

Diversity in the Genus *Apis*

Edited by

Deborah Roan Smith



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To the memory of my mother

*Ruth Evangeline Dix Smith,
11 November 1921 - 31 January 1989*



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Preface

Honey bees constitute a single genus in the family Apidae. One species, *Apis mellifera*, is native to Europe, Africa and the Middle East; the rest, a handful of species, are found in Asia. All honey bees are similar in morphology, social biology, nest architecture, foraging behavior, and the use by foragers of a complex "dance" to signal direction and distance to food sources. *Apis mellifera* is one of the best-studied insects in the world, though many basic questions about the biology of this species remain unanswered. However, the similarities among honey bee species have, to a certain extent, blinded us to the tremendous diversity of behavior and ecology found among the Asian species. Even the number of *Apis* species is not known with any certainty.

A major question in social biology, the single or multiple origins of highly eusocial behavior in the Apidae, is still hotly debated. Its resolution hinges on deciphering the phylogenetic relationships of the four apid subgroups (orchid bees, bumble bees, stingless bees and honey bees). This problem is addressed by three studies in this collection, using morphological data, nuclear DNA characters and mitochondrial DNA characters. The fact that no consensus is reached reflects the difficulty of the problem and shows the need for additional research.

This volume grew out of an informal conference, "Diversity in the Genus *Apis*," which took place in 1989 at the annual meeting of the Entomological Society of America. The conference was inspired largely by the work of Dr. Friedrich Ruttner, whose research on *Apis mellifera* spans many decades. In *The Biogeography and Taxonomy of Honey Bees* (1988) Ruttner reviewed and summarized published research on the comparative ecology, behavior, morphology and biogeography of *Apis mellifera* and the Asian honey bee species. The contrast between the voluminous literature on the western honey bee, *Apis mellifera*, and the relatively scanty data on the Asian honey bee species highlighted the need for further research on Asian *Apis*.

Conference participants were Fred C. Dyer (diversity in dance language), Thomas Seeley (comparative energetics in Asian *Apis*), Gudrun Koeniger (diversity in *Apis* mating systems), Jean-Marie Cornuet (genetic diversity in *Apis mellifera*), Gard W. Otis (isozyme variability in the genus *Apis*), Deborah Smith (mitochondrial DNA diversity in *Apis*) and Walter

S. Sheppard (ribosomal RNA diversity in *Apis*). Here, I have collected together (in expanded form) the papers presented by most of the original conference participants as well as additional chapters by other authors. These additional chapters include a discussion of the phylogenetic relationships among *Apis* species by Byron Alexander; a summary of species diversity in *Apis* by Gard Otis, which includes information on the biology of two newly recognized species, *A. andreniformis* and *A. koschevnikovi*; two studies of phylogenetic relationships within the Apidae by Sydney Cameron and Michael Prentice; and a synthesis of recent studies on honey bee energetics by Fred Dyer.

Together, these chapters present a well-rounded picture of current research on honey bee biology. These studies are for the most part very recent; in fact many are still in progress. They are meant to serve as introductions to various aspects of honey bee biology and as guides for future research, especially on Asian honey bees. In aid of this, each author has presented a discussion of the rationale for his or her study and the techniques and methodologies involved. I hope that this collection of chapters will be the starting point for many new research projects on the systematics, biogeography and comparative biology of the Asian honey bee.

Deborah Roan Smith

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A Cladistic Analysis of the Genus *Apis*

Byron Alexander

Introduction: Historical Review of Ideas About Honey Bee Phylogeny

The literature on honey bees is so vast and incorporates contributions from such a wide range of different disciplines that few, if any, working scientists even pretend to be familiar with all of it (the author of this chapter certainly does not). Consequently, a given publication's pathways of influence are apt to be very strange and convoluted. As a case in point, consider a paper by A. Gerstäcker with the formidable nineteenth-century German title, "Über die geographische Verbreitung und die Abänderungen der Honigbiene nebst Bemerkungen über die ausländischen Honigbienen der alten Welt." This paper was apparently originally presented as a plenary address at a meeting of German beekeepers in Potsdam in 1862. It was printed in the form of a Festschrift, or commemorative volume, that also served as an identification badge indicating who had registered to attend the conference (Buttel-Reepen, 1906). Therefore, the number of copies originally printed was presumably only 524 -- the number of documented participants at the meeting -- and most of these copies were probably soon discarded (or perhaps converted to kindling for smokers to calm aggressive German honey bees). Nevertheless, this paper managed to form the groundwork for the prevailing orthodoxy concerning honey bee phylogeny for at least half a century. Gerstäcker presented a careful and comprehensive survey of variation in a number of morphological traits

