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Molecular profiling of different morphotypes under the genus *Rhynchophorus* (Coleoptera: Curculionidae) in Central and Southern Philippines

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Rhynchophorus ferrugineus* Olivier and *Rhynchophorus schach* Olivier have long been reported as the two most common species of palm weevils in the Philippines. In 2008, surveys conducted in Agusan del Sur, Davao del Sur, Davao City and Cebu revealed more erstwhile unreported palm weevil morphotypes which led to perplexity in taxonomic classification. This study was conducted to determine and ascertain the genetic variation and phylogenetic relationship among 10 collected morphotypes, including *R. ferrugineus* and *R. schach,* using sequences of cytochrome oxidase I (COI) gene to resolve their taxonomic status. Results show that there were no marked morphometric differences among the palm weevils as supported by the COI characterization. Nucleotide sequences were subjected to phylogenetic analysis using bootstrapping, namely: neighbor-joining, parsimony and maximum likelihood. Among the phylogenetic trees generated, parsimony and maximum likelihood had similar results. Parsimony yielded low bootstrap values ranging from 2.5 to 10 while the maximum likelihood were from 1 to 7, indicating that these morphotypes can be clustered into one species. In support of these findings, low genetic variation was also observed, from 0 to 5.82% and high genetic similarity of 94.18 to 100% among morphotypes. Based on these results, it can be deduced that the morphotypes are cases of polymorphism, including the synonymy of *R. ferrugineus* and *R. schach.

Key words: Morphotypes, polymorphism, *Rhynchophorus ferrugineus*, *Rhynchophorus schach*, synonymy.

INTRODUCTION

Palm weevils under the genus *Rhynchophorus* have been reported as one of the major pests of coconut (*Cocos nucifera* L.) in the tropics (Menon and Pandalai, 1960). Two species, namely *R. ferrugineus* (Olivier) and *R. schach*

(Olivier) were identified as common in India (Menon and Pandalai, 1960) and the Philippines (Blancaver et al., 1977; Gabriel, 1976; Loyola, 1994; Pacumbaba, 1972). Palm weevils have also been reported as major herbivores of sago (*Metroxylon sagu* Rottb.) in Agusan del Sur (Abad, 1983) and, on the basis of morphological descriptions from literature accounts, identified *R. ferrugineus* and *R. schach* as the most frequently encountered species. Further surveys of sago areas in the Caraga Region, Southern Mindanao and Argao, Cebu in 2008 showed weevil collections comprising not only of the two aforementioned species but of 37 other erstwhile unreported morphotypes (Abad et al., 2008). Such situation has created confusion in terms of nomenclatural systematics of weevils at hand. It could not be determined whether there are distinct species in collections or that all collections are cases of polymorphism; likewise, it casted doubt over the species status of *R. ferrugineus* and *R. schach*. While 39 morphotypes were arbitrarily categorized into five (5) major *Rhynchophorus* groups for convenient classification and reference (Salamanes, 2010), the need to resolve whether or not and if some or all of the phenotypic forms are genotypic in nature was considered to be in order.

Pest identity is a basic knowledge that needs to be acquired so that appropriate pest management strategies can be devised in accordance with the nature and habits of the pest species. Outside the Philippines, the synonymy of *R. ferrugineus* and *R. vulneratus* has been reported at the molecular level with the aid of RAPD markers and cytochrome oxidase I (COI) sequences (Hallett et al., 2004). In fact, a recent study comparing DNA sequences of the mitochondrial COI gene confirmed *R. vulneratus* as distinct species from *R. ferrugineus* (Rugman-Jones et al., 2013). The genetic diversity of *Rhynchophorus* weevils has not yet been explored in the Philippines, particularly in the Central and Southern areas, and there are no current reports on how far its diversity has reached. New and sophisticated techniques such as molecular characterization can greatly aid in resolving species identification and phylogenetic relationship of *Rhynchophorus* in the Philippines. The COI gene from the mitochondrial DNA is usually utilized as the source for molecular characterization of different *Rhynchophorus* morphotypes to determine whether the cause of variations in form is genotypic or a case of polymorphism; with the possibility of looking at the emergence of one or few novel species. This pioneer work in the Philippines was conducted to verify the species/-subspecies categories of selected *Rhynchophorus* weevils found in Central and Southern Philippines using sequences of COI gene to determine the genetic variation among morphotypes of *Rhynchophorus*; and to ascertain the phylogenetic relationship of the morphotypes within the phylogeny of the genus *Rhynchophorus*.

