Morphological Variations and New Species Description of Genus *Rousettus* Bat from Gunung Duasudara Sanctuary, North Sulawesi, Indonesia

1,2Hanry Jefry Lengkong, 3Endang Arisoesilaningsih, 3Luchman Hakim and 4Sudarto

1Doctoral Student of Biology Department, Faculty of Mathematics and Natural Sciences, Brawijaya University, Malang, Indonesia
2Faculty of Mathematics and Natural Sciences, Sam Ratulangi University, Manado, North Sulawesi, Indonesia
3Department of Biology, Faculty of Mathematics and Natural Sciences, Brawijaya University, Malang, Indonesia
4Faculty of Agriculture, Brawijaya University, Malang, Indonesia

Abstract: Bats belongs to Pteropodidae Family that spreaded evenly in Indonesia. Genus *Rousettus* have their morphological variances among its own species based on characteristics on each species. Among them there is fruit-feeding bats of from genus *Rousettus* (Chiroptera: Pteropodidae) that have many variances of morphology among its own species. This study was aimed to identify the morphological variations and its sex type influence of genus *Rousettus* bats from Gunung Duasudara Sanctuary, North Sulawesi. The locations were consisted 7 types of major vegetations at altitude range from 0 to 1351 m above sea level (asl). All habitat types were observed using Mist-net method at 1 and 3 m above the ground. There were found 452 individuals, including *R. amplexicaudatus* (224), *R. celebensis* (219) and *R. tangkokoensis* n. sp. (9). Nine individuals of *Rousettus tangkokoensis* n. sp. were newly found in lowland forest and coastal forest. These newly-found species were different from other *Rousettus*. There was discovered that sex type had influenced the skull and external body characters on *R. amplexicaudatus*, *R. tangkokoensis* n. sp. *R. celebensis*. However, most of other characters were statistically not-significant that indicated there was not any sexual dimorphism. According to the Discriminant Function Analysis (DFA), these morphological groups possess different specification. Therefore, the three species of genus *Rousettus* have statistically variation of skull and external body characters one to another.

Keywords: North Sulawesi, Gunung Duasudara Sanctuary, *Rousettus*

Introduction

Indonesia contains many species of nocturnal animals, especially bats. Bats belong to Pteropodidae Family that spreaded evenly in Indonesia. There are many research reports about bats spreads along Sulawesi Island. Bergmans and Rozendaal (1988) reported that in Sangihe islands and Selayar, Sulawesi, there were 21 species of Pteropodidae Family. On the other hand, according to Suyanto (2001) and Suyanto et al. (2002), there were 25 species of Pteropodidae in Sulawesi.

Lee et al. (2001) found 9 species of bats in 6 natural vegetations in Gunung Duasudara Sanctuary, Sulawesi, the bat species were found are: *Rousettus celebensis*, *Cynopterus brachyotis*, *Thoopterus nigrescens*, *Nyctimene cephalotes*, *Macroglossus minimus*, *Megaderma spasma*, *Pteropus hypomelanus*, *Rhinolopus* sp. and *Myotis* sp. Furthermore, Lengkong (2009) also reported the total of 9 species presence from 4 vegetations in the same location, they found *Boneia bidens* and *Dobsonia exolta* species that previously unreported. However, Lengkong (2009) did not find 3 species that previously reported by Lee et al. (2001) i.e., *M. spasma*, *P. hypomelanus* and *Rhinolopus* sp.

Among them there is fruit-feeding bats from genus *Rousettus* (Chiroptera: Pteropodidae) that have many
variances of morphology among its own species. Genus *Rousettus* has 9 species which are widely distributed from Indonesia to Southeast Asia, southwest Africa, Madagascar (Corbet and Hill, 1992), Philippine, Papua New Guinea and Solomon (Koopman, 1993). Corbet and Hill (1992) reported that there were 5 species of *Rousettus* from Indonesia, i.e.: *Rousettus (Rousettus) amplexicaudatus* (Saint-Hilaire, 1810) which are distributed in Sumatra, Kalimantan, Sulawesi, Malucas, Java, Bali, Sunda Lesser and S. Burma; *R. (R.) celebensis* (Andersen, 1907) and *Rousettus (Boneia) bidens* (Jentink, 1879) in Sulawesi; *R. (R.) leschenaultii* (Desmarest, 1820) in Java and Pakistan; and *R. (R.) spinalatus* (Bergmans and Hill, 1980) in Sumatra and Kalimantan.

There are 5 species of *Rousettus* in Sulawesi, i.e.: *Rousettus amplexicaudatus, Rousettus celebensis, Rousettus leschenaultii, Rousettus spinalatus* and *Rousettus linduensis*. Therefore, Sulawesi is an area that possessing high level of endemic fauna and the hot spot of *Rousettus* species diversity (Suyanto et al., 2002); (Maryanto and Yani, 2003).

Genus *Rousettus* have small body size compared to other bats genus. They have long snout (Suyanto, 2001; Payne et al., 2000), second finger claw, short tail, wing membrane attached to the body side, except in *R. spinalatus* (Andersen, 1912; Bergmans and Hill, 1980; Payne et al., 2000; Suyanto, 2001). They have wings attached to the hindfoot base of the first or second foot finger, or around the first or second metatarsal, or between both (Bergmans, 1997).

Genus *Rousettus* have their morphological variances among its own species based on characteristics on each species. Therefore, this study was aimed to identify the morphological variations in bat species of genus *Rousettus* in Gunung Duasudara Sanctuary, North Sulawesi.

### Materials and Methods

#### Study Site

The study was conducted from November 2013 to August 2014 in Gunung Duasudara Sanctuary, North Sulawesi (Fig. 1). It covered 7 major vegetations at altitude 0-1351 m above sea level (asl). Samples were obtained using 2 bat nets sized 12×3.6 m, it was set at 1 and 3 m above the ground. The net setting locations are presented in Fig. 1.

