield poor and erratic results in at least some phylogenetic studies (Chan et al. 2022). Therefore, we tried our best to avoid short 16S rRNA gene fragments in our analysis and used all the available homologous genes to generate what should be at least a mostly resolved and meaningful tree.

Actually, Reilly et al. (2022) also found that the Lesser Sunda *H. florensis* and *H. elberti* form a clade that is sister to the rest of the Australo-Papuan *Papurana* assemblage. However, the phylogenetic placement of *H. moluccana* has not been confirmed due to the unavailability of molecular data. With our molecular data on *H. moluccana*, we realized that *H. moluccana* represents a separate clade, and the existence of these two distantly related sympatric lineages on Halmahera Island (*papua* clade and *celebensis* clade) suggests an independent dispersal towards this island. The dispersal of *H. moluccana* (*celebensis* clade) on Halmahera Island seems more recent compared to *H. nigroverrucosa* sp. nov. (*papua* clade). The previous studies (e.g., Reilly et al. 2022) suggested that the Lesser Sundas clade of subgenus *Papurana* is the sister group to the Australo-Papuan clade, which is compatible with a biogeographical scenario in which the Lesser Sundas acted as a stepping stone between the Asian and Australo-Papuan biogeographic realms.

Furthermore, based on morphology and phylogenetic analysis, both populations of *Hylarana moluccana* on Halmahera and Ternate belong to the same species and are only separated by sequence distances (uncorrected *p* distances) between 0 and 0.2% of 16S rRNA. Although, Mertens (1967) mistakenly identified one of the syntypes (SMF 6562) as a holotype, in the original description of *R. moluccana*, the distribution (i.e., type locality) was given as "Halmaheira und Ternate" (Boettger 1895) referring to two specimens, hence syntypes. However, according to Article 74.6 of the International Code of Zoological Nomenclature (ICZN 1999), here we accept the specimen SMF 6562 as the lectotype of *R. moluccana* and the designator was Mertens (1967). The designated lectotype was collected from Ternate Island; hence we restrict the type locality of *R. moluccana* to Ternate, though it is a widely distributed species in both Halmahera and Ternate.

Our study is still preliminary, but it could be hypothesized that the ancestral population of *H. nigroverrucosa* sp. nov. perhaps split into two sympatric lineages on Halmahera Island and one lineage dispersed along the highlands of Papua New Guinea towards Australia. The other lineages seem to have dispersed towards Lesser Sunda, through the lower peneplains, evidently evolved as *H. elberti* and *H. florensis*. Sympatric evolutionary lineages (in this case Halmahera) of several morphologically cryptic frogs have obfuscated their taxonomy over a long period implying that the species diversity of Southeast Asian frogs remains significantly underestimated (see Stuart et al. 2006; Inger et al. 2009).

The colonization of islands and subsequent isolation across oceanic barriers is assumed to be the primary diversification mechanism in insular frogs (e.g., Gonzales et al. 2014; Yu et al. 2020). Similar to recent discoveries of lizard and snake fauna from Sulawesi and Moluccas (e.g., Amarasinghe et al. 2021; Riyanto et al. 2022), the discovery of a genetically and morphologically distinct *Hylarana* suggests additional unrecognized amphibian species await discovery and description on other islands of the region. Our results provide new insights into insular isolation in a previously unstudied region and further demonstrate that the actual diversity of frogs is vastly underestimated in Indonesia.

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Supplemental Material

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APPENDIX

Other Specimens Examined

***Hylarana aurata*.**—Indonesia: West Papua: ZMB 58694 (holotype).
***Hylarana celebensis*.**—Indonesia: North Sulawesi: Gorontalo: ZMB 5745 (holotype).
***Hylarana celebensis*.**—Indonesia: North Sulawesi: Gorontalo: MZB Amph 23365, 24220.
***Hylarana daemeli*.**—Indonesia: West Papua: MZB Amph 24439–41, 24443; Raja Ampat: 17695, 17700–01; Waigeo: 13347.
***Hylarana elberti*.**—Indonesia: East Nusa Tenggara: Sumba: MZB Amph. 26711–12, 26719, 26723.
***Hylarana florensis*.**—Indonesia: West Nusa Tenggara: Sumbawa: MZB Amph. 32049, 32680; Flores: MZB Amph. 201, 21795.
***Hylarana garritor*.**—Papua New Guinea: UPNG 1647 (holotype).
***Hylarana grisea*.**—Indonesia: West Papua: ZMA.RENA.5704 (holotype)
***Hylarana jimiensis*.**—Indonesia: West Papua: MZB Amph. 6879–80.
***Hylarana moluccana*.**—Indonesia: North Halmahera: MZB Amph. 12640, 12760–62; West Halmahera: 12635–38, 12653–54, 12758, 12773, 33553; East Halmahera: 12643–47, 12651, 12765, 12767–68, 12771–72; Central Halmahera: 12656, 16040–41, 16043–51, 16053–54, 21451; Ternate: 12633, 12757, 15382–90, 15392–93; Obi Island: 28529; Tidore Island: 12648–50, 12769, 12793; Moti Island: 16415–19, 16421–24.
***Hylarana novaeguineae*.**—Indonesia: West Papua: MZB Amph. 24410–11, 24437.
***Hylarana papua*.**—Papua New Guinea: UPNG 2717 (holotype).
***Hylarana supragrisea*.**—Papua New Guinea: UPNG 3586 (holotype).
***Hylarana arfaki*.**—Indonesia: West Papua: Waigeo: MZB Amph. 13309, 13316, 13319, 15229.
***Hylarana volkerjane*.**—Indonesia: West Papua: ZMB 64005 (holotype).
***Hylarana volkerjane*.**—Indonesia: West Papua: MZB Amph. 24403–04, 24407, 24409, 24412, 24438, 24941.