MATERIALS AND METHODS

Sample collection

The weevil specimens studied by Salamanes (2010) were used for molecular characterization. Samples were obtained from different areas in Central and Southern Philippines (Figure 1). Laboratory procedures were conducted from December 2010 to January 2011 at the Molecular Biology Laboratory of the University of the Philippines Mindanao, Davao City.

Rhynchophorus species verification

Species verification of *Rhynchophorus* was done based on morphological characteristics according to the parameters set by White and Borror (1970). They were classified according to the following features: distinct coloration on its pronotum and elytra; clubbed antennae which rise near the eyes, the first segment of its antennal club being enlarged and shiny; and an exposed pygidium. Members of the genus have sizes ranging from 3 to 31 mm.

Morphometrics

The 140 weevils collected, comprising 39 morphotypes, were arbitrarily classified into five categories, namely: striped, dotted, maple leaf-shaped, black-shaded and bilateral symmetry (Salamanes, 2010). Two variants were selected from each category for a total of 10 morphotypes, which were then subjected to DNA extraction. The samples used are shown and described in Figures 2-11. The morphometric data gathered by Salamanes (2010) on the 10 selected palm weevils were summarized and incorporated in the study to reconcile it to the results obtained from the molecular characterization of *Rhynchophorus* variants using COI gene. Measurements of antenna, rostrum, head, thorax, legs, and abdomen of each weevil sample were also included. Sexes of the samples were determined by examining the presence or absence of hair-like structures at the anterior part of the rostrum. Presence of these structures indicates that the weevil is a male; otherwise, the specimen is a female.

DNA extraction

DNA samples from 10 palm weevils in storage were processed following the protocol of Bastian et al. (2008). Briefly, the genomic DNAs were extracted from the leg muscle tissues of each weevil

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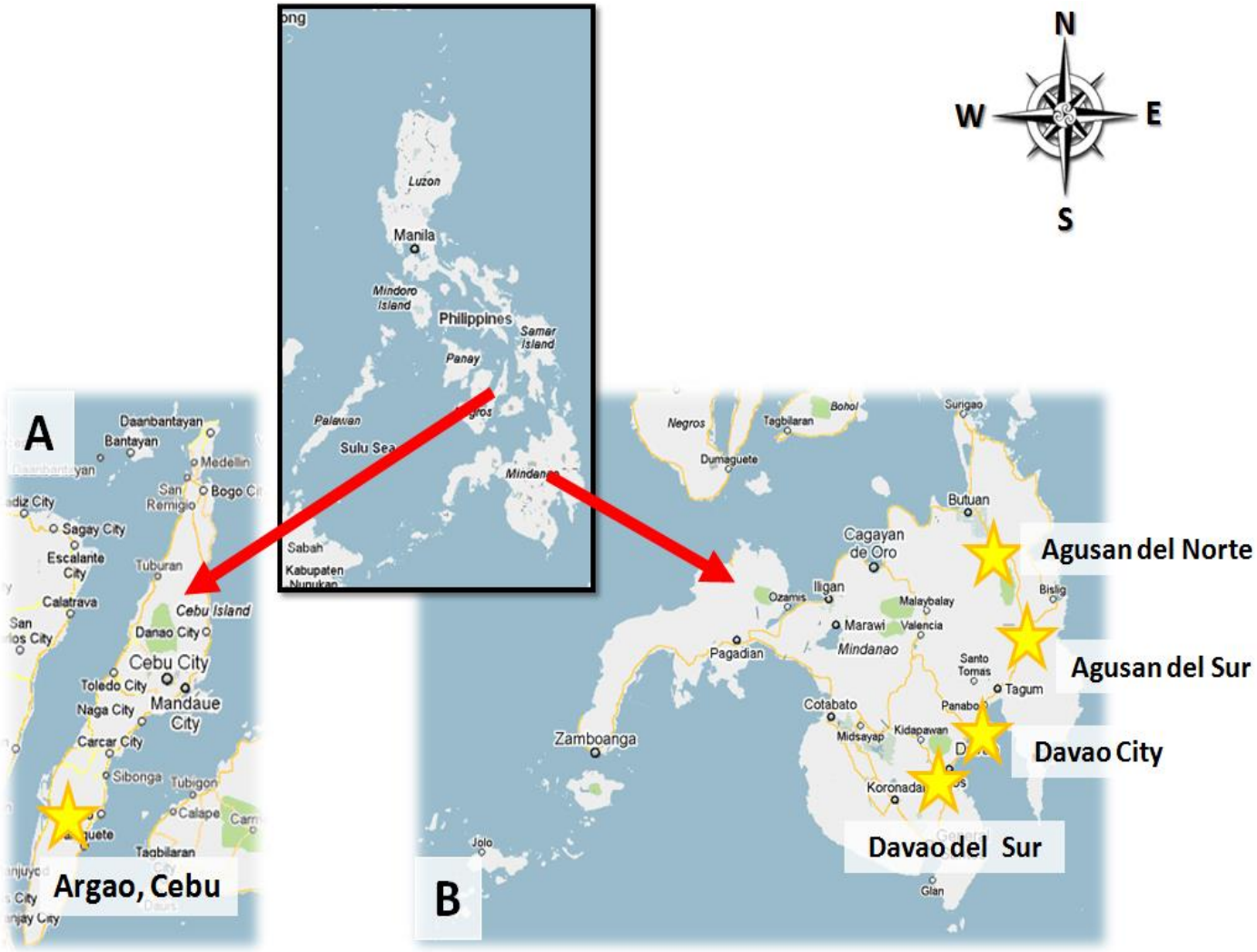