#### Methods

The samples were 452 adult bats that had been netted in Gunung Duasudara Sanctuary. Twenty-nine bats were preserved in 70% alcohol and their skull bones were taken out as specimens (Table 1). The measurements of the specimen’s skull, jaw and tooth were done on 18 skull characters and 7 external body characters collected from adult individuals at the accuracy of 0.1 cm (Fig. 2).

Table 1. ANOVA of skull and external body characters of genus *Rousettus* based on sex type influence. The significance levels of F used: * = p<0.05, ** = p<0.01 dan *** = p<0.001

| Characters | *R. amplexicaudatus* | *R. celebensis* | *R. tangkokoensis n. sp.* |
|------------|---------------------|----------------|--------------------------|
| a. Skull   |                     |                |                          |
| GSL        | 1.96                | 7.66*          | 0.35                     |
| ZB         | 2.21                | 14.00**        | 1.32                     |
| LIW        | 2.41                | 0.10           | 2.27                     |
| POW        | 0.00                | 2.81           | 3.48                     |
| BCW        | 0.72                | 0.30           | 0.05                     |
| BL         | 0.14                | 0.47           | 1.58                     |
| MSF        | 1.42                | 0.23           | 14.19**                  |
| PL         | 0.09                | 0.01           | 0.09                     |
| CBL        | 0.62                | 7.02*          | 0.01                     |
| CCL        | 2.63                | 4.44           | 0.01                     |
| C1-M3      | 0.79                | 0.00           | 0.08                     |
| C1, C1     | 2.26                | 8.20*          | 1.80                     |
| M3-M3      | 5.61*               | 0.01           | 2.12                     |
| DL         | 0.41                | 0.03           | 0.25                     |
| RAP        | 4.13                | 6.94*          | 1.12                     |
| C2-M3      | 0.02                | 1.83           | 1.70                     |
| C2-C1      | 2.62                | 2.35           | 0.01                     |
| M2-M3      | 4.39                | 0.00           | 0.32                     |
| b. External body | 8.19**        | 21.16***        | 0.37                     |
| SV         | 18.48****          | 3.52           | 0.37                     |
| FL         | 0.01                | 9.55**         | 0.05                     |
| FA         | 14.65****          | 3.91*          | 0.01                     |
| HF         | 2.69                | 3.41           | 0.25                     |
| EAR        | 0.71                | 1.09           | 0.39                     |
| TAIL       | 1.24                | 2.47           | 0.14                     |
Fig. 1. Study site of genus *Rousettus* bat in Gunung Duasudara Sanctuary, North Sulawesi

Fig. 2. Measurements of skull and external body of genus *Rousettus*
The skull characters measured were: (1) Greatest Skull Length (GSL), (2) Zygomatic Breadth (ZB), (3) Least Interorbital Width (LIW), (4) Postorbital Width (POW), (5) Braincase Width (BCW), (6) Bullae Length (BL), (7) Mesopterygoid Fossa Width (MSF), (8) Palatal Length (PL), (9) Condyleobasal Length (CBL), (10) Condylar Length (CCL), (11) distance between upper canine and third upper molar (C\textsuperscript{3}M\textsuperscript{3}), (12) distance between outside upper canine (C\textsuperscript{3}C\textsuperscript{1}), (13) distance between outside third upper molar (M\textsuperscript{3}M\textsuperscript{3}), (14) Dentary Length (DL), (14) Ramus Angular Process (RAP), (15) distance between lower canine and third lower molar (C\textsubscript{1}M\textsubscript{3}), (16) distance between outside lower canine (C\textsubscript{1}C\textsubscript{3}) and (17) distance between outside third lower molar (M\textsubscript{3}M\textsubscript{3}).

The external body characters measured were: (1) Snot-Vent length (SV), (2) Face Length (FL), (3) Forearm (FA), (4) Tibia (TIB), (5) Hindfoot Length (HF), (6) Ear (EAR) and (7) Tail (TAIL).

Each species was statistically tested based on 18 skull characters and 7 external body characters using ANOVA to determine the influence of *Rousettus* sp. sex to their morphological variations. Discriminant Function Analysis (DFA) was done by combining most of skull characters and all external body characters separately using species as priority group. This interspecific relationship in the discriminant function between skull characters and external body characters was highly similar. The DFA could also identify important characters in separating species/group. To minimize Wilk’s Lambda value, there were only 6 characters were used (GSL, ZB, MSF, LIW, C\textsuperscript{1}C\textsuperscript{1}, FA).

**Results and Discussion**

Supp.Data.1 shows the value of sample size, mean, standard deviation, maximum and minimum of several skull and external body character measurements (mm) of genus *Rousettus*. It describe that *R. celebensis* has statistically the largest skull characters among three *Rousettus* species in this study. Despite only represented by sex. To avoid the “over fitting” to the DFA result, there were not all characters were employed. This study used only 6 characters (GSL, ZB, MSF, LIW, C\textsuperscript{1}C\textsuperscript{1}, FA) selected to minimize the Wilk’s Lambda value. Although the DFA of these three species groups had used reduce characters, the DFA value was similar to that using complete 25 skull and external body characters. This result was only based on 6 selected characters (Fig. 3 and Table 2).

The plot of genus *Rousettus* comprising 3 species, *R. amplexicaudatus, R. celebensis* and *R. tangkokoensis* n. sp. DFA was initially done to all skull and external body characters that were not significantly influenced by sex. To avoid the “over fitting” to the DFA result, there were not all characters were employed. This study was used only 6 characters (GSL, ZB, MSF, LIW, C\textsuperscript{1}C\textsuperscript{1}, FA) selected to minimize the Wilk’s Lambda value. Although the DFA of these three species groups had used reduce characters, the DFA value was similar to that using complete 25 skull and external body characters. This result was only based on 6 selected characters (Fig. 3 and Table 2).