across the entire geographic range of honey bees. He summarized his findings by recognizing only four species in the genus *Apis*. For the record, the names he used were *mellifica*, *dorsata*, *indica*, and *florea*; a detailed discussion of nomenclature is beyond the scope of this paper and can be found elsewhere (Maa, 1953; Ruttner, 1988)). Thus, the idea that the genus *Apis* consists of a small number of widespread species exhibiting considerable geographic variation considerably predates Mayr's (1942) formulation of the biological species concept. Gerstacker also proposed splitting *Apis* into two informal groups, with his "First Group" comprised solely of *dorsata*, and his "Second Group" holding the other three species. He convincingly defended the merits of his system relative to a simpler one first proposed by Latreille in 1804 and adopted by Lepeletier in 1836, which grouped species solely on the basis of the color of the scutellum. An English version of Gerstacker's work was published in 1863 in the *Annals and Magazine of Natural History*. Thus, Frederick Smith, a well-known English taxonomist at the British Museum (Natural History), was familiar with Gerstacker's work when he published his own revision of honey bees in 1865. However, Gerstacker's work was virtually unknown in Germany until it was resurrected in 1906 by von Buttel-Reepen, an influential German systematist and evolutionary biologist who was particularly interested in the evolution of social behavior in bees. An attentive official at the Berlin Zoological Museum had managed to acquire an original copy of Gerstacker's 1862 Festschrift paper and showed it to von Buttel-Reepen, who was sufficiently impressed with it that he arranged to have it reprinted in its entirety as part of a comprehensive paper he published in 1906 on the systematics, biology, and biogeography of honey bees. As far as I have been able to determine (always keeping in mind the possibility of other undiscovered equivalents of Gerstacker's papers), this 1906 publication by von Buttel-Reepen was the first serious attempt to discuss the evolution of the genus *Apis*, drawing on evidence from paleontology, biogeography, and comparative behavioral studies. It even included a phylogenetic diagram for all social Apidae (honey bees, stingless bees, and bumble bees) that not only indicated postulated ancestor-descendant relationships but also specified the geological age and current geographical distribution of each taxon. By our present standards, the justification for this elaborate phylogenetic scenario is extremely vague, so that it may seem like little more than grandiose story-telling. However, if considered in the context of the time when it was written, it represents an innovative and comprehensive attempt to provide a coherent conceptual framework for explaining observed patterns of variation among different species of honey bees, and it was certainly influential with other honey bee

researchers. Furthermore, it was not entirely without logical or empirical support. To a large extent, von Buttel-Reepen based his proposed phylogeny of *Apis* on ideas about the most likely sequence of changes in the progressive development of social behavior. He considered the European honey bee to have the most advanced form of society, and *Apis dorsata* to be the least socially advanced. Specific features of *dorsata* behavior that he considered to be primitive included the rearing of workers and drones in cells of the same size, and a tendency to exhibit migratory behavior. The observation that meliponines also rear workers and drones in the same types of cells was cited as evidence that this is a primitive behavior in *dorsata*, since he considered the societies of meliponines to be less advanced than, and ancestral to, those of *Apis*.