Figure 1. Map of the Philippines showing the sites where samples were obtained: (A) Argao, Cebu; (B) Davao City, Davao del Sur, Agusan del Norte and Agusan del Sur (Google Earth, 2011).

and preserved in 95% ethanol. Three out of six legs of each weevil were prepared by washing six times with 100 mM EDTA pH 8.0 solution prior to extraction. Proteinase K digestion, phenol/chloroform extraction and ethanol precipitation were then employed.

Polymerase chain reaction

PCR conditions were optimized using Veritti Thermal Cycler (Applied Biosystems) and the PCR mix was prepared according to the instructions stated in the PCR GoTaq® PCR Core System I manual (Promega, USA). The primers used were short F and short R, to amplify the COI gene with approximately 220 bp product (Gilbert et al., 2007). The final optimized conditions used were 94°C for 1 min for pre-denaturation, 35 cycles of denaturation at 94°C for 1 min, annealing at 45°C for 1 min, extension at 72°C for 1 min and a final elongation at 72°C for 5 min and finalized with a holding temperature of 4°C. PCR products were then subjected to electrophoresis and DNA bands were visualized under ultraviolet transilluminator (UVITEC Limited, UK).

DNA sequencing and phylogenetic analysis

PCR products were purified using Purification Wizard SV Gel and PCR Clean-up System (Promega, USA) following the manufacturer's protocol. Purified PCR products were shipped to Macrogen Inc., Korea for DNA sequencing services. Alignment of DNA sequences was done to identify regions of similarity on *Rhynchophorus* species using the WSSADI program (Polinar, 2005). After the alignment,

Figure 2. Striped B ♀ (Arl4-16) collected from Lansones, Rosario, Agusan del Norte.

Figure 4. Dotted B ♀ (Arl6-D5) collected from Lansones, Rosario, Agusan del Norte.

Figure 3. Striped C ♀ (Anli-8) collected from Libas, Agusan del Norte.

Figure 5. Dotted J ♀ (Ala3-33) collected from Lamacan, Argao, Cebu.

Figure 6. Maple leaf-shaped E ♀ (Alu3-23) collected from Luwak, Argao, Cebu.

Figure 7. Maple leaf-shaped G ♀ (Alula-21) collected from Luwak-Lamacan, Argao, Cebu.

Figure 8. Black-shaded B ♀ (Ala2-25) collected from Lamacan, Argao, Cebu.

Figure 9. Black shaded H♂ (Ala3-35) collected from Lamacan, Argao, Cebu.

Figure 10. Bilateral symmetry B♂ (Ala2-28) collected from Lamacan, Argao, Cebu.

Figure 11. Bilateral symmetry E♂ (Ata1-11) collected from Talaytay, Argao, Cebu.

mutations that might be useful in the analysis of the phylogenetic tree of the species *Rhynchophorus* were identified.

Aligned nucleotide sequences were subjected to SEQBOOT with 1000 bootstrap replications. To obtain the genetic similarity (percentile) matrix, the bootstrap replicates were subjected to a program called DNADIST software using the two-parameter model (Kimura, 1980). For the phylogenetic tree regeneration, the programs DNAPARS, DNAML and NEIGHBOR were used for parsimony, maximum likelihood and neighbor-joining, respectively. All softwares are included in the Phylogenetic Inference Package (PHYLIP) v3.68.

DNA sequences of the COI gene of different weevil samples were processed and submitted to the DNA databank.