**Analysis of Variance (ANOVA) of sex influence upon entire skull and external body characters** were measured in all genus shows that sex significantly influenced one skull character (M\textsuperscript{3}M\textsuperscript{3}) and 3 external body characters (SV, FL, TIB) of *R. amplexicaudatus* (p<0.05). It was influenced 5 skull characters (GSL, ZB, CBL, C\textsuperscript{1}C\textsuperscript{1}, RAP) and 3 external body characters (SV, FA and TIB) of *R. celebensis*. Yet it was influenced one skull character (MSF) of *R. tangkokoensis* n. sp. Thus, most of the analyzed skull characters and external body characters were not affected by sex. Therefore, both skull and external body characters of the three species were not sexually dimorphism (Table 1).

DFA used the species group represented by taxa *R. amplexicaudatus, R. celebensis* and *R. tangkokoensis* n. sp. DFA was initially done to all skull and external body characters that were not significantly influenced by sex. To avoid the “over fitting” to the DFA result, there were not all characters were employed. This study was used only 6 characters (GSL, ZB, MSF, LIW, C\textsuperscript{1}C\textsuperscript{1}, FA) was employed to minimize the Wilk’s Lambda value. Although the DFA of these three species groups had used reduce characters, the DFA value was similar to that using complete 25 skull and external body characters. This result was only based on 6 selected characters (Fig. 3 and Table 2).

The plot of genus *Rousettus* comprising 3 species, *R. amplexicaudatus, R. celebensis* and *R. tangkokoensis* n. sp. on the basis of GSL, ZB, MSF, LIW, C\textsuperscript{1}C\textsuperscript{1} and FA (Fig. 3). It is shows that those 3 species were dispersing, indicates that their kindship was close. The classification failures mostly occurred in *R. amplexicaudatus*. *R. amplexicaudatus* is close to either *R. celebensis* or *R. tangkokoensis* n. sp., so that some members of *R. amplexicaudatus* could be statistically misclassified as either *R. celebensis* or *R. tangkokoensis* n. sp. For *R. celebensis* and *R. tangkokoensis* n. sp. with sufficiently low similarity, these data did not result in error of species classification.

The DFA characters (>0.5) set in function 1 are GSL, ZB, MSF, LIW, C\textsuperscript{1}C\textsuperscript{1} and FA set in function 2 were GSL, ZB, MSF, LIW, C\textsuperscript{1}C\textsuperscript{1} and FA (Table 2).

The DFA of the reduced characters was grouped into 2 significance functions explaining all variances. Function 1 explicates, whereas function 2 eludicate the variance with a total number of, respectively, 93.6 and 6.4% (Table 2). The data diversity of function 1 is higher than that of function 2, it shows that various morphological differences involving skull and external body characters are entirely an important character in separating members of genus *Rousettus*. These morphological groups are considered having different specifications, since these 3 species of genus *Rousettus* possess different skull and external body characters one to another.
Table 2. Standard and unstandard discriminant function coefficient of 3 species of genus *Rousettus*

| Character | Function 1 | Function 2 |
|-----------|------------|------------|
| GSL       | -1.129 (-0.632) | 0.257 (0.144) |
| ZB        | 0.378 (0.245)  | -0.141 (-0.091) |
| MSF       | -0.282 (-0.754) | -0.096 (-0.258) |
| LIW       | 0.445 (0.964)  | 1.201 (2.601)  |
| C'C'      | -0.117 (-0.179) | -0.612 (-0.937) |
| FA        | 0.799 (0.228)  | -0.209 (-0.060) |

Eigen Value
- Function 1: 1.806
- Function 2: 0.113

Constant
- Function 1: -2.150
- Function 2: -10.451

Variance explained (%)
- Function 1: 93.6
- Function 2: 6.4

Fig. 3. Discriminant function 1 and 2 plot of genus *Rousettus* based on 6 selected characters

Species Systematics
- Class: Mammal
- Order: Chiroptera
- Suborder: Megachiroptera
- Family: Pteropodidae Gray, 1821
- Genus: Rousettus Gray, 1821

*Rousettus Amplexicaudatus* Saint-Hilaire (1810)

**Holotype**

Hanry Lengkong collected specimen from Manado, Indonesia (HL 11131 [field no. RA-2]). It was caught on November 7th 2013 over the grassland, at 14 m above the sea level, Gunung Duasudara Sanctuary, North Sulawesi, Indonesia (N 01°32.262', E 125°12.281'). The specimen was adult male, testes swollen, 74 grams BW, skull and dentary separated. Fresh specimens were preserved in 70% alcohol, meters

**Parasite**

Nine specimens were collected by Hanry lengkong from moss forest, submontana forest, owl and forest, casuarine forest, coastal forest, shrub and grassland near the holotype location.

The specimens were:
- HL11132 (field no. RA-19): adult male, testes swollen, 73 grams BW, caught at 15 m above sea level (N 01°32.883', E 125°12.543')
Distribution

The specimens were found in Gunung Duasudara Sanctuary, North Sulawesi, Indonesia.

Diagnosis

*R. amplexicaudatus* has bigger size than average of *R. tangkokoensis* n. sp. but smaller than the average of *R. celebensis* for all characters, including POW, FA, HF and EAR (Table 1).

Description

All bats were described directly in the sanctuary of Gunung Duasudara. *R. amplexicaudatus* specimen was compared with *R. celebensis* and *R. tangkokoensis* n. sp. (Fig. 1).

Pelage

The color of shoulder pelage was greyish brown to brown, chest to belly was slightly bright grey brown, neck was grey brown with yellow hair bundle on both neck sides, top of head was slightly dark grey brown. There was no dense hair up to around the thigh, the wing is hairy and no wing attached onto shoulder side.

**External Body**

*R. amplexicaudatus* bats possessed longer SV, FL, TIB and TAIL than *R. celebensis* and *R. tangkokoensis* n. sp.; meanwhile the FA, HF and EAR are bigger than those of *R. celebensis* but smaller than those of *R. tangkokoensis* n. sp.