Von Buttel-Reepen's ideas about honey bee phylogeny were generally either accepted or ignored, but not contested, for the next fifty years. When Maa published his comprehensive and extremely thorough taxonomic revision of honey bees in 1953, he had relatively little to say about phylogeny and did not challenge any of von Buttel-Reepen's major conclusions. It was Maa's opinion (1953, pp. 633-634) that "The genus *Megapis* [= *Apis dorsata*, or the *dorsata* species group] beyond doubt includes the most primitive forms. This assumption is fully supported both by morphological and biological facts. The relative positions of *Apis* [= *A. mellifera*, *A. cerana*, and *A. koschevnikovi*] and *Micrapis* [= *Apis florea* and *A. andreniformis*], however, are open to controversy." In his Table 6 (Maa, 1953, pp. 629-630), he presented a list of morphological characters "believed to be of phylogenetic significance," in which he contrasted "generalized" and "specialized" extremes of characters, although he presented no discussion at all of how he determined which extreme was generalized and which was specialized for each character.

An alternative proposal for the phylogeny of *Apis*, and the one most widely accepted today, grew out of Martin Lindauer's (1956) comparative study of the dance language in three species of *Apis* in Sri Lanka (formerly Ceylon). The explicit goal of this study, which is now regarded as a classic work in comparative ethology, was to understand how the dance language of *Apis mellifera* could have evolved. Lindauer was the first to show that a dance language containing information about the direction and distance to resources occurs throughout the genus *Apis*. Although the basic form of the dance language was the same in all species he studied, he noted interspecific differences in details of the dance. He argued that this variation held clues to the evolution of the dance language, provided that one could identify homologous components of the language in each species and establish which condition was ancestral.

Lindauer's procedure for deciphering the history of the dance language was to analyze it from a functional perspective, under the assumption that a more direct, simpler communication system would be ancestral to a more indirect and complex system. He placed particular emphasis on the mode of communicating directional information, since the distinction between simpler and more complex signals seemed most obvious in this system. In *Apis florea*, the waggle dance is performed on a horizontal surface, and the straight run portion of the dance is pointed right in the direction foragers should fly when they leave the nest. The other honey bee species that Lindauer studied perform the dance on the vertical face of a honeycomb, and directional cues are presented with reference to the pull of gravity. Bees reading the dance must transpose these gravity-related directional cues in order to fly in the proper direction when they leave the nest. Lindauer reasoned that the method of indicating direction was clearly simpler and more direct, and thus more likely to represent the ancestral condition, when the dance was performed on a horizontal surface.

Lindauer's ideas gained wide acceptance, not only among German-speaking scientists, but also among English-speakers when presentations in English (Lindauer, 1961; von Frisch, 1967) became available. Additional evidence supporting Lindauer's evolutionary scenario was provided when the Janders conducted a comparative study of geotaxis in numerous families and genera of Malaysian bees. They found that the species of *Apis* that perform their dances on a vertical surface have a unique form of geotaxis, which they termed metageotaxis. This metageotaxis is essential for the accurate presentation of directional information in the waggle dance when it is performed on a vertical surface. Consequently, it seemed especially significant that the dwarf honey bee (the Janders were probably working with *Apis andreniformis*), which performs its dance on a horizontal surface, does not exhibit metageotaxis.

Biochemical data presented by Kreil (1973, 1975) have also been interpreted as supporting Lindauer's hypothesized phylogeny (Kreil, 1975; Ruttner, 1988). Kreil determined the complete sequence of 26 amino acids in the peptide melittin from the venom glands of four species of *Apis*. He found that the sequence was identical in *mellifera* and *cerana*, and *florea* was least like the other three species.

The first author to call attention to uncertainty about the polarity (i.e., primitive vs. derived condition) of various characters in Lindauer's scenario was N. Koeniger (1976). He pointed out that data such as Kreil's provided information about phenetic similarity, but were not informative about phylogenetic relationships in the absence of information about character polarity (Appendix 1 contains a detailed explanation of the

ambiguity of Kreil's data). Koeniger also called attention to further studies of geotaxis by Horn (1975) which raised doubts about whether the form of geotaxis in *Apis florea* was the same as that of other bees. The Janders had shown that *Apis florea*, along with all the other bees they examined except *Apis cerana*, *dorsata*, and *mellifera*, has a form of geotaxis that they called progeotaxis. Horn's work indicated that, although the form of the behavior called progeotaxis was the same in *Bombus terrestris* and *Apis florea*, the sensory receptors mediating the behavior are different. Without further study in the outgroup, it is not clear whether the geotaxis of *Apis florea* is plesiomorphic or an autapomorphy, perhaps derived from the metageotaxis of other *Apis* species.