RESULTS

Morphotypes of *Rhynchophorus*

The 10 samples, two from each morphotype groupings, included in this study are presented in Table 1 and described in Figures 2-11. These palm weevils, bearing their own unique markings on the thorax region, were collected from areas in Central and Southern Philippines.

Palm weevils categorized under this morphotype are characterized by having a bottle like stripe at the center of its thorax. The stripe is smoothly curved at the mid and upper portion. Forty specimens of both sexes were collected (Figure 2). This morphotype group has a central stripe with curvatures on both sides of its base with a distinct bulge at the middle. The anterior part of the stripe near the rostrum has a tapered like appearance which resembles a bottle neck. A total of four individuals comprising both sexes were collected (Figure 3). They are characterized as those having two pairs of dots placed symmetrically, the upper pair of dots smaller than the lower diamond-shaped dots. A pair of faint black vertical lines can also be observed along the center of the thorax; and its elytra have colored striations. A total of two female individuals were collected (Figure 4). Weevils under this group have similarities with the Dotted B except that it has a distinct marking at the mid right portion of its thorax. Its two pairs of dots are located symmetrical to one another with the lower pair bigger and shaped like a diamond compared to the upper pair. Colored striations can be observed on its elytra. Only one female weevil was collected (Figure 5). This group of weevils has a pair of blotches at the upper periphery of the thorax. Its maple leaf-shaped pattern is smooth and rounder at the side with the tip of the central blotch being sharp. A small streak of red-orange can be observed at the center of the thorax from posterior to mid part. Only one female weevil was collected (Figure 6). Palm weevils under this group have a maple leaf-like outline and bean-shaped markings on their upper peripheral part; a slit can also be seen on the posterior end to the mid portion of the thorax. Colored striations on the elytra are also present. One female weevil was collected (Figure 7).

Table 1. *Rhynchophorus* morphotypes collected from Agusan del Norte and Cebu, Philippines.

Morphotype	Code	Location
Striped B♀	Arl4-16	Lansones, Rosario, Agusan del Norte
Striped C♀	Anli-8	Libas, Agusan del Norte
Dotted B♀	Arl6-D5	Lansones, Rosario, Agusan del Norte
Dotted J♀	Ala3-33	Lamacan, Argao, Cebu
Maple leaf-shaped E♀	Alu3-23	Luwak, Argao, Cebu
Maple leaf-shaped G♀	Alula-21	Luwak-Lamacan, Argao, Cebu
Black-shaded B♀	Ala2-25	Lamacan, Argao, Cebu
Black-shaded H♂	Ala3-35	Lamacan, Argao, Cebu
Bilateral symmetry B♂	Ala2-28	Lamacan, Argao, Cebu
Bilateral symmetry E♂	Ata1-11	Talayatay, Argao, Cebu

Source: Salamanes (2010).

The thoraces of the weevils that belong to this group have a paired pattern that appear like a stalactite with a thin flare-like orange marking around it and an arch-like pattern prevailing it. No colored striations were observed. Only one female weevil was collected (Figure 8). A pair of boot-like blotches on the anterior region of the thorax was observed. The blotches resemble the land outline of Italy. A minute diamond-shaped space among the black markings at the mid portion of the thorax and a faint orange vertical line at the posterior region can be observed. Six weevils of both sexes were collected (Figure 9). Weevils under this group have a pair of markings, symmetrically placed on both sides and resemb-ling foliage on their thoraces. Another pair of elongated blotches is present at the mid upper portion of the thorax. Its elytra have colored striations. Two indivi-duals of both sexes falling under Bilateral Symmetry B were collected (Figure 10). This group of weevils is charac-terized with a pair of black markings superimposed on their prominently orange colored thorax. These markings give an orange-colored stripe along the mid-thoracic region. A total of four individuals, male and female were collected (Figure 11)

Morphometrics

No remarkable differences were observed on the sizes of the head, thorax, abdomen, rostrum and antennae among the 10 *Rhynchophorus* morphotypes, including *R. schach* and *R. ferrugineus* (Table 2). The same is true for the femur, tibial and tarsal lengths (Table 3).

Genetic variation of *Rhynchophorus* morphotypes

The amplified COI genes of the 10 *Rhynchophorus* morphotypes were 220 bp and produced low genetic variation among the 10 samples (Figure 12 and Table 4). Genetic variation and genetic similarity were then determined based on the sequences.

The highest genetic variation yielded only 5.82% between Anli-8 and Ala2-25 while the lowest genetic variation yielded 0.88% between Arl4-16 and Anli-8. Arl4-16 and Ata1-11, Anli-8 and Ata1-11, Anli-8 and Alula-21, Arl6-D5 and Alu3-23 showed no genetic variation.

