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Original article

A morphometric analysis of *Apis cerana* F and *Apis nigrocincta* Smith populations from Southeast Asia

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Summary — *Apis cerana* F and *Apis nigrocincta* Smith, two species of Asian cavity nesting honey bees, were analyzed for interpopulation morphological variation using conventional morphometric techniques. The well-known honey bee subspecies *Apis cerana cerana* and *A c japonica* are confirmed as being distinct from the rest of *A cerana*, even though the analysis removed the confounding influence of 'overall size' from the data set. There are at least three other morphologically distinct groups: one ranging from Sri Lanka through to Flores and central Sulawesi, a second group so far only found in southern Sulawesi, and a group from Timor. A large phenetic shift has occurred in the second Sulawesi group, indicating that it may have evolved in isolation for a long period of time, or undergone rapid divergence following a more recent bottleneck event. *A nigrocincta* is shown to be present over most of western Sulawesi, on Mindanao and on Sangihe, a small island between Mindanao and Sulawesi. The Mindanao and Sulawesi forms are distinct, but the small sample size did not allow for subspecific recognition. Bees from Luzon, previously identified as *A c philippina*, are shown to be very distinct from both *A cerana* and *A nigrocincta*, and the possibility of their belonging to the species *A nigrocincta* is discussed.

***Apis cerana* / *Apis nigrocincta* / morphometrics / Southeast Asia / honey bees**

INTRODUCTION

The honey bees of the world (*Apidae*: *Apis* spp) are widespread over their native range of Europe, Africa and Asia, and have been successfully transplanted to other regions and continents (Ruttner, 1988). They have been well studied over most of their range, and have been assigned to many subspecies (races or geographical variants) on the basis of morphometric analyses. Understanding of honey bee morphological variation is incomplete, however. New species (*Apis nigrocincta*, Hadisoesilo and Otis, 1996;

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Apis nuluensis, Tingek et al, 1996) have recently been discovered from Sulawesi and Borneo, respectively, and their ranges have not yet been adequately determined. No inclusive study of other Southeast Asian island bee populations has been carried out either, although Ruttner (1988) said that if new forms were to be discovered, that is where they were most likely to be found.

Recently, Smith and Hagen (1996) published an analysis of mitochondrial DNA variation among populations of cavity nesting honey bees in Asia, which attempted to phylogenetically link the various populations. They were able to resolve four monophyletic lineages that are relevant to this study: 1) *Apis cerana indica* from Sri Lanka and other parts of India; 2) *A c cerana* from China and *A c japonica* from Japan; 3) *A c indica* from Malaysia and Indonesia; and 4) *A nigrocincta* (see Hadisoesilo and Otis, 1996) from Sulawesi with *A c philippina* (sensu Ruttner, 1988) from the Philippines. Unfortunately, the variation between the samples was too great to root the resulting tree, and consequently they were not able to discern phylogenetic relationships between them (Smith and Hagen, 1996). Both Smith and Hagen (1996) and our study used many of the same samples. Therefore this can be seen as a companion study to theirs, investigating morphological variation as opposed to genetic variation. It was our intention to survey the major islands of Southeast Asia where cavity nesting honey bees are known to exist, and to do a preliminary assessment of their interpopulation morphometric variation.

MATERIALS AND METHODS

Collection data and measurement techniques

Honey bees were collected from natural nests and semi-managed hives from many sites in Malaysia, Indonesia and eastern Asia (see *Appendix* and fig 1). For the analysis, one sample consisted of ten bees from a single colony. Many of the samples were collected by the authors, but some were collected by others and sent to us (see *Acknowledgments*). The bees were killed with ether and preserved in 75% ethanol. The right fore and hind wings were removed and mounted on glass slides with glycerin. The right hind legs were removed, disarticulated and mounted in the same manner. A second glass slide was placed on top to ensure that the parts stayed parallel to the plane of measurement (coverslips were not heavy enough to prevent the parts rolling and the wings folding). No distortion due to compression was detectable.

The specimens were placed on the stage of an inverted microscope and an image of the body parts was projected onto paper. Magnifications were ×45 for most measurements, or ×21.5 for the wing lengths. Distance measurements were made with a ruler and were accurate to the nearest 0.01 mm, and wing angles were accurate to the nearest tenth of a degree, measured with a protractor. The values of the variables for each sample were calculated as the arithmetic average over the ten bees from that hive. Consistency was achieved in that one person (MSD) performed all of the measurements. In total, 98 samples were analyzed.

The characters measured are listed in table I. The fore wing and leg characters are the same as described by Ruttner (1988) and Ruttner et al (1978), while the hind wing characters are those of Mattu and Verma (1984). One new character, width of the tibia, was introduced. It was defined as the width of the distal end of the metatibia.

Morphometric analyses

Plots of the raw data, taken two variables at a time, were made to determine if they could discriminate between the samples taken from the different localities.

Cluster analysis (using UPGMA and Ward's minimum variance and Euclidean distance options in SYSTAT; Wilkinson 1990) assigned samples to groups of overall similarity, and these groups were entered into a multiple-group principal components analysis, which determined the characters most correlated with 'size' [see Thorpe (1988) and Daly (1992)]. The MG-PCA was used simply to determine those variables correlated with 'size', and not to compare popu-