Skull

*R. amplexicaudatus* had longer GSL, BL, LIW and POW than those in *R. celebensis* and *R. tangkokoensis* n. sp.; meanwhile the ZB, BCW, MSF, PL, CBL and CCL are bigger than those in *R. tangkokoensis* n. sp. but smaller than those in *R. celebensis*.

Teeth and Dentary

*R. amplexicaudatus* specimen had longer M1 M2, M3 M4 and RAP than *R. celebensis* and *R. tangkokoensis* n. sp.; the C1 M3, C2 M1, C3 C1, C4 C1 and DL are also bigger than those in *R. tangkokoensis* n. sp. but smaller than those in *R. celebensis*.

Biological Condition

Ninety-eight adult males possessed reproductive condition (63 unswollen testes, 9 slightly swollen testes and 26 swollen testes) and 126 adult females had reproductive condition (60 unswollen nipples, 16 slightly swollen, 23 swollen nipples without bite marks, 24 swollen nipples with bite marks and 3 pregnant).

Habitat

Two-hundred seventy-nine bats were caught using bat nets at 1 and 3 m above the ground in moss forest, submontana forest, lowland forest, casuarine forest, shrub and grassland in rainy and dry seasons.

Rousettus Celebensis Andersen (1907)

Holotype

Hanry Lengkong collected the specimens from Manado, Indonesia (HL) 111311 (field no. RC-1). It was caught on December 6th, 2013, in moss vegetation, at 1.037 m above sea level, Gunung Duasudara Sanctuary, North Sulawesi, Indonesia (N 01°31', 271', E 125°11', 169°). The specimens were adult male, unswollen testes, 44 grams BW. Skull and dentary were seperated then preserved in 70% alcohol.

Parasitype

Nine specimens were collected by Hanry lengkong from vegetations of moss forest, submontana forest, lowland forest, casuarine forest, coastal forest, shrub and grassland near the holotype location. The specimens were:
• HL111312 (field no. RC-84), adult female, swollen nipples without bite marks, 41 grams BW, caught at 1.109 m above sea level (N 01°31.115', E 125°11.093')
• HL 111313 (field no. RC-94), adult male, slightly swollen testes, 95 grams BW, caught at 1.109 m above sea level (N 01°31.115', E 125°11.093')
• HL111314 (field no. RC-129), adult female, unswollen nipples, 42 grams BW, caught at 996 m above sea level (N 01°53.809', E 125°11.076')
• HL 111315 (field no. RC-137), adult male, slightly swollen testes, 90 grams BW, caught at 712 m above sea level (N 01°31.193', E 125°12.266')
• HL 111316 (field no. RC-173), adult female, unswollen nipples, 55 grams BW, caught at 514 m above sea level (N 01°30.872', E 125°13.516')
• HL 111317 (field no. RC-190), adult male, unswollen testes, 75 grams BW, caught at 389 m above sea level (N 01°30.620', E 125°13.201')
• HL 111318 (field no. RC-198), adult female, unswollen nipples, 58 grams BW, caught at 1.351 m above sea level (N 01°28.991', E 125°10.291')
• HL 111319 (field no. RC-199), adult male, unswollen testes, 67 grams BW, caught at 1.345 m above sea level (N 01°29.022', E 125°10.344')
• HL 111320 (field no. RC-204), adult female, unswollen nipples, 56 grams BW, caught at 55 m above sea level (N 01°33.429', E 125°09.359')

Distribution
Those specimens found in Gunung Duasudara Sanctuary, North Sulawesi, Indonesia.

Diagnosis
R. celebensis has the biggest skull and external body characters. It is different from R. amplexicaudatus and R. tangkokoensis n. sp. since it possesses many larger skull characters despite represented only by the following skull characters, including: GSL of 37.30, ZB of 22.20, BCW of 14.48, MSF of 5.07, PL of 17.99, CBL of 35.41, CCL of 33.44, C'M of 13.10, C'C of 6.74, DL of 27.67, C'M of 14.17, C'C of 4.09 (Table 1).

Description
All bats were described directly in Gunung Duasudara Sanctuary. R. celebensis specimen was compared with that of R. amplexicaudatus and R. tangkokoensis n. sp. (Fig. 4).

Pelage
The color of shoulder pelage was brown to yellowish brown, chest to belly was slightly bright yellowish brown, neck was dark orange, top head was slightly dark yellowish brown. They had dense pelage around the thigh, hairy wings and no wing attaching on to the shoulder side.

External Body
R. celebensis had shorter SV, FA, TIB, HF and EAR than those in R. amplexicaudatus and R. tangkokoensis n. sp.; FL and TAIL are longer than those in R. tangkokoensis n. sp., but smaller than those in R. amplexicaudatus.

Skull
R. celebensis had longer GSL, ZB, BCW, BL, MSF, PL, CBL and CCL than those in R. amplexicaudatus and R. tangkokoensis n. sp.; meanwhile the LIW was larger than that in R. tangkokoensis n. sp. but smaller than that in R. amplexicaudatus; POW was larger than that in R. amplexicaudatus but smaller than that in R. tangkokoensis n. sp.

Teeth and Dentary
R. celebensis had longer C'M, C'M, C'C, C'C, and DL than those in R. tangkokoensis n. sp. and R. amplexicaudatus; the M'M, M'M, and RAP were bigger than those in R. tangkokoensis n. sp. but smaller than those in R. amplexicaudatus.

Biological Condition
Ninety adult males had reproductive condition (76 had unswollen testes, 3 had slightly swollen testes and 11 had swollen testes) and 123 adult females had reproductive condition (76 had unswollen nipples, 9 had slightly swollen nipples and 20 had swollen nipples without bite marks, 9 had swollen nipples with bite marks and 9 pregnant).

Habitat
Two-hundred sixty-one individuals were caught one time in rainy and dry seasons by using bat nets at 1 and 3 m above the ground in moss forest, submontana forest, lowland forest, casuarine forest, coastal forest and shrub.

Rousettus Tangkokoensis new. sp.