Low genetic variation among 10 *Rhynchophorus* morphotypes suggests that there may have been insufficient time for these morphotypes to completely diverge into two significantly different species. Genetic similarity ranged from 94.18 to 100% which is very high in terms of likeliness for specific morphotypes to be considered as distinct species belonging to *R. ferrugineus* or *R. schach* (Table 5). Furthermore, samples with genetic variation ranging from 3 to 5% suggest that these morphotypes may be exemplifying the emergence of a novel subspecies.

Phylogenetic tree of *Rhynchophorus* morphotypes

To ascertain the phylogenetic relationship among *Rhynchophorus* morphotypes, three methods were used: neighbor-joining, parsimony and maximum likelihood. Among the three, parsimony and maximum likelihood presented similar results thus, they were subjected to further analysis and the outcomes were compared. *Drosophila melanogaster*, a member of the class Insecta was used as outgroup for the phylogenetic tree reconstruction (Figure 13). An outgroup was needed for the first outbranching of the tree. Bootstrap values, as indicated at each node of the tree were the basis for determining the relatedness of the 10 morphotypes. For

Table 2. Mean length and width of head, thorax, abdomen, rostrum and antennae of the different *Rhynchophorus* morphotypes[z].

Morphotypes	Length (mm)					Width (mm)			
	H	T	Ab	R	An	H	T	Ab	R
Arl4-16	2.53 (±0.50)	12.43 (±1.65)	19.04 (±2.73)	10.59 (±1.24)	7.81 (±1.51)	3.85 (±0.49)	10.83 (±1.42)	13.48 (±1.69)	1.61 (±0.21)
Anli-8	1.85 (±0.15)	10.75 (±1.58)	16.08 (±1.67)	9.30 (±1.64)	6.90 (±0.78)	3.50 (±0.22)	9.23 (±1.46)	11.33 (±1.86)	1.43 (±0.15)
Arl6-D5[y]	2.90	12.80	20.20	11.90	8.10	4.60	11.70	14.30	1.80
Ala3-33[y]	1.70	11.50	16.50	9.90	6.20	3.30	9.90	12.20	1.30
Alu3-23[y]	2.40	12.00	17.90	9.80	6.80	3.50	10.20	12.40	1.40
Alula-21[y]	2.00	10.30	15.30	8.80	6.10	3.50	9.10	11.10	1.30
Ala2-25[y]	2.20	11.20	16.60	9.90	6.10	3.60	9.70	11.80	1.40
Ala3-35	1.72 (±0.22)	10.88 (±0.80)	16.06 (±1.68)	8.83 (±1.05)	6.58 (±0.57)	3.33 (±0.18)	9.53 (±0.85)	11.74 (±1.08)	1.37 (±0.11)
Ala2-28[y]	1.90	10.60	15.80	8.80	6.40	2.90	9.30	11.30	1.50
Ata1-11	1.85 (±0.23)	10.75 (±0.58)	15.80 (±1.27)	8.45 (±0.41)	6.25 (±0.17)	3.13 (±0.16)	9.13 (±0.57)	11.33 (±0.69)	1.33 (±0.11)

H, Head; T, thorax; Ab, abdomen; R, rostrum; An, antennae. .[z]Source: Salamanes (2010). [y]only 1 or 2 specimens.

Table 3. Mean length of femur, tibia and tarsus of the different *Rhynchophorus* morphotypes according to segments z.

Morphotypes	Femur Length (mm)			Tibia Length (mm)			Tarsus Length (mm)		
	FL	ML	HL	FL	ML	HL	FL	ML	HL
Arl4-16	6.66 (±0.96)	6.72 (±0.94)	7.44 (±1.06)	6.76 (±1.20)	5.37 (±1.10)	6.37 (±0.93)	5.69 (±0.92)	5.44 (±1.00)	5.57 (±0.83)
Anli-8	5.75 (±0.90)	5.83 (±1.20)	6.48 (±0.99)	5.73 (±1.31)	4.28 (±1.02)	5.80 (±1.10)	4.68 (±0.87)	4.68 (±1.02)	4.73 (±0.59)
Arl6-D5[y]	7.10	7.10	8.00	6.70	5.40	6.00	5.90	6.00	5.80
Ala3-33[y]	5.10	5.40	6.30	5.10	4.50	5.40	4.30	4.20	---
Alu3-23[y]	4.90	5.70	6.80	5.70	4.70	6.20	5.10	5.10	5.00
Alula-21[y]	5.10	5.00	6.10	5.00	4.40	5.00	4.40	4.40	4.40
Ala2-25[y]	5.00	5.20	5.60	4.80	4.10	5.10	3.90	---	4.50
Ala3-35	5.15 (±0.43)	5.38 (±0.50)	5.80 (±0.48)	5.35 (±0.37)	4.48 (±0.50)	5.23 (±0.50)	4.50 (±0.59)	4.42 (±0.47)	4.48 (±0.52)
Ala2-28[y]	5.50	5.40	6.20	5.60	4.10	5.10	4.30	4.60	4.40
Ata1-11	5.25 (±0.32)	5.40 (±0.12)	5.90 (±0.12)	5.15 (±0.15)	4.30 (±0.12)	5.25 (±0.25)	4.60 (±0.22)	4.63 (±0.23)	4.58 (±0.22)

