

DNA STUDIES REVEAL PROCESSES INVOLVED IN THE SPREAD OF NEW WORLD AFRICAN HONEYBEES

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ABSTRACT

African honeybees, imported to South America thirty-four years ago, have spread throughout most of the neotropics and have replaced the resident European bees. Two controversial views concern the nature of the neotropical African population. One view is that the African bees have spread primarily by paternal introgression into European colonies and the resulting population is mixture of all African-European hybrid genotypes. The other view is that the bees have spread primarily by maternal migration of feral African swarms and the feral population has retained, to a large extent, an African genetic integrity. Results from recent studies, using mitochondrial and nuclear DNA markers, support the latter view. Asymmetries in both maternal and paternal gene flow between feral African and managed European populations favor the African genotype. The replacement of the large extant European gene pool by a tiny introduced African gene pool could not have occurred if the African and European populations were panmictic and the African genotype were not favored by selective mechanisms. Some separation of the parental genotypes and/or selection against hybrid genotypes had to be realized. Superior fitness in a tropical environment is probably largely responsible for the African bee success, but reduced fitness of hybrids due to genetic factors may be involved also. As African bees approach temperate climatic regions where European bees are better adapted, a persisting hybrid zone may be established. With DNA markers, hybrid zone dynamics can be studied which may reveal the nature of selective processes.

RESUMEN

Las abejas africanas, importadas a America del Sur 34 años atras, se han expandido a traves de la mayor parte del Neotrópico y han reemplazado las abejas europeas. Dos controversiales puntos de vista se relacionan con la naturaleza de la población de abejas africanas en el neotrópico. El primer punto de vista sostiene que las abejas africanas se han esparcido en las colonias europeas en base a una introgresión paterna dando por resultado una población mixta de genotipos híbridos Europeo-Africanos. El segundo punto de vista es que las abejas se han esparcido primeramente por migración materna de emjambres salvajes de abejas africanas y que la población salvaje ha retenido, una integridad genetica africana. Resultados de estudios recientes, utilizando marcas de mitocondria y DNA nuclear, soportan esta teoria. Asimetrías del flujo genético materno y paterno entre poblaciones salvajes africanas y poblaciones europeas favorecen el genotipo africano. El reemplazo de un fondo genético minúsculo no pudo haber ocurrido si las poblaciones africanas y europeas fueran panmiticas y el genotipo africano no estuviera favorecido por mecanismos de selección. Puede haber ocurrido una separación de los genotipos paternos y la selección en contra de genotipos híbridos. El ambiente tropical es probablemente la razón por la cual la abeja africana se ha adaptado tan bien en este medio, pero la reducción de la adecuación de los híbridos puede deberse también a factores geneticos. Cuando las abejas africanas colonicen regiones climaticas templadas donde las abejas europeas están mejor adaptadas, un híbrido persistente puede establecerse en esta zona. Con marcas de DNA, la dinámica de híbridos en esta zona puede ser estudiada revelando la naturaleza del proceso selectivo.

The honeybee, *Apis mellifera* (L.), is represented by a number of subspecies, or races, indigenous to Europe, the Middle East and Africa (Ruttner 1988). Among the races are profound differences in ecological adaptation between temperate and tropical environments (Fletcher 1978, 1991, Winston et al. 1983, Rinderer 1988, Ruttner 1988). Because African bees are adapted to the tropics, queens of the race *A.m. scutellata* (Lepeletier) (initially believed to be *A.m. adansonii*, Latreille, Ruttner 1988) were brought to Brazil in the late 1950s with the intention of improving commercial honey production (Kerr 1967). African swarms that had escaped from apiaries established a large self-sustaining feral population. The African bee population has since expanded through most tropical regions of South, Central and North America (Michener 1975, Taylor 1977, 1985a).

The honeybees first introduced to the Americas were primarily of the west European subspecies *A.m. mellifera* (L.) and *A.m. iberica* (Goetze). These bees were notorious for their stinging tendency. The more docile east European races, *A.m. ligustica* (Spinola), *A.m. carnica* (Pollmann) and *A.m. caucasia* (Gorbachev), were imported later and became the predominant stock used for beekeeping (Pellet 1938, Oertel 1976, Shepard 1989). In temperate but not tropical regions, European bees established feral populations (Michener 1975, Taylor 1977, 1985a, 1988). In the neotropics, European bees were confined largely to apiaries. Nevertheless, they vastly outnumbered the imported African bees. Yet within thirty-four years, African bees replaced the European subspecies over most of one continent and part of another, without noticeable modification of the African behavioral characteristics (Taylor 1985a). The African bee takeover has been dramatic, with major biological and economic consequences.

The tendencies to invest honey resources into brood production, to swarm, to abscond, and