Furthermore, Koeniger pointed out that *Apis* belongs to a monophyletic group (the family Apidae of Michener, 1974) in which cavity-nesting is probably the groundplan state. Hence, the cavity-nesting of certain honey bee species may not be a derived condition, as Lindauer's evolutionary hypothesis requires. Taken by itself, this character suggests that the species that nest in more open situations may have arisen later than the cavity-nesting honey bees. Dyer (1985, 1987) has recently published detailed analyses of the dance language in *A. florea* that show that its method of communicating directional information is not as simple or straightforward as Lindauer's more cursory studies had suggested.

Although Koeniger called attention to the need for a rigorous examination of characters in the light of the principles of phylogenetic systematics (Hennig 1950, 1966), and he discussed how a cladistic interpretation of one character would alter widely-held opinions about *Apis* phylogeny, he did not present a comprehensive cladistic analysis himself. Instead, he ended his paper with the statement (my translation) that "a clarification [of the phylogeny] will require studies involving characters of *Apis* species that can be compared with homologous characters of other groups of Apidae." To my knowledge, the only previously published study of *Apis* claiming to be a quantitative cladistic analysis is that of Sakai et al. (1986). They evaluate 23 species and subspecies (or races) from the entire natural range of the genus. Their data matrix includes morphological, behavioral, and biochemical characters, and they present both cladistic and phenetic analyses. However, their cladistic analysis is difficult to understand or evaluate, because they do not indicate how the characters were polarized or how coding decisions were made for continuously varying characters. Nevertheless, their conclusions about cladistic relationships are concordant with Lindauer's hypothesis.

In view of Koeniger's (1976) remarks about the need for additional characters whose polarities could be clearly established, it is ironic that he

must have been unaware that just such a set of characters had been discovered by Snodgrass in 1941, in a comparative morphological study of the male genitalia throughout the order Hymenoptera. In this wide-ranging survey, Snodgrass examined three species of *Apis* (*mellifera*, *cerana* (which he called *indica*), and *floreana*), identified several similarities shared by *mellifera* and *cerana*, and presented cogent arguments indicating that they were *derived* homologous similarities.

The analysis presented in this chapter is an extension of the study begun by Snodgrass and Koeniger. It is a quantitative cladistic analysis based upon a comparative study of adult morphology and, to a much smaller extent, behavior. Its objective is to determine if a cladistic analysis, using characters whose polarity can be established by outgroup comparison and which are independent of those involved in Lindauer's hypothesis of the evolution of the dance language, will support the cladistic relationships among the species of *Apis* implied by his evolutionary scenario. My data matrix (Table 1.2) does include one character that is clearly an integral part of Lindauer's evolutionary scenario. This character is the choice of nest site location (character number 20 in Tables 1.1 and 1.2). I included it in my analysis because Koeniger has made a convincing case that its polarity can be determined by outgroup comparison, and because he specifically mentioned it as supporting an alternative phylogenetic hypothesis for the species of *Apis*. In reporting my results, I have tried to explain the meaning of specialized terminology and quantitative measurements that are unlikely to be familiar to readers not versed in the methodology of quantitative cladistic analysis. However, a complete explanation and justification of parsimony methods is beyond the scope of this chapter. An introduction to the philosophical, biological, and mathematical issues involved in parsimony methods as used in phylogenetic systematics can be found in Farris (1983), Felsenstein (1983), and Sober (1983).