FL, Foreleg; ML, Middle leg; HL, Hind leg. [z]Source: Salamanes (2010). [y]only 1 or 2 specimens.

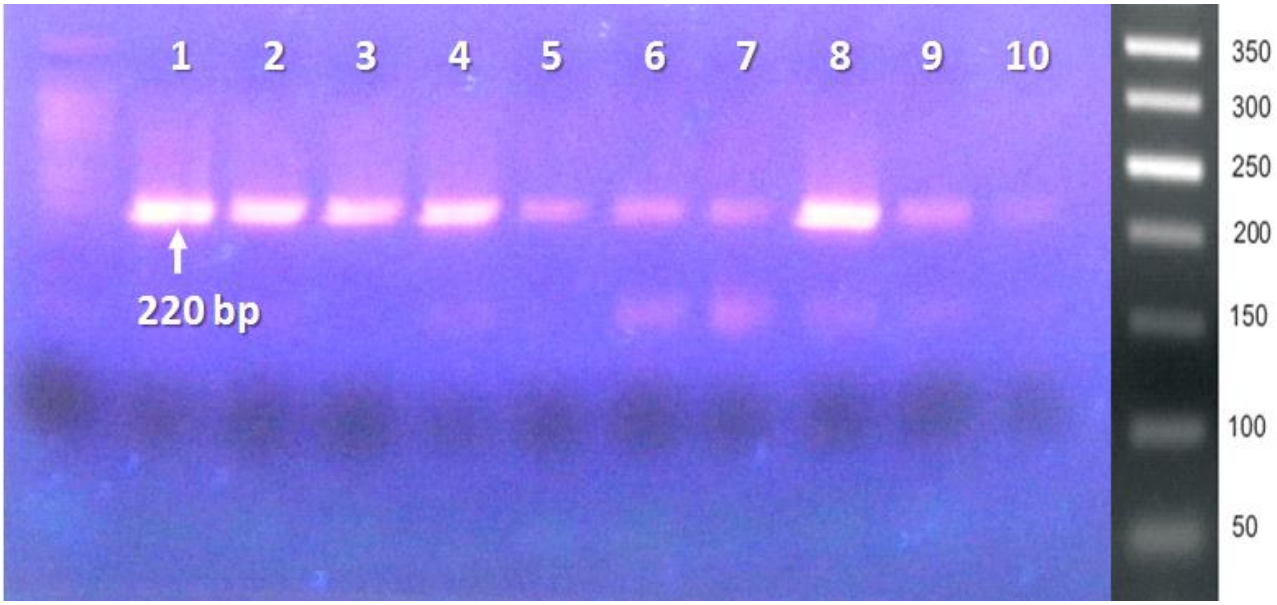

Figure 12. The amplified COI genes of the 10 *Rhynchophorus* morphotypes: 1, Arl4-16; 2, Anli-8; 3, Arl6-D5; 4, Ala3-33; 5, Ala2-28; 6, Ata1-11; 7, Ala2-25; 8, Ala3-35; 9, Alu3-23; 10, Alula-21 using the short F and short R primers (Gilbert et al., 2007).

Table 4. Genetic variation (%) of 10 morphotypes of *Rhynchophorus* from different locations based on COI gene sequences.