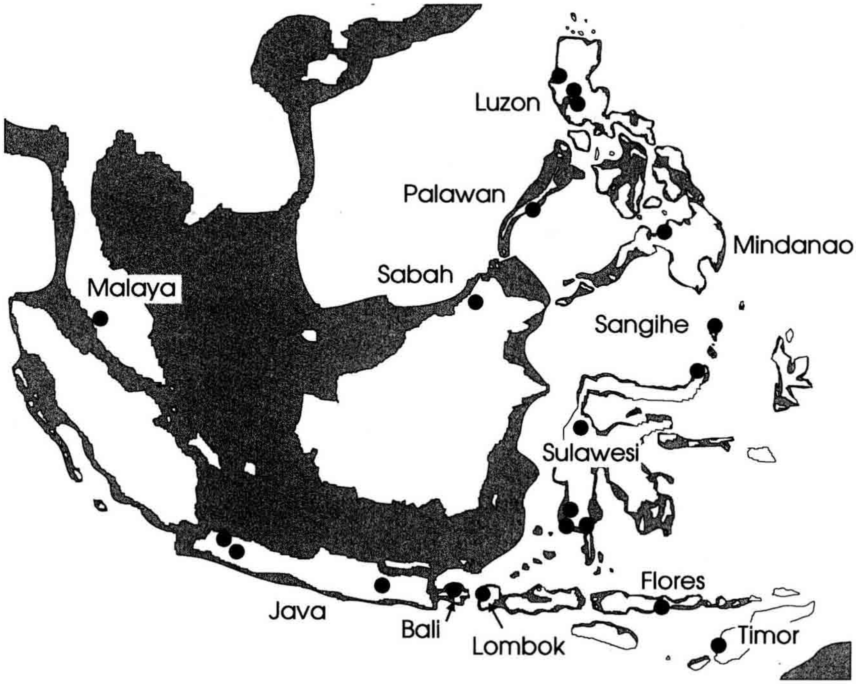

Fig 1. Map of Southeast Asia showing collection sites. Sri Lanka, Hong Kong and Japan not shown. Dark area indicates 200 m isobath, or approximate extent of maximum land exposure during past ice age.

lations. Variables identified as 'size' variables were regressed on hind wing length (the variable with the highest correlation, > 0.95, with PCA 1 in single-group analyses), and those that had high significant correlations with hind wing width ($R^2 > 0.6$, $P < 0.05$) were classified as size-related. The overall influence of size was removed from those characters using the computer routine 'Burnaby' recommended by Rohlf and Bookstein (1987) and available by anonymous ftp from life.bio.SUNYSB.edu/morphmet as part of the 'Macleod' package. The new 'size-free' variables were rejoined with the original variables that were shown to be uncorrelated with 'size' into a new data set. This was the size-free data set used for the rest of the analyses.

The size-free data set was subjected to a simple principal component analysis as an exploratory analysis to determine whether any of the recognized subspecies or samples from localities not studied or reviewed by Ruttner (1988) formed distinct clusters.

Smith and Hagen's (1996) results suggested that the bees known as *A c philippina* from the Philippines [both Luzon and Mindanao according to Ruttner (1988)] may be more closely related to *A nigrocincta*, since they have similar DNA sequences in the area of the mitochondria they investigated. A discriminant analysis (DA) was designed, assigning those bees known to be *A cerana* (from Ruttner, 1988) and *A nigrocincta* (from Hadisoesilo and Otis, 1996) to two groups, and testing to which of the two groups the bees from the Philippines and one other group from the island of Sangihe (half-way between Sulawesi and Mindanao, see fig 1) belonged. A second DA was performed in which the Philippine samples were placed into their own a priori group, to determine their relative fit to each other.

Table I. List of characters measured and their preliminary statistical properties. Those in **bold** were size-related and were entered into the program 'Burnaby'. Leg and fore wing characters are as described in Ruttner et al (1978) and Ruttner (1988); hind wing characters as described in Mattu and Verma (1984). Metatibia width was a new character, and is defined as the width of the distal tip of the metatibia.

Metafemur length
Metatibia length
Metatibia width
Metatarsus length
Metatarsus width

Fore wing length
Fore wing width
Length of first abcissa of vein M_{3+4}
Length of second abcissa of vein M_{3+4}
Angle A4
Angle B4
Angle D7
Angle E9
Angle G18
Angle J10
Angle J16
Angle K19
Angle L13
Angle N23
Angle O26

Hind wing length
Hind wing width
Number of hamuli
Extent of hamuli
Length of vannal lobe
Length of jugal lobe
Length of basal portion of radial vein
Length of apical portion of radial vein
Length of 'indica' vein
Length of discoidal vein

For further analysis of the two species *A cerana* and *A nigrocincta*, non-metric multidimensional scaling ordinations were performed as recommended by de Queiroz and Good (1997), and confirmatory analysis was carried out with discriminant analysis.

All statistics were performed with SYSTAT version 5.0 for DOS (Wilkinson, 1990).

RESULTS

Several plots of the raw variables were searched for patterns of variation between the groups. Plots of lengths versus widths of wings and leg segments showed significant discrimination between the samples, and separated the two species *A cerana* and *A nigrocincta* from one another and the subspecies *A c cerana* (the samples from Hong Kong) and *A c japonica* (from Japan) from the rest of *A cerana* (fig 2). They also highlighted the reason behind the desire to remove the effect of 'size' from the variables. The smallest bees measured were the samples from Luzon, the largest from Japan, and they occupy opposite ends of the graph (fig 2). It was not desired that populations be classified by their size differences, and the removal of overall size from the variables allowed us to analyse samples by their shape, instead of by size.

UPGMA clustering of the data identified 16 groups, mostly arranged by location of collection. One sample from Timor was grouped into the cluster of Malay peninsula samples, another Timorese sample clustered with the samples from Flores, and one Flores sample clustered with the south Sulawesi samples. The Javanese samples (western and eastern) were all in one group, and one Balinese sample clustered with them. One sample from Sulawesi *A nigrocincta* clustered with the Mindanao group, and two from Sulawesi *A nigrocincta* clustered with the Sangihe group. A discriminant analysis, performed with the 16 groups, reassigned these samples into their respective islands (with probability > 0.95). The 16 groups entered into the MGPCA were: Java, Bali, Lombok, Flores, Timor, South Sulawesi (Sulawesi *a*), central Sulawesi (Sulawesi *b*), Sulawesi *A nigrocincta*, Sangihe, Malay peninsula, Sabah/Palawan,