Materials and Methods

Before a cladistic analysis of the species of *Apis* can be undertaken, it is necessary to establish what the species are. Taxonomists working on honey bees have differed widely in the number of species they recognize. Recent treatments have ranged from Maa's (1953) upper extreme of 24 species in three genera to a much more conservative list of four, or possibly five, species in one genus (Ruttner, 1988). However, until very recently, the prevailing practice was to ignore Maa's work entirely or

dismiss it as an example of extreme splitting, and recognize four species of *Apis*, namely *florea*, *dorsata*, *cerana*, and *mellifera*, following the practice first championed by Gerstäcker. Even the most recent general summaries on honey bee biology or taxonomy (e.g. Seeley 1985, Winston 1987, Ruttner 1988) follow this practice, with some brief discussion of a few unresolved taxonomic questions concerning certain Asian populations. Serious consideration of these unresolved questions is now under way, with the result that opinions about the number of species of *Apis*, especially in Southeast Asia, are changing. The analysis presented here deals with six species or species groups: *andreniformis*, *cerana*, *florea*, *koschevnikovi*, *mellifera*, and the "*dorsata* group". Evidence for the recognition of *andreniformis* and *koschevnikovi* as valid biological species is discussed elsewhere (Wu and Kuang, 1986, 1987; Wongsiri et al., 1989 for *andreniformis*; Tingek et al., 1988; Mathew and Mathew, 1988; Rinderer et al., 1989; Ruttner et al., 1989 for *koschevnikovi*). The species status of various populations of *Apis* with obvious affinities to *dorsata* remains more controversial. The cladistic analysis presented here is not intended to resolve whether *dorsata* is one widespread species exhibiting considerable geographic variation (similar to that of *cerana* and *mellifera*), or a group of closely related species. Cladistic relationships among taxa can be resolved only if one can identify characters that exhibit more than one state in the taxa being compared. The characters that could be used to resolve relationships among the six taxa analyzed in this study did not vary within the *dorsata* group. If systematists studying *Apis* reach a consensus that there are several species in the *dorsata* group, it will be necessary to find additional characters to resolve the cladistic relationships among them. The resolution of this study will be sufficient to provide an independent test of the hypothesized phylogeny of the species studied by Lindauer, and it can provide a framework for future studies using other characters that might provide a finer level of resolution.

In selecting characters for this analysis, the principal criteria were that there be two or more discrete states for each character, and that one of these states also occur in the outgroup (i.e. species outside the genus *Apis*, but considered to be close relatives of *Apis*), so that there would be a basis for determining which of the alternate states in *Apis* represents the ancestral condition. Care was taken to examine specimens from throughout the known range of each species, to verify that the characters chosen are not restricted to local populations of a species. Although all castes have been examined, drones were found to have most of the variation that was potentially informative about phylogenetic relationships within the genus. Outgroup taxa used for character polarization were

chosen from all the major lineages in the family Apidae (*sensu* Michener, 1974), whose monophyly is well established (Winston and Michener, 1977; Sakagami and Michener, 1987), and from two basal clades within the Xylocopinae, which is the sister group of the Apidae (Sakagami and Michener, 1987). Additional details of the taxa examined in this study are documented elsewhere (Alexander, 1991). Although several cladistic analyses of the family Apidae have been published (Winston and Michener, 1977; Kimsey, 1984; Plant and Paulus, 1987; Chapters 3, 4 and 5, this volume), there is still uncertainty as to the sister group of *Apis* (C.D. Michener, personal communication). This did not present a problem with most of the characters used in this study, since the outgroup exhibited only one of the states found within *Apis*, so that polarity decisions were unequivocal (*sensu* Maddison et al., 1984). The hind wing venational character for which this was a significant consideration will be discussed below.

Dissections of male genitalia and female stings were cleared overnight at room temperature in 10% KOH. These dissections are in vials mounted with the pinned, dried specimens from which they were dissected. Specimens with dissections that were examined for this study are identified as voucher specimens in the collections of the Snow Entomological Museum and the Cornell University Insect Collections. Cladistic analyses were done with the Hennig86 computer program written by James S. Farris (Farris, 1988).