Holotype
Hanry Lengkong was collected the specimens from Manado, Indonesia (HL) 111321 (field no.RT-1). It was caught on June 5th, 2013, lowland forest at 514 m above sea level, Gunung Duasudara Sanctuary, North Sulawesi, Indonesia (N 01°30.872', E 125°13.516'). The specimens were adult female, swollen nipples with bite marks), 84 grams BW. Their skull and dentary were separated then preserved in 70% alcohol.
**Parasitype**

Eight specimen collected by Hanry Lengkong in lowland forest and coastal forest near holotype location. The specimens were:

- HL 111322 (field no. RT-2): adult female, unswollen nipples, 72 grams BW, caught at 495 m above sea level (N 01°30.779', E 125°13.543')
- HL 111323 (field no. RT-3): adult male, swollen testes, 86.0 grams BW, caught at 514 m above sea level (N 01°30.872', E 125°13.516')
- HL 111324 (field no. RT-4): adult male, unswollen nipples, 72 grams BW, caught at 514 m above sea level (N 01°30.872', E 125°13.516')
- HL 111325 (field no. RT-5), adult male, swollen testes, 83 grams BW, caught at 514 m above sea level (N 01°30.872', E 125°13.516')
- HL 111326 (field no. RT-6), adult male, swollen nipples with bite marks, 83 grams BW, caught at 514 m above sea level (N 01°30.872', E 125°13.516')
- HL 111327 (field no. RT-7), adult male, unswollen testes, 77 grams BW, caught at 514 m above sea level (N 01°30.872', E 125°13.516')
- HL 111328 (field no. RT-8), adult female, swollen nipples without bite marks, 82 grams BW, caught at 514 m above sea level (N 01°30.872', E 125°13.516')
- HL 111329 (field no. RT-9), adult male, unswollen testes, 72 grams BW, caught at 192 m above sea level (N 01°30.872', E 125°13.516')

**Ethimology**

The new species which is proposed using the name of Tangkoko Mt. occurring in the sanctuary area was collected by Hanry Lengkong from Manado, Indonesia. The new species is (HL) 111321 had been found in Gunung Dusasudara, which the only area known where this species was collected.

**Diagnosis**

The color of outer appearance of *R. tangkokoensis* n. sp. was blackish orange shoulder, while *R. amplexicaudatus* was greyish brown to brown and *R. celebensis* was brown to yellowish brown. The color of chest to belly of *R. tangkokoensis* n. sp. was slightly bright orange, while *R. amplexicaudatus* was slightly bright grey brown and *R. celebensis* was slightly yellowish brown. The top head of *R. tangkokoensis* n. sp. was colored slightly dark orange, while *R. amplexicaudatus* was slightly dark grey brown and *R. celebensis* was slightly yellowish brown. The neck of *R. tangkokoensis* n. sp. was colored dark orange, while *R. amplexicaudatus* was grey brown with yellow hair bundle on both sides of the neck and *R. celebensis* was dark orange.

The pelage of *R. tangkokoensis* n. sp. and *R. amplexicaudatus* was similar since it did not possessing dense pelage up to around the thigh, while *R. celebensis* had dense pelage around the thigh. Next similarity between *R. tangkokoensis* n. sp., *R. amplexicaudatus* and *R. celebensis* is they had hairy wings attaching on the back side, not on the their shoulder sides (Fig. 4).

*R. tangkokoensis* n. sp. had bigger and smaller skull characters than those in *R. amplexicaudatus* and *R. celebensis*. The former is POW (8.99 mm; 8.60-9.70 mm); the latter is GSL (34.67 mm; 30.30-37 mm). The other characters are ZB (20.79 mm; 18.00-24.40 mm), LIW (7.18 mm; 6.70-8.30 mm), BCW (14.15 mm; 13.70-14.70 mm), BL (2.60 mm; 2.40-2.80 mm), MSF (4.54 mm; 4.10-5.00 mm), PL (16.34 mm; 14.60-17.60 mm), CBL (33.11 mm; 28.00-36.50 mm), CCL (31.84 mm; 27.50-34.90 mm), CCI1 (11.27 mm; 9.30-12.60 mm), CCI2 (6.12 mm; 5.50-7.30 mm), M3M1 (9.13 mm; 8.80-9.80 mm), DL (25.72 mm; 23.00-28.30 mm), RAP width (10.38 mm; 9.50-11.80 mm), C3M3 (12.33 mm; 10.10-14.90 mm), C1C1 (3.79 mm; 3.50-4.20 mm) and M3M4 (8.84 mm; 8.20-9.50 mm).

The external body characters of *R. tangkokoensis* n. sp., (FA, HF, EAR and TAIL) were bigger than those of *R. amplexicaudatus* and *R. celebensis*. TIB of *R. tangkokoensis* n. sp. is also bigger than that of *R. celebensis*.
bigger than \textit{R. amplexicaudatus}. Otherwise, the FL of \textit{R. tangkokoensis} n. sp. is smaller than \textit{R. amplexicaudatus}. Overall, \textit{R. tangkokoensis} n. sp. had small-sized characters, except for POW, FA, HF and EAR, which are bigger than those of \textit{R. amplexicaudatus} and \textit{R. celebensis} (Table 1).

Beside differences among species above, Suyanto (2001) stated that these were only based on the size lower arm wing and the presence or absence of wing attaching on the middle of shoulder. In both characters, \textit{R. tangkokoensis} n. sp. is consistent with typical characteristics of \textit{Rousettus}. Under this reason, the species is classified in a member of \textit{Rousettus}.

Comparing \textit{R. tangkokoensis} n. sp. with other studies on \textit{R. amplexicaudatus} based on one the body sizes, their FA length is about 82.22–86.76 mm (Payne and Francis, 1985) and FA length is about 77–87.2 mm (Suyanto, 2001). Apparently, the \textit{R. tangkokoensis} n. sp. is nearly similar to \textit{R. amplexicaudatus}.