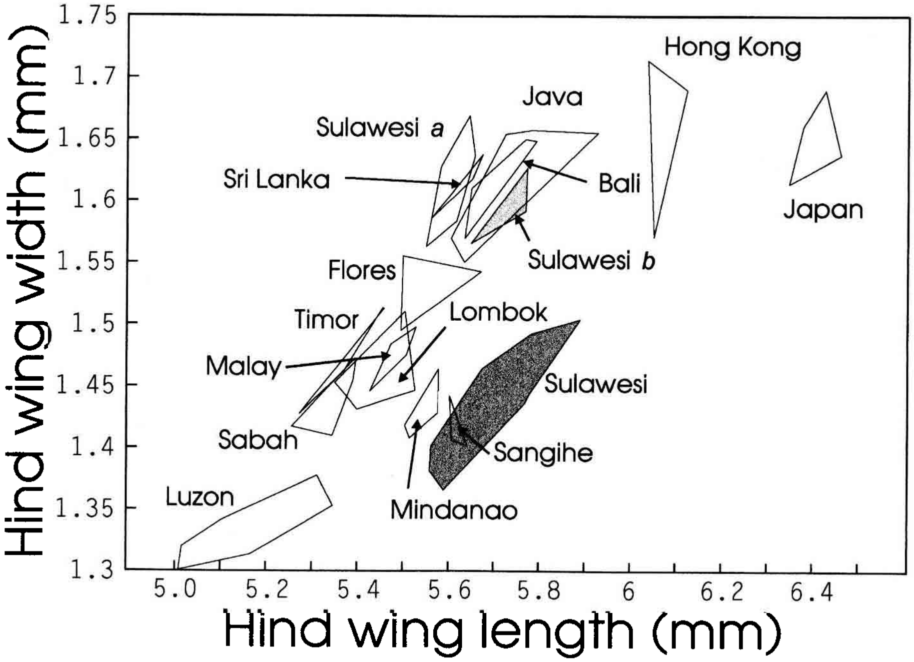

Fig 2. Graph of hind wing length versus width. Several groups are apparent even in this two variable scatterplot. Dark polygonal shape is *A nigrocincta* (sensu Hadisoesilo and Otis, 1996), pale grey shape is Sulawesi *b* from central Sulawesi.

Luzon, Mindanao, Hong Kong, Japan and Sri Lanka. Those variables that had their highest absolute correlations with factor 1 of the MG-PCA and had high significant correlations with hind wing length were classified as size related. They are highlighted in table I.

The principal component analysis of the size-free data set, which compressed 62% of the variation in the samples into the first two components (fig 3), revealed many of the previously known subspecies of *A cerana*. Both *A c cerana* from Hong Kong and *A c japonica* from Japan were well isolated from the rest of the samples as was *A nigrocincta* from Sulawesi (dark grey polygonal shape). The Mindanao and Sangihe samples plotted with the samples of *A nigrocincta*, and the samples from Luzon were nearby. The samples of Sulawesi *A cerana* were isolated into two distinct groups, one (Sulawesi *a* from southern Sulawesi; clear polygonal shape) was separate from but adjacent to the Javanese samples, and one from central Sulawesi (Sulawesi *b*; light grey polygonal shape) plotted within the Javanese samples. The samples from Sabah and Timor formed distinct groups, and the samples from Sri Lanka overlapped slightly with the samples from Java.

The discriminant function analysis performed to identify the Philippine and Sangihe bees placed the Sangihe and Mindanao samples and four of the seven Luzon samples in with *A nigrocincta* (probability

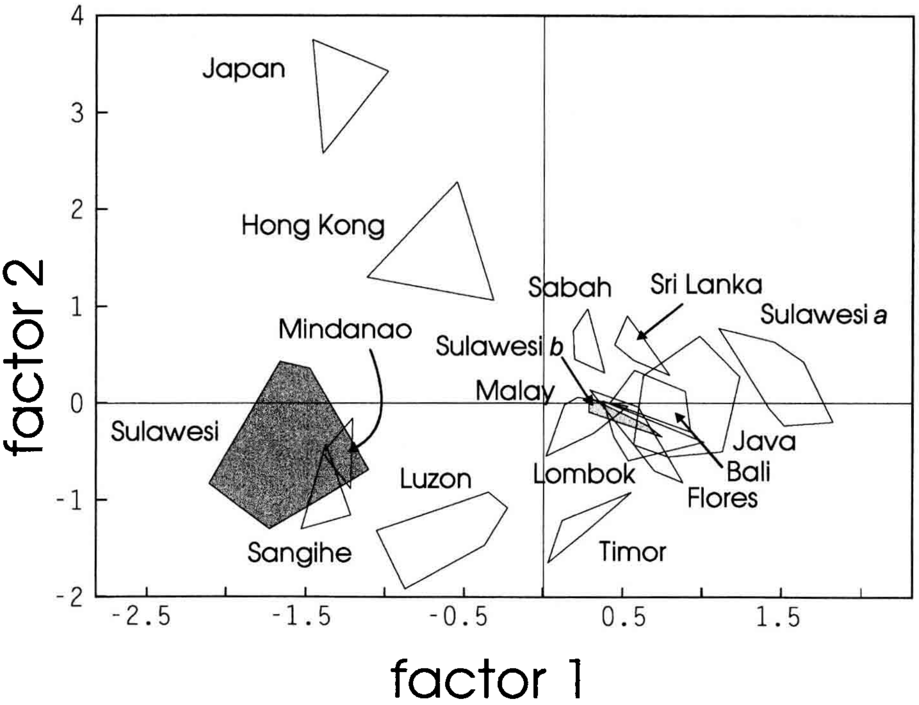

Fig 3. Principal components analysis of entire size-free data set. Factor 1 accounts for 48% of total variation, factor 2 accounts for 14%. Dark polygonal shape is *A nigrocincta* (sensu Hadisoesilo and Otis, 1996), pale grey shape is Sulawesi *b* from central Sulawesi.