Results

The analysis used 21 characters (Table 1.1; matrix in Table 1.2) and 7 taxa, including the outgroup. The general objective of a cladistic analysis based upon parsimony methods is to arrange taxa in a pattern that minimizes the number of transitions among character states. In biological terms, parsimony can be viewed as an attempt to maximize the number of shared similarities that can be explained by homology, or inheritance from a common ancestor, while minimizing the number of similarities that must be explained by *ad hoc* hypotheses of homoplasy (= convergence and parallelism) (Farris, 1983). In mathematical terms, this amounts to finding the shortest possible pathway, or tree, connecting the taxa. Tree "length" is expressed as the number of "steps", with a step being a transition from one character state to another. Consequently, it is important to specify how characters that exhibit more than two states, such as characters 11, 12, 13, and 17 in Table 1.1, are to be treated in the algorithms used to find the

shortest tree. If a character is treated as *additive*, a transition between states 0 and 2 must proceed through state 1, so that the transition between states 0 and 2 requires two steps. If a character is treated as *non-additive*, the transition between states 0 and 2 need not pass through state 1, and any transition between any two character states requires only one step. Non-additive characters place fewer restrictions on character transformations, but they also have less power to resolve cladistic relationships (Mickey, 1982). Thus, in an analysis aimed at resolving cladistic relationships, it is generally preferable to treat multistate characters as additive whenever possible. In this analysis, only character 17, the vestiture of the male tarsi, was coded as non-additive, since there was no independent biological rationale (such as a morphocline) for arranging the three alternate states in a linear transformation series. The other multistate characters (11-13) were coded as additive, since one could logically consider them to be ordered in a linear sequence of transformations (or morphocline), with the starting point, or plesiomorphic condition, determined by noting which character state occurs in the outgroup.

A parsimony analysis of the matrix in Table 1.2 found a single most parsimonious tree (Figure 1.1), with a length of 27 steps and a consistency index of 93. The consistency index for a tree is the ratio of the shortest possible tree for a data set (its length if *all* shared similarities are homologous and *no* character states arise independently in unrelated taxa) to the observed length of the tree whose consistency index is being calculated, taking into account multiple origins of a given character state on that tree (Kluge and Farris, 1969). The higher the consistency index, the greater the agreement among different characters in supporting the same pattern of cladistic relationships. Compared to other published quantitative cladistic analyses, the consistency index of 93 found in this analysis is unusually high.

One possible reason for such a high consistency index is that over half of the characters in the data matrix are synapomorphies shared by *all* species of *Apis*. Such characters support the non-controversial hypothesis that honey bees are a monophyletic assemblage. (Although this hypothesis is not controversial, it is not valid to simply assume that *Apis* is monophyletic.) However, these characters supporting the monophyly of *Apis* provide no information at all about the question of major interest in this study, namely the phylogenetic relationships among the species *within* the genus. If one is to use the consistency index to assess how much agreement or disagreement there is among different characters in supporting the same pattern of phylogenetic relationships, characters that

Table 1.1 Characters and alternate states used in the quantitative cladistic analysis. Characters 19 and 20 refer to the behavior of workers or larvae; all other characters are features of adult morphology. Characters 0-3 apply to both sexes and all castes, 4 applies to queens, 5-7 apply to workers, and 8-18 apply to drones. All characters except 17 are coded as additive (see text). Alexander (1991) contains illustrations of Characters 0 and 5-9, which are not illustrated here or in other references cited below.

CHARACTERS AND CHARACTER STATES

0. Compound eyes hairy:
 0. no
 1. yes
1. Angle ABC of forewing:
 0. $> 45^\circ$ (Figure 1.2D)
 1. $< 45^\circ$ (Figure 1.2A)
2. Angle BDE of forewing:
 0. $> 45^\circ$ (Figure 1.2D)
 1. $< 45^\circ$ (Figure 1.2A)
3. Distal abscissa of hindwing vein M (indica vein):
 0. present (Figure 1.2C)
 1. absent (Figure 1.2B)
4. Ovariole number:
 0. 3 or 4*
 1. > 50
5. Barbed sting:
 0. absent
 1. present
6. Sting sheath:
 0. pigmented and bearing distinct setae
 1. unpigmented, with short, inconspicuous setae
7. Venter of metasomal segment 8 a conspicuous membranous bulb surrounding base of sting shaft:
 0. no
 1. yes
8. Compound eyes of males meeting at top of head
 0. no
 1. yes
9. Male proboscis:
 0. same length as in female (worker in social species)
 1. much shorter than in worker
10. Male endophallus (Figure 3.5 in Ruttner 1988):
 0. not greatly enlarged
 1. enormously enlarged