Bergmans and Rosendaal (1988) measured 14 male \textit{R. amplexicaudatus} from Sulawesi. Their FA length is about 81.55 (77.3–85.6 mm), ZB length is 22.3 (20.7–23.3 mm), GSL length is 36.85 (35.2–38.5 mm) and CBL length is 35.4 (34.2–37.2 mm). It is apparently \textit{R. amplexicaudatus} have similarity with the \textit{R. tangkokoensis} n. sp body size, yet it has bigger skull than \textit{R. tangkokoensis} n. sp., but with \textit{R. spinalis}, it had similar FA, 85.1 mm.

Maryanto and Yani (2003) measured the several characters of 12 males \textit{R. celebensis} in Museum Zoologicum Bogoriense and determined their length for these several characters: FA 75.81 mm (72.61-79.41 mm), GSL 39.46 mm (38.75-40.64 mm), ZB 23.59 mm (22.02-25.12 mm), CBL 37.57 mm (36.48-38.66 mm) and POW 7.69 (7.26-8.47 mm). They also measured the several characters from 4 males \textit{R. linduensis} from Sulawesi. The measured of the length for these characters were: FA 76.73 mm (75.64-77.54 mm), GSL 39.38 mm (38.81-39.70 mm), ZB 24.08 mm (23.93-24.25 mm), CBL 37.47 mm (37.2-37.63 mm) and POW 7.07 mm (7.02-7.14 mm). In ferential, \textit{R. celebensis} and \textit{R. linduensis} have smaller body size and entirely larger skull, except the POW size.

Rookmaker and Bergmans (1981) measured the body size of \textit{R. leschenaultii} from Indonesia. \textit{R. tangkokoensis} n. sp. has smaller body size and skull compared with \textit{R. leschenaultii}. \textit{R. leschenaultii} has FA length about 84.0–90.36 mm, GSL 40.3–43.6 mm and ZB 24.8–27.6 mm, respectively.

Moreover, \textit{R. tangkokoensis} n. sp. compared with \textit{R. egyptiacus} (FA of 85-101.9 mm) (Grzimek, 2003; Kwiecinski and Griffin, 1999) and \textit{R. madagascar} (FA of 119-140 mm) (Bergmans, 1997; Bush Warriors, 2013; Jenkins and Racey, 2008; McNab, 1969), it is apparent that \textit{R. tangkokoensis} n. sp. be smaller than \textit{R. egyptiacus} and \textit{R. madagascar}.

**Description**

All bats were described directly in Gunung Duasudara Sanctuary. \textit{R. tangkokoensis} n. sp. specimens were compared with those of \textit{R. amplexicaudatus} and \textit{R. celebensis}.

**Pelage**

The color of shoulder pelage is blackish orange to orange, chest to belly is slightly bright, neck is dark orange, top head is slightly dark orange. There is no dense pelage around the thigh. They have unhaired wings and no wing attaching on the shoulder sides.

**External Body**

\textit{R. tangkokoensis} n. sp. has longer FA, HF, EAR. They have smaller FL and shorter TAIL than those of \textit{R. amplexicaudatus} and \textit{R. celebensis} and longer Snot-Vent (SV) than that of \textit{R. celebensis}.

**Skull**

\textit{R. tangkokoensis} n. sp. has big POW and small GSL, ZB, LIW, BCW, BL, MSF, PL, CBL and CCL.

**Teeth and Dentary**

\textit{R. tangkokoensis} n. sp. has small C¹M³, small C³M³, small C¹C¹, small M³M³, small M¹M¹, short DL and small RAP.

**Biological Condition**

Four male specimens have reproductive condition (two of them have unswnollen testes and the rest have swollen testes) and 5 adult female specimens have reproductive condition (two have unswnollen nipples, 1 have swollen nipples without bite mark and 2 have swollen nipples with bite marks).

**Habitat**

Nine bats were caught when rainy and dry seasons using bat nets at 1 and 3 m above the ground in lowland forest and coastal forest.

**Discussion**

New bat species was found in Sulawesi. There have been 5 species known more than those of other islands in Indonesia or any other areas. It proves that Sulawesi island has high diversity of bats and as (Lee et al., 2001) a hot spot (natural habitat) (Maryanto and Yani, 2003) of this genus. North Sulawesi is the only well-known natural habitat of \textit{Rousettus (Boneia) bidens}, Central Sulawesi is the well-known natural habitat of \textit{Rousettus linduensis}.

In fact, the bats were hunted local people consumption or were sald in the market in North Sulawesi and other areas. That could caused seriously hazard for the entire species and endanger the study basis.
on bats in this island which has unique evolutionary perspectives. In Gunung Duasudara Sanctuary, specifically, there are 12 active hunting areas, including the caves and tree holes where the bats are usually found. Thus, sustainability of bat population in Indonesia could be developed by environmental awareness of bat hunters, tour guide and local communities. Government involvement is also expected in the case of law enforcement of natural habitats protections against illegal bat hunters. Gunung Duasudara Sanctuary needs to be protected due to its unique ecosystem, the presence of endangered species, high natural diversity, the potential function of hydrological protection and natural tourism destination.

This study was conducted in all types of vegetation at the range of altitude 0-1351 m above sea level, included moss forest, submontana forest, lowland forest, casuarine forest, coastal forest, shrub and grassland. Nevertheless, *R. tangkokoensis* n. sp. was only found in lowland forest (8 types) and coastal forest (one type) at 0-600 m above sea level. This species was considered as lowland origin adjacent to casuarine forest of Batuangus Mt., with many genera. Bats could be found in various places possessing vegetated terrestrial ecosystem. Lowland forest is a typical forest holding the highest biodiversity over other types of forests (Medellin et al., 2000). Our research revealed that the area was very important for bats to make a nest and fulfilling their diet due to the availability of preferred food, such as kongkoriang tree (*Adinandra dumosa vidual*) and council tree (*Ficus altissima*). These trees bearing fruits in May to June so that *R. tangkokoensis* n. sp. is only found in lowland forest and coastal forest during that period. It is important that the government establish the zonation to protect bat habitats, especially lowland forest and coastal forest. In this study, there were found possessing high bat diversity and the places where new species was identified. Major concerns on this location are hunting, deforestation and land conversion to coconut plantation area.