1.0), while the other three Luzon samples were identified as being more similar to *A cerana* (probability 1.0). However, when any three of the seven Luzon samples were placed into their own a priori group, all of the others were classified with them (probability 1.0), indicating that they are more like each other than either *A cerana* or *A nigrocincta* sensu strictu. The Mindanao and Sangihe samples were always classified with *A nigrocincta* samples from Sulawesi, not into the Luzon a priori group, indicating that they are more similar to Sulawesi *A nigrocincta* than to the Luzon samples.

For further analyses, the data set was split into the two groups representing *A cerana* and *A nigrocincta*. The morphologically ambiguous samples from Luzon were included in both analyses, to compare their phenetic relationship with the two species.

A nigrocincta set

This data set included all of the Philippine bees, as well as *A nigrocincta* from Sulawesi as described by Hadisoesilo and Otis (1996). The multidimensional scaling analysis (MDS) resolved 96% of the original variability into four dimensions (stress = 0.08) and split the samples into three clear clusters (fig 4): *A nigrocincta* from Sulawesi and Sangihe, the Mindanao bees, and the sam-

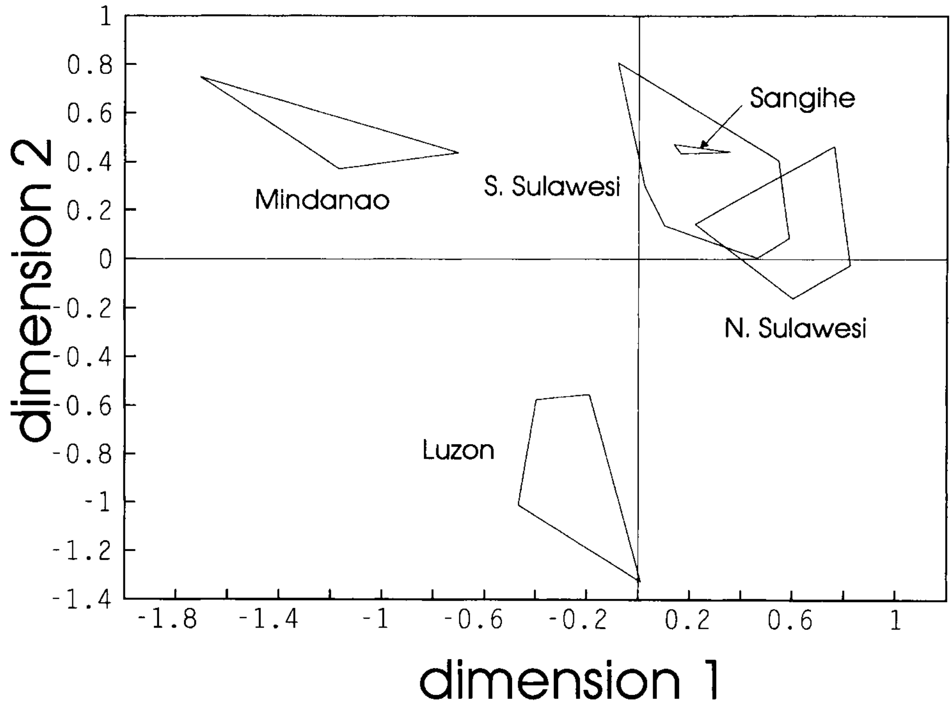

Fig 4. First two dimensions of multidimensional scaling analysis, *A nigrocincta* and Luzon bees only. Polygonal shapes enclose all specimens from named group.

ples from Luzon. These were the a priori groups included in a discriminant analysis (DA). Figure 5 shows the graph of the two factors (= discriminant functions) derived from the analysis. Ellipses are at the 75% confidence level (Cornuet, 1982; Ruttner, 1988), and define 'discrete groups'. The three groups identified by the MDS were verified as being distinct.

A cerana set

This data set included all of the *A cerana* samples from Japan, Hong Kong, Sri Lanka, Malay peninsula, Sabah/Palawan, Java, Bali, Lombok, Flores, Timor and Sulawesi (Sulawesi *a* from southern Sulawesi and Sulawesi *b* from central Sulawesi) and the samples from Luzon. The MDS was able to resolve 96% of the original variation into four dimensions, with stress 0.09. Dimension 1 (fig 6) split the samples into the three well-established subspecies *A c cerana* (Hong Kong), *A c japonica* and *A c indica* (sensu Ruttner, 1988). Dimension 2 subdivided *A c indica*, removing Sri Lanka, Timor and Sulawesi *a*, and clearly isolated the Luzon samples. Dimension 3 (not shown) separated Timor from Sri Lanka, and the Malay peninsula together with Sabah/Palawan from the rest of *A c indica*. These were the groups then entered into the DA: Sulawesi *a*, the combined samples from Java + Bali + Lombok + Flores + Sulawesi *b*, Timor, Malay + Sabah, Hong Kong,