(continues)

Table 1.1, continued

CHARACTERS AND CHARACTER STATES

11. Ventral gonocoxite:
 0. present, sclerotized throughout
 1. sclerotized portion reduced to a transverse bar (Figure 1.3A, gc)
 2. membranous throughout (Figure 1.3C)
12. Dorsal gonocoxite:
 0. not conspicuously reduced (Figure 1.4C, gc)
 1. reduced, widely separated mesally, about half as long as penis valves (Figure 1.4E, gc)
 2. greatly reduced, less than half as long as penis valves (Figure 1.4D, gc)
13. Gonobase:
 0. present as a distinct ring
 1. an incomplete ring, or isolated fragments of sclerotization (Figure 1.4C, br)
 2. absent (Figure 1.4D, E)
14. Male metasomal tergum 8:
 0. with two long arms of about the same length (Figure 1.3A, C, T8)
 1. vertical arm much longer than horizontal arm (Figure 1.4A, T8; also Plate 31T of Snodgrass 1941)
15. Male metasomal sterna 7 & 8
 0. not fused mesally (Figure 1.3A-D, S7, S8)
 1. fused mesally (Figures 1.4A, B, S7, S8)
16. Thumblike process on male hind basitarsus:
 0. absent
 1. present (Figure 7.4 in Ruttner, 1988; Figures 1 & 2 in Wu and Kuang, 1987)
17. Vestiture of male tarsi:
 0. not specially modified
 1. dense pads of frond-like setae on middle and hind tarsi (Figures 8.8, 8.9 in Ruttner, 1988)
 2. dense pads of stiff bristles on inner surface of thumblike process of hind basitarsus (Figure 7.5 in Ruttner, 1988)
18. Flagellum of male antenna:
 0. "long", i.e. about as long as distance from vertex to apical margin of clypeus
 1. "short", i.e. about half as long as distance from vertex to apex of clypeus
19. Capping of drone cells:
 0. without a central pore
 1. with a central pore (Figures 9.13, 9.14 in Ruttner, 1988)
20. Nest Site
 0. within a cavity
 1. not within a cavity

*In the outgroup taxa examined for this analysis, the socially parasitic genus *Psithyrus* shows considerable intra- and interspecific variability in ovariole number (Cumber, 1949), although the number of ovarioles never approaches that found in *Apis* queens. This is presumably an autapomorphy for *Psithyrus*, associated with its socially parasitic way of life.