	1	2	3	4	5	6	7	8	9	10
Arl4-16	0.000000									
Anli-8	0.878470	0.000000								
Arl6-D5	4.312502	4.188519	0.000000							
Ala3-33	4.641839	4.922578	2.741262	0.000000						
Ala2-28	3.596298	4.136224	3.618501	3.744180	0.000000					
Ata1-11	-1.000000	-1.000000	2.128683	3.532978	5.092266	0.000000				
Ala2-25	4.627843	5.820000	3.368906	4.177738	3.680079	4.944946	0.000000			
Ala3-35	4.848904	3.891408	3.323647	4.194904	4.187688	2.409259	4.594211	0.000000		
Alu3-23	4.779708	4.908820	-1.000000	4.708742	4.987939	4.679949	5.267461	5.058018	0.000000	
Alula-21	4.812748	-1.000000	4.750243	1.315442	3.666670	4.958101	2.036880	3.571055	4.009349	0.000000

(1), Arl4-16; (2), Anli-8; (3), Arl6-D5; (4), Ala3-33; (5), Ala2-28; (6), Ata1-11; (7), Ala2-25; (8), Ala3-35; (9), Alu3-23; (10), Alula-21.

morphometrics between the two *Rhynchophorus* "species", *R. schach* (categorized under striped group) and *R. ferrugineus* (under the dotted group), except for the body coloration or markings along with the other morphotypes, the characterization of COI gene of the 10 morphotypes revealed the relatedness of these weevils. the parsimony, low bootstrap support was observed with values ranging from 2.50 to 10.00. The same was observed for maximum likelihood with bootstrap values ranging from 1.00 to 7.00. Since trees generated from the two methods presented bootstrap values lower than 50, all of the 10 morphotypes of *Rhynchophorus* can be clustered into one group thus, supporting opinions that these morphotypes belong to one species of *Rhynchophorus*.

DISCUSSION

Variations in the morphology, specifically on thoracic markings of *Rhynchophorus* may be attributed to genetic

Table 5. Genetic similarity percentile matrix among *Rhynchophorus* morphotypes using COI gene sequences.

	1	2	3	4	5	6	7	8	9	10
Arl4-16	100.00									
Anli-8	99.12	100.00								
Arl6-D5	95.69	95.81	100.00							
Ala3-33	95.36	95.08	97.26	100.00						
Ala2-28	96.40	95.86	96.38	96.26	100.00					
Ata1-11	100.00	100.00	97.87	96.47	94.91	100.00				
Ala2-25	95.37	94.18	96.63	95.82	96.32	95.06	100.00			
Ala3-35	95.15	96.11	96.68	95.81	95.81	97.59	95.41	100.00		
Alu3-23	95.22	95.09	100.00	95.29	95.01	95.32	94.73	94.94	100.00	
Alula-21	95.19	100.00	95.25	98.68	96.33	95.04	97.96	96.43	95.99	100.00

(1), Arl4-16; (2), Anli-8; (3), Arl6-D5; (4), Ala3-33; (5), Ala2-28; (6), Ata1-11; (7), Ala2-25; (8), Ala3-35; (9), Alu3-23; (10), Alula-21.

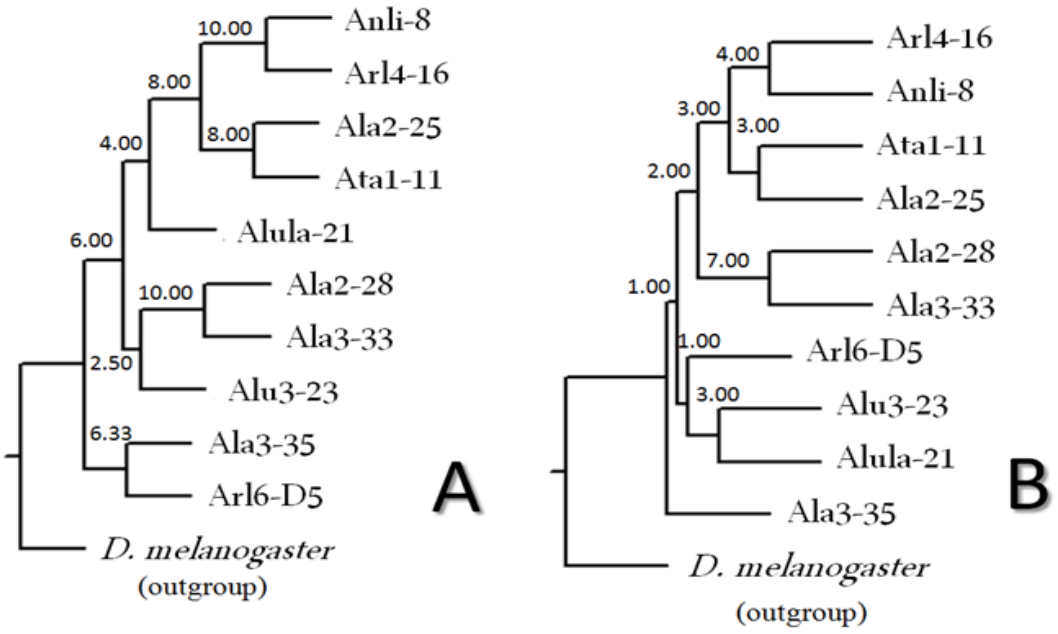

Figure 13. Phylogenetic tree derived from (A) parsimony and (B) maximum likelihood for 10 *Rhynchophorus* morphotypes with *Drosophila melanogaster* as an outgroup. Bootstrap values are indicated at each node.