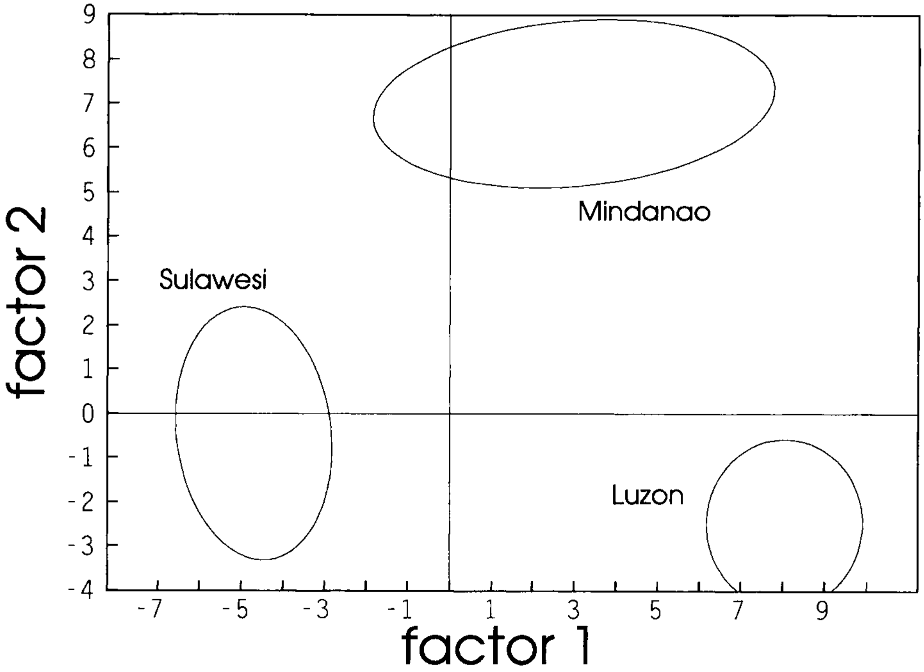

Fig 5. Only two factors (= discriminant functions) of discriminant analysis of *A nigrocincta* and Luzon bees. Ellipses at the 75% confidence level surround members of and identify morphologically distinct groups.

Japan, Sri Lanka, and Luzon. The graphical results of the DA are presented in figures 7 and 8. The first three factors separated all seven of the groups. Factor 1 (fig 7) identified *A c cerana* and *A c japonica* as being distinct. Factor 2 in combination with factor 1 separated Malay+Sabah and Timor from the Java et al group, but they were all overlapped by *A c indica* from Sri Lanka. The Luzon bees were well separated, and Sulawesi *a* was distinct but adjacent to the Java et al group. Factor 3 (fig 8) divided Sri Lanka, Malay+Sabah and Timor, and further isolated Sulawesi *a*. The Timor and Malay+Sabah samples always remained close to the Java et al group. All ellipses are at the 75% level of confidence.

DISCUSSION

This survey was able to document a high degree of morphological differentiation among the island populations of *A cerana* and *A nigrocincta* in Southeast Asia. The previously recognized subspecies A *cerana cerana* and *A c japonica* (Ruttner 1988) are upheld, and the tentative subspecies *A c philippina* is shown to be divergent from both *A cerana* and *A nigrocincta*. In view of Smith and Hagen's (1996) results, in which mitochondrial DNA sequencing placed *A c philippina* in a monophyletic branch with *A nigrocincta*, and not with any other *A cerana* groups, we suggest that *A c philippina* may, in fact, be more closely related to *A nigrocincta* than to *A cerana*.

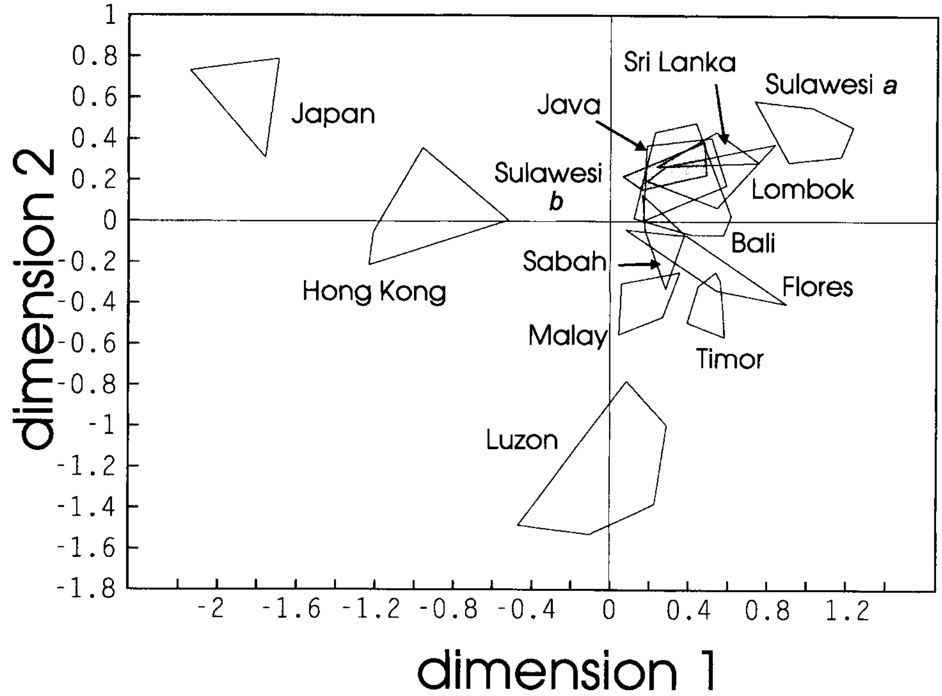

Fig 6. First two dimensions of multidimensional scaling analysis, *A cerana* and Luzon bees only. Polygonal shapes enclose all specimens from named group. Light grey shape is Sulawesi *b*.

Further analyses need to be made, particularly from other islands of the Philippines, and they should combine genetic, behavioural and morphological studies.

A c indica from Sri Lanka is shown to be distinct from the Malaysian and Indonesian samples, but as we lack geographically intermediate samples between Sri Lanka and Malaysia, we cannot at this time dispute Ruttner's (1988) identification of the Malaysian and Indonesian *A cerana* as belonging to *A c indica*. Smith and Hagen's (1996) data showed *A c indica* from Sri Lanka to have a very different mtDNA haplotype from Malaysian or Indonesian *A cerana*. The transition between Indian and Southeast Asian *A cerana* remains unstudied.