*R. tangkokoensis* n. sp. was different from other *Rousettus* since it has blackish orange to orange colored shoulder, orange colored chest to belly, large orange colored neck, darker orange colored top head, unhairy wing attaching on shoulder sides, big and wide FA and large POW. The influence of sex type on all measured parameters using ANOVA. *R. amplexicaudatus* had one skull character (M$^3$M$^1$) and 3 external body characters (SV, FL and TIB) significantly affected; *R. celebensis* had 5 skull characters (GSL, ZB, CBL C$^2$C$^1$ and RAP); and 3 external body characters (SV, FA and TIB) significantly affected; and *R. tangkokoensis* n. sp. has only one skull character (MSF) significantly affected. It means that there is no sexual dimorphism.

The morphology of *R. tangkokoensis* n. sp. was distinguished from *R. amplexicaudatus* and *R. celebensis* based on 2 significance functions. Function 1 describes the variation represented by total number of 93.6% and function 2 explains the variation represented by 6.4% (Table 2). It shows that data diversity of function 1 is higher than that of function 2, refers that various morphological differences involving the entire skull size and external body were an important character to distinguish the members of genus *Rousettus*.

**Conclusion**

Three species of genus *Rousettus* have morphological differences involving the entire skull size and external body. There was discovered that sex type had influenced the skull and external body characters on *R. amplexicaudatus*, *R. tangkokoensis* n. sp. and *R. celebensis*. However, most of other characters were statistically not-significant that indicated there was not any sexual dimorphism.

**Acknowledgement**

Many people have contributed to this research completion. We would specifically appreciated Joyo, Pipo, Johannes, Budi, Nyong and Anto, for the support of bat specimen collection in Gunung Duasudara Sanctuary, North Sulawesi. We would thank to Mahadaratunkamsi as well in measurement and identification techniques during our work in Biological Research and Development Center- LIPI Bogor.

**Author’s Contributions**

Hanry Jefry Lengkong: Participates in all experiment, coordinated the data-analysis and contributed to the writing manuscript.

Endang Arisoesilaningsih, Luchman Hakim and Sudarto: Designed the research plan, organized the study and contributed to the writing manuscript.

**Ethics**

This article is original and contains unpublished material. The corresponding author confirms that all of the other authors have read and approved the manuscript and no ethical issues involved.

**References**

Andersen, K., 1907. On pterocyon, rousettus and myonycteris. Ann. Mag. Nat. Hist., 19:501-515.

Andersen, K., 1912. Catalogue of the Chiroptera in the Collection of the British Museum. 2nd Edn., British Museum Publications, London, pp: 854.

Bergmans, W. and F.G. Rozendaal, 1988. Notes on a Bergmans, W. and F.G. Rozendaal, 1988. Notes on a collection of fruit bats from Sulawesi and some off-lying islands 9 Mammalia, Megachiroptera). Zoologische Verhandelingen, 248: 1-14.
Bergmans, W. and J.E. Hill. 1980. On a new species of *Rousettus* Gray, 1821, from Sumatra and Borneo (Mammalia: Megachiroptera). Bull. Brit. Museum Natural History, 38: 95-104.

Bergmans, W., 1994. Taxonomy and biogeography of African fruit bats (Mammalia, Megachiroptera). 4. The genus *Rousettus* Gray, 1821. Beaufortia, 44: 79-126.

Bergmans, W., 1997. Taxonomy and biogeography of African fruit bats (Mammalia, Megachiroptera). 5. Genera *Lissonycteris* Andersen, 1912, *Myoncterus* Matschie, 1899 and *Megaloglossus*, Pagentecher 1885; general remarks and conclusions; annex: Key to all species. Beaufortia, 47: 11-90.

Rookmaker, L.C. and W. Bergmans, 1981. Taxonomy and geography of *Rousettus amplexicaudatus* (Geoffroy, 1810) with comparative notes on sympatric congeners (Mammalia, Megachiroptera). Beaufortia, 31: 1-29.

Bush Warriors, 2013. “IUCN Species of the Day: African fruit bats (Mammalia, Megachiroptera). 4. The genus *Rousettus*.” Beaufortia, 50: 1-90.

Koopman, K., 1993. Order Chiroptera. In: Mammals of the World, Wilson, D.E. and D.M. Reeder (Eds.), Smithsonian Institution Press, Washington, pp: 137-241.

Kwiecinski, G. and T. Griffiths, 1999. *Rousettus egyptiacus*. Mammalian Species, 611: 1-9.

Lee, R.J., J. Riley and R. Merrill, 2001. Biodiversity and Conservation in Northern part of Sulawesi. WCS-IP and NRM, Prima Centra, Jakarta.

Lengkong, H.J., 2009. Bat diversity in tangoko-duasudara sanctuary based on position above sea level. J. Ilmiah Sains, 9: 218-229.

Maryanto, I. and M. Yani, 2003. A new species of *Rousettus* (Chiroptera: Pteropodidae) from Lore Lindu, Central Sulawesi. Mammal Study, 28: 111-120. DOI: 10.3106/mammalstudy.28.111

McNab, B., 1969. The economics of temperature regulation in Neotropical bats. Comparative Biochem. Physiol., 31: 227-268.

Medellin, R.A., M. Equihua and M.A. Amin, 2000. Bat diversity and abundance as indicators of disturbance in neotropical rainforests. Conserv. Biol., 14: 1666-1675. DOI: 10.1111/j.1523-1739.2000.99068.x

Payne, J and C. Francis, 1985. "IUCN Species of the Day: Madagascan rousette."

Payne, J., C.M. Francis, K. Phillipps and S.N. Kartikasari, 2000. *Rousettus* amplexicaudatus from Celebes. Notes Leyden Mus. 1: 117-119.