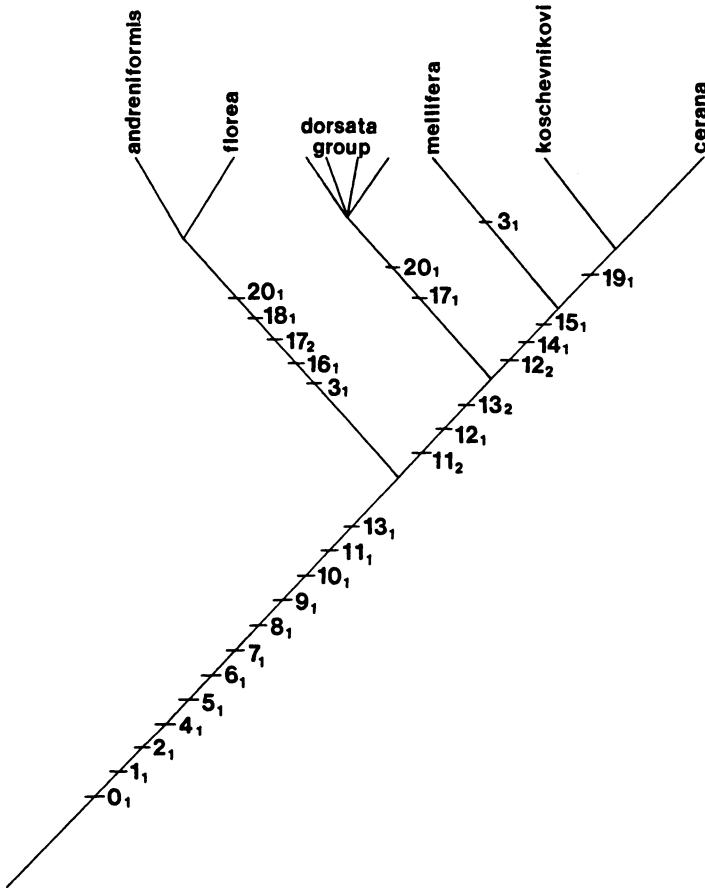


Figure 1.1 (A) The most parsimonious cladogram for the species of *Apis*. This cladogram is based upon the data matrix in Table 1.2, and explanations of characters and character states are given in Table 1.1. The pairs of numbers on the cladogram indicate inferred transformations in character states. In each pair, the large number on the left is the number of the character, as listed in Table 1.1, and the smaller subscript on the right represents the derived character state. See text for further discussion.

have the same derived state in all taxa in the group of interest, or characters in which the derived state occurs in only one taxon, will inflate the consistency index in a potentially misleading way. No matter how the taxa are grouped on a cladogram, these particular characters will never suggest an arrangement of taxa that would contradict the grouping of taxa supported by another character.

If the consistency index is recalculated with all the autapomorphies for the genus *Apis* excluded (characters 0-2 and 5-10), its value drops from 93 to 88. The only homoplastic (convergent) characters are the distal abscissa

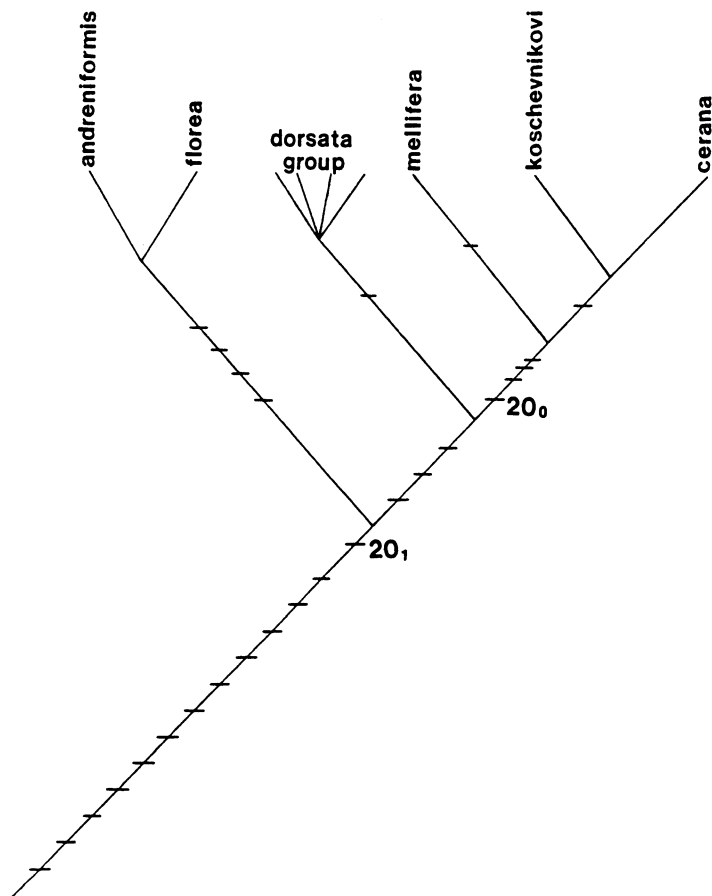


Figure 1.1 (B) An alternate transformation series for character 20 (nest location). See text for further discussion.

of vein M on the hind wing (character 3), which is hypothesized to have been independently lost in *mellifera* and the *andreniformis-floreana* lineage, and nest site location, which will be discussed below.

Discussion

The results of this analysis are unusually clear and unequivocal. This does not guarantee that the hypothesis of common ancestor relationships summarized in Figure 1.1 is true, but the available evidence strongly favors this hypothesis over any others. The hypotheses of homoplasy required by this phylogenetic hypothesis are plausible. Two equally