The southern Sulawesi samples (Sulawesi *a*) showed an exceptionally high level of morphological divergence (see figs 2, 3, 6 and 8) from their nearest neighbours (Malaya+Sabah and the Java et al group), even though they have mtDNA haplotypes identical to those found on Java through Timor (Smith and Hagen, 1996). Although not shown graphically, this population is distinct from others at the 95% level of confidence (95% confidence ellipse). This suggests that this is a population that has undergone significant selection and/or drift in isolation from the other populations. Conversely, a bottleneck experienced during

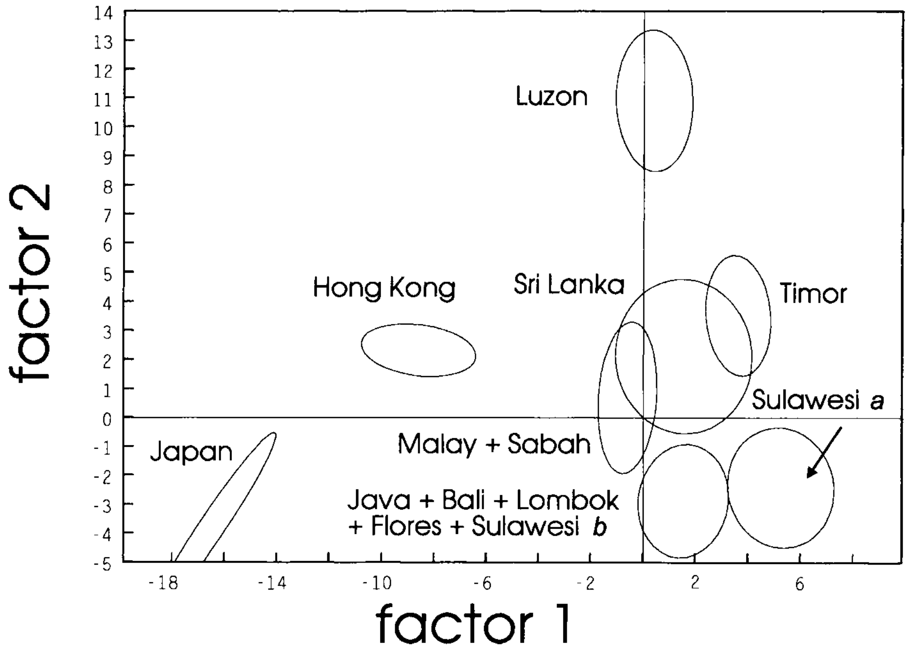

Fig 7. First two factors (= discriminant functions) of discriminant function analysis of *A cerana* and Luzon bees. Ellipses at the 75% confidence level surround members of and identify morphologically distinct groups.

past migration to Sulawesi may have accelerated phenetic diversification, as Bryant and Meffert (1996) have shown to occur in experimentally bottlenecked house flies. Such a migration may have been recent, ie, perhaps aided by humans. Unfortunately, we do not know of a way to test these alternatives with the material we have at present.

The second *A cerana* population on Sulawesi, Sulawesi *b*, seems to be indistinguishable from Javanese *A cerana*, and it may be a very recent import, as it seems to be confined to only a few sites in central Sulawesi (Hadisoesilo and Otis, 1996).

The five samples from Timor were identified as being morphologically different from other *A cerana* samples, although they are close to the samples from Java et al (figs 6–8). Like the Sulawesi *a* samples, they may represent a divergent form, clearly derived from the Javanese group (and with identical mtDNA; Smith and Hagen, 1996), but isolated for a period of time. Again due to the paucity of samples, we refrain from making subspecies judgments until more samples from a wider area of the island are analysed.

We have shown that the samples of cavity nesting honey bee from the Philippine island of Mindanao and from the small island of Sangihe between Mindanao and Sulawesi are most similar phenetically to *A nigrocincta* (fig 3), although the Mindanao population represents a morphologically different group (figs 4 and 5). Smith and Hagen (1996) place them within the *A nigrocincta* clade as well, based on molecular evidence.

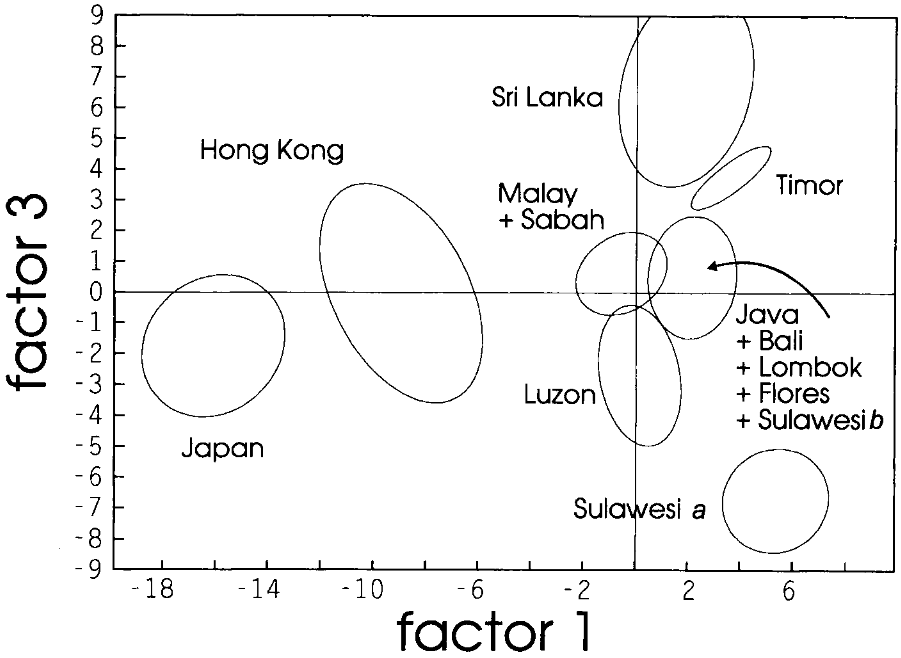

Fig 8. Factors 1 and 3 of discriminant function analysis of *A cerana* and Luzon bees. Ellipses at the 75% confidence level surround members of and identify morphologically distinct groups.

We refrain from judging the Mindanao samples to be a separate subspecies owing to inadequate samples (four) but believe that more sampling will confirm them as being significantly different from Sulawesi *A nigrocincta*. This report and that of Damus (1995) are the basis for the assertion by Otis (1996) that *A nigrocincta* inhabits Mindanao and Sangihe.