drift, polymorphism or phenotypic plasticity wherein geographical distribution may have contributed to its distinct modification. Sexual dimorphism cannot be a factor among these morphological transformations since both male and female sexes were observed on the majority population of the five main categories of *Rhynchophorus* morphotypes. Nonetheless, several groups exemplified genetic variation percentile ranging from 3 to 5% suggesting an emerging of a subspecies.

Based on the results obtained, the synonymy of *R. ferrugineus* and *R. schach* may be deduced. The findings parallel that of Hallett et al. (2004) who, by using RAPD markers and COI sequences, showed that the *R. ferrugineus* and *R. vulneratus* from Java are of the same

species. Thus, molecular characterization was a defining work to elucidate the species status of the morphotypes. With the trivial differences in Molecular characterization utilizing the COI gene was successfully carried out. Among the subunits of cytochrome oxidase, COI was preferred due to its highly conserved yet variable regions closely associated to the mitochondria (Lunt et al., 1996). The COI is unique in its ability to discriminate closely related species in all animal phyla except Cnidaria (Hebert et al., 2003).

Phenotypic plasticity is a phenomenon that covers all types of environmentally- induced phenotypic variation (Stearns, 1989). It is the ability of a single genotype to produce more than one phenotype, be it morphological, physiological or behavioral in response to environmental conditions (West-Eberhard, 1989). However, epigenetic systems generally drive such changes. Mechanisms other than DNA sequences are responsible for the phenotypic variation in epigenetics. Polymorphism occurs when a species have more than one morph existing in the same population that inhabits the same time and space. These morphs must belong to a panmictic population (Ford, 1965). Such population assumes random mating and that every individual is a potential mate. Polymorphism can be passed on to progenies with modifications by natural selection. The genetic make-up or DNA sequences determine which morphs will be expressed by a population.

From the characterization of the COI gene of the 10 different *Rhynchophorus* morphotypes, it is more likely that polymorphism could be the main factor which caused such morphological modifications. Genetic variation among the morphotypes is too high to be considered as due to phenotypic plasticity driven by epigenetic systems whereas, its high similarity with one another still indicates that they belong to one species.

Geographical barrier leading to reproductive isolation is speculated to be a factor for the emergence of different morphotypes. Cebu, has only small patches of sago plantings in the sites visited, the biggest area of which was just about two hectares. This may have limited the breeding area of the weevils leading to more inbreeding thus giving greater chances for rare morphotypes to emerge. Consequently, large sago areas in Mindanao, especially in the provinces of Agusan, are common. This should allow gene flow over large populations of weevils to operate where rare traits could seldom emerge.

It is of interest to note that the photographs of the collections by Rugman-Jones et al. (2013) from the northern island of the Philippines contained both the erstwhile considered *"ferrugineus"* type (with black dots on the thorax region) and the *"shach"* type (with orange to red stripe on the thorax region) and now classified on the basis of results of their molecular study as all belonging to the *R. ferrugineus* species. While the outcome of this study concurs with their findings, Rugman-Jones et al. (2013) found no genetic evidence of *R. ferrugineus* in the 150 palm weevils collected from Java. They added that Hallett et al. (2004) may not have encountered *R. ferrugineus* in Java at all and instead worked only with a *R. vulneratus* population showing high levels of color variation. Rugman-Jones et al. (2013) also hinted the possibility of finding *R. vulneratus* in Mindanao, obviously due to the relative proximity of the southern Philippine island to Java.

The revealed synonymy of *R. ferrugineus* and the commonly referred to as *R. schach* in the Philippines has great implications for farmers specifically those who grow crops such as *C. nucifera* and *M. sagu*, plants that are both susceptible to palm weevils. Preventive and mitigating procedures for their destructive effects can now be formulated with relative ease as both morphotypes are of the same species and have the same biological properties. As a result, an applicable pest management strategy can now be devised to both morphotypes. This could also mean that in the long run, less input would be entailed in dealing with *Rhynchophorus* infestation.

Conflict of Interests

The author(s) have not declared any conflict of interests

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