**Supp. Data**

Table 1. Skull and external body characters of genus *Rousettus* bats.

| Species | GSL | ZB | LIW | POW | BCW | BL | MSF | PL | CBL | C1' | C4' | C3' | M1' | M2' | DL | RAP | C3 | C2 | C1 | M3 | M4 |
|---------|-----|----|-----|-----|-----|----|-----|----|-----|-----|-----|-----|-----|-----|----|-----|----|----|----|-----|-----|
| *R. amplexicaudatus* | N | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| Min | 34.60 | 20.30 | 6.90 | 6.40 | 13.90 | 2.60 | 4.00 | 15.10 | 33.10 | 31.40 | 11.10 | 4.60 | 8.70 | 26.50 | 10.50 | 12.80 | 3.00 | 8.10 |
| Max | 37.40 | 23.90 | 8.00 | 9.80 | 15.40 | 4.20 | 5.70 | 19.70 | 37.10 | 35.00 | 13.90 | 7.30 | 11.00 | 29.40 | 12.90 | 16.00 | 5.10 | 10.30 |
| X | 36.14 | 21.85 | 7.49 | 7.39 | 14.42 | 3.63 | 4.79 | 17.54 | 34.63 | 32.99 | 12.49 | 6.40 | 9.57 | 27.58 | 11.62 | 13.97 | 3.88 | 9.10 |
| SD | 1.09 | 1.11 | 0.37 | 0.96 | 0.43 | 0.55 | 0.48 | 1.34 | 1.26 | 1.23 | 0.96 | 0.82 | 0.81 | 0.96 | 0.74 | 0.91 | 0.60 | 0.83 |
| *R. celebensis* | N | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 |
| Min | 35.50 | 19.60 | 6.40 | 6.80 | 13.90 | 2.60 | 4.50 | 16.50 | 32.20 | 30.10 | 12.00 | 6.10 | 8.80 | 25.20 | 10.10 | 13.50 | 3.30 | 8.00 |
| Max | 39.30 | 23.50 | 7.90 | 9.60 | 15.00 | 3.85 | 5.70 | 19.10 | 37.40 | 34.70 | 13.80 | 7.60 | 9.60 | 29.60 | 12.70 | 15.30 | 4.80 | 9.40 |
| X | 37.30 | 22.20 | 7.27 | 8.01 | 14.48 | 3.17 | 5.07 | 17.99 | 35.41 | 33.44 | 13.10 | 6.74 | 9.27 | 27.67 | 11.40 | 14.17 | 4.09 | 8.89 |
| SD | 1.75 | 1.21 | 0.48 | 0.76 | 0.33 | 0.52 | 0.31 | 0.86 | 1.43 | 1.30 | 0.52 | 0.50 | 0.27 | 1.31 | 0.80 | 0.61 | 0.51 | 0.49 |
| *R. tangokoensis* | n. sp. | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 | 9 |
| Min | 30.30 | 18.00 | 6.70 | 8.60 | 13.70 | 2.40 | 4.10 | 14.60 | 28.00 | 27.50 | 9.30 | 5.50 | 8.80 | 23.00 | 9.50 | 10.10 | 3.50 | 8.20 |
| Max | 37.80 | 24.40 | 8.30 | 9.70 | 14.70 | 2.80 | 5.00 | 17.60 | 36.50 | 34.90 | 12.60 | 7.30 | 9.80 | 28.30 | 11.80 | 14.90 | 4.20 | 9.50 |
| X | 34.67 | 20.70 | 7.18 | 8.99 | 14.15 | 2.60 | 4.54 | 16.34 | 33.11 | 31.84 | 11.27 | 6.12 | 9.13 | 25.72 | 10.38 | 12.33 | 3.79 | 8.84 |
| SD | 2.74 | 2.17 | 0.52 | 0.39 | 0.33 | 0.16 | 0.30 | 1.00 | 3.14 | 2.50 | 1.26 | 0.58 | 0.33 | 1.79 | 0.76 | 1.64 | 0.24 | 0.37 |
### 1b. External body characters

| Species                  | SV   | FL   | FA   | TIB  | HF   | EAR  | TAIL |
|--------------------------|------|------|------|------|------|------|------|
| *R. amplexicaudatus*     | N    | 224.00 | 224.00 | 224.00 | 224.00 | 224.00 | 224.00 |
| Min                      | 70.00 | 18.80 | 71.00 | 23.50 | 16.00 | 13.30 | 8.70 |
| Max                      | 95.40 | 41.30 | 81.00 | 42.00 | 30.30 | 33.40 | 39.00 |
| X                        | 83.62 | 36.55 | 74.87 | 34.43 | 22.30 | 17.22 | 28.11 |
| SD                       | 5.08 | 2.24 | 2.13 | 1.65 | 2.09 | 1.63 | 4.03 |
| *R. celebensis*          | N    | 219.00 | 219.00 | 219.00 | 219.00 | 219.00 | 219.00 |
| Min                      | 60.60 | 30.50 | 67.00 | 25.90 | 13.50 | 10.70 | 16.60 |
| Max                      | 97.00 | 44.30 | 81.00 | 37.30 | 26.60 | 26.30 | 33.80 |
| X                        | 78.41 | 36.23 | 73.25 | 33.64 | 20.62 | 16.76 | 26.09 |
| SD                       | 7.17 | 2.10 | 2.29 | 1.83 | 2.07 | 1.67 | 3.12 |
| *R. tangkokoensis* n. sp.| N    | 9.00  | 9.00  | 9.00  | 9.00  | 9.00  | 9.00  |
| Min                      | 65.50 | 31.30 | 72.00 | 29.30 | 20.70 | 15.10 | 16.70 |
| Max                      | 91.90 | 38.80 | 86.00 | 38.30 | 23.60 | 20.10 | 25.90 |
| X                        | 80.87 | 35.71 | 79.00 | 34.30 | 22.33 | 18.07 | 21.30 |
| SD                       | 8.94 | 2.72 | 5.77 | 3.60 | 1.11 | 1.47 | 3.26 |