The congruence of our phenetic results with Smith and Hagen's (1996) genetic results is encouraging, and it illustrates that continued use of both genetic and morphological studies of the cavity-nesting honey bees of Southeast Asia should clear up many questions, while at the same time leading to additional, more complex ones. Further collections and studies of bees from the islands of Sulawesi, Sumatra, the islands of the Philippines and neighbouring islands are sure to result in exciting discoveries concerning the variability and evolutionary history of the honey bees of this region. Such studies will hopefully clarify the species status (*A cerana*, *A nigrocincta* or *A philippina*) of the bees from Luzon.

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Résumé — Analyse morphologique des populations d'*Apis cerana* F et d'*Apis nigrocincta* Smith d'Asie du Sud-Est. Depuis l'article de Ruttner et al (1978) détaillant les méthodes et les caractères utiles, la classification des abeilles mellifères repose sur les techniques morphométriques. Toutes les sous-espèces reconnues d'*A mellifera* et d'*A cerana* ont été décrites à l'aide des techniques de morphométrie. Seules la Malaisie, l'Indonésie et les Philippines n'ont pas été étudiées de façon appropriée. Il est encore possible d'y trouver des sous-espèces et même des espèces non décrites à ce jour (Ruttner, 1988). Notre étude donne un aperçu de la variabilité morphométrique qui existe en Asie du sud est.

Nous avons utilisé une combinaison des caractères de l'aile antérieure et de la patte de Ruttner (1988) et des caractères de l'aile postérieure de Mattu et Verma (1984), mais la principale différence avec les études précédentes est d'avoir exclu l'influence de la taille sur les variables en utilisant la technique de Burnaby, telle qu'elle a été décrite par Rohlf et Bookstein (1987). Cela nous a permis d'analyser les populations d'abeilles en fonction de leur forme et non de leur taille.

De nombreux échantillons ont été utilisés à la fois par nous et par Smith et Hagen pour leur étude (1996) portant sur l'analyse génétique de la variation de l'ADN mitochondrial, si bien que nos résultats peuvent être considérés comme le pendant morphométrique de leur étude génétique. Cette dernière a montré que les abeilles connues couramment sous le nom d'*A c philippina* (Ruttner, 1988) sont sur la même branche monophylétique qu'*A nigrocincta* de Sulawesi et que les abeilles de Mindanao et de Sangihe (Smith et Hagen, 1996). Nos analyses discriminantes ont attribué tous les échantillons de Mindanao et de Sangihe, ainsi que quatre échantillons de Luzon, directement à *A nigrocincta*, les trois autres échantillons de Luzon étant attribués à *A cerana* (probabilité de 1,0 pour tous les échantillons). Lorsqu'on a formé a priori un groupe à partir de trois échantillons de Luzon quels qu'ils soient, tous les autres échantillons ont été attribués à ce groupe, alors que les échantillons de Mindanao et de Sangihe restaient avec *A nigrocincta* (probabilité 1,0). Ce résultat, ainsi que ceux de Smith et Hagen (1996), suggèrent que les échantillons de Mindanao et Sangihe sont phénétiquement différents d'*A nigrocincta* (figs 4 et 5) et d'*A cerana* (figs 6 et 8) et représentent peut-être une sous-espèce distincte qui pourrait être, non pas *A c philippina* comme l'avait suggéré Ruttner (1988), mais *A n philippina* ou une espèce distincte propre. D'autres études sont nécessaires.

Les sous-espèces bien connues *A c cerana* (de Hong Kong) et *A c japonica* (du Japon) ont été séparées dans toutes les analyses et sur tous les graphiques (figs 2, 3, 6–8). *A c indica* est composée d'au moins trois groupes : l'un comporte les populations du Sri Lanka, de Malaisie et des îles indonésiennes de Java, Bali, Lombok, Flores, Timor et une petite population dans le centre de Sulawesi (Sulawesi *b*), le second n'a été trouvé jusqu'à présent que dans le sud de Sulawesi (Sulawesi *a*) et le troisième est celui de Timor (figs 2, 3, 6–8). Le groupe Sulawesi *a* a subi une dérive phénétique qui l'a éloigné du reste d' *A c indica*. Cela indique soit qu'il a été isolé pendant une longue période, soit qu'il a subi un goulot d'étranglement (« bottleneck ») important, peut-être dans les temps récents, peut-être aussi sous l'influence de l'Homme.

A nigrocincta peut être divisé en deux groupes : l'un de Mindanao, l'autre de Sulawesi et de Sangihe (figs 4 et 5). Nous

n'avions pas suffisamment d'échantillons de Mindanao pour être certains de couvrir correctement la variation qui existe sur l'île; aussi nous nous abstenons de considérer ce groupe comme une sous-espèce.

Il est certain que d'autres récoltes d'abeilles et d'autres études faites sur les îles de Sulawesi, de Sumatra, des Philippines et des îles voisines fourniront des découvertes passionnantes concernant la variabilité et l'histoire évolutive des abeilles mellifères de cette région. De telles études devraient clarifier, nous l'espérons, le statut d'espèce des abeilles de Luzon (*A cerana, A nigrocincta* ou *A philippina*).

***Apis cerana* / *Apis nigrocincta* / morphométrie / Asie du sud est / abeilles mellifères**

Zusammenfassung — Eine morphometrische Analyse von südostasiatischen Populationen von *Apis cerana* F und *Apis nigrocincta* Smith. Morphometrische Techniken waren seit der detaillierten Darlegung der Methoden und verwendbaren Merkmale durch Ruttner et al (1978) die Hauptstütze der Klassifierung der Honigbienen. Alle anerkannten Rassen von *Apis mellifera* und *Apis cerana* wurden mit morphometrischen Techniken beschrieben. Die Gebiete von Malaysia, Indonesien und den Philippinen wurden bislang nicht angemessen untersucht. Es ist immer noch nicht ausgeschlossen, daß bisher nicht beschriebene Rassen oder sogar Arten in diesem Bereich gefunden werden (Ruttner 1988). Unsere Studie soll einen Einblick über die morphometrischen Variabilität in Südostasien geben. Wir kombinierten eine Reihe von Ruttner's (1988) Merkmalen an Vorderflügeln und Beinen mit den Hinterflügelmerkmalen von Mattu und Verma (1984). Ein Hauptunterschied unserer Methode ist der Ausschluß des Einflusses von Größe auf die Merkmale mit Burnaby's Technik, entsprechend der Beschreibung in Rohlf und Bookstein (1987). Das ermöglichte uns, die Bienenpopulationen nach ihrer Form und nicht ihrer Größe zu analysieren. Neben unseren morphometrischen Studien wurde eine genetische Analyse der Variation mitochondrialer DNA von Smith und Hagen (1996) durchgeführt, die weitgehend dieselben Proben benutzten, sodaß unsere Ergebnisse als morphologische Gegenseite der genetischen Untersuchung angesehen werden können. Nach den genetischen Befunden gehören die bisher als *A c philippina* (Ruttner, 1988) beschriebenen Bienen zu dem gleichen monophyletischen Zweig wie *Apis nigrocincta* aus Sulawesi und die Bienen von Mindanao und Sangihe (Smith und Hagen, 1996). In unserer Diskriminanzanalyse wurden alle Proben aus Mindanao und Sangihe, sowie vier der Proben aus Luzon der Art *A nigrocincta* aus Sulawesi, die anderen drei Proben aus Luzon wurden *A cerana* zugeordnet (alle mit einer Wahrscheinlichkeit von 1.0). Wenn „a priori" Gruppen aus drei der Luzon-Proben gebildet wurden, wurden die restlichen Proben aus Luzon dieser Gruppe zugeordnet, die Proben aus Mindanao und Sangihe blieben aber weiterhin bei *A nigrocincta* (Wahrscheinlichkeit 1.0). Die Proben aus Luzon dagegen sind phenetisch sowohl von *A nigrocincta* als auch von *A cerana* (Abb 6–8) verschieden und stellen vielleicht eine eigene Unterart dar, die möglicherweise nicht die von Ruttner (1988) vorgeschlagenen *A c philippina* ist, sondern eher *A n philippina* oder sogar einer anderen Art entspricht. Dazu sind aber weitere Studien erforderlich. Die gut bekannten Unterarten *A c cerana* (aus Hong Kong) und *A c japonica* (aus Japan) waren in allen Analysen und Graphen gut getrennt (Abb 2, 3, 6–8). Für *A c indica* konnte gezeigt werden, daß sie sich aus drei Gruppen zusammensetzt: eine wird von Sri Lanka, Malaysia, den indonesichen Inseln Java, Bali, Lombok, Flores und Timor und einer kleinen Population im zentralen Sulawesi (Sulawesi *b*) gebildet, eine 2. Gruppe wurde bislang nur im südlichen Sulawesi gefunden (Sula-

wesi *a*) und die 3. Gruppe auf Timor (Abb 2, 3, 6–8). Die Gruppe Sulawesi a zeigte eine phenetische Merkmalsverschiebung von den restlichen *A c indica,* was einen Hinweis entweder auf eine lange Isolation gibt oder auf eine Population, die einen Engpaß (bottle neck) durchlaufen hat, möglicherweise vor kürzerer Zeit und möglicherweise durch menschlichen Einfluß. Bei *A nigrocincta* können zwei Gruppen unterschieden werden: eine von Mindanao und eine von Sulawesi und Sangihe (Abb 4 und 5). Wir hatten nicht genügend Proben von Mindanao um sicher zu sein, daß die Variation auf der Insel ausreichend erfaßt wurde und verzichteten daher darauf, diese Gruppe als eigene Rasse anzusehen. Weitere Probensammlungen von den Inseln Sulawesi, Sumatra, den philippinischen und den benachbarten Inseln werden mit Sicherheit zu weiteren interessanten Entdeckungen über die Variabilität und Evolutionsgeschichte der Honigbienen in dieser Region führen. Solche Studien werden dann hoffentlich auch die Artzugehörigkeit der Bienen von Luzon (*A cerana, A nigrocincta* oder *A philippina)* klären.

***Apis cerana / Apis nigrocincta* / Morphometrie / Südostasien / Honigbienen**

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APPENDIX

Samples were collected from the following sites: *Philippines: Luzon,* Laguna Province, College (2), Quezon Province, Quezon City (3), Zambales Province, Santa Cruz (2); *Palawan,* Babuyan (1); *Mindanao,* Laguna del Norte, Kauswagan (4); *Indonesia: Sangihe,* 5 km E of Tahuna (3); *Sulawesi North,* Teeb (near Amurang) (4), Sonder (1); *Sulawesi Central,* Kamarora (70 km SE Palu) (5), Bobo (1); *Sulawesi South,* Tabo Tabo (60 km N Ujung Pandang) (1), Jeneponto (5), Manipi (4), Bontobulaeng (4); *Java West,* Parungpanjang (5), Cianjur (3); *Java East,* Tretes (5); *Bali,* Tunjungan (2 km N Ubud) (6); *Lombok,* Senggigi (1), Gunungsari (4 km W of Senggigi) (6); *Flores*, Ende and surrounds (4); *Timor*, 3 km SW Kupang (5); *Malaysia: Malay Peninsula*, Selangor, Universiti Pertanian Malaysia (5), *Sabah*, Tenom (5); *China: Hong Kong* (4); *Guangdong*, Guangzhou (1) — in the body of the paper this sample is referred to as one from Hong Kong, for simplicity and because of its geographic proximity to and phenetic similarity with those samples; *Japan: Honshu*, Machida Shi (near Tokyo) (4); *Sri Lanka:* Colombo (4). *Total:* 98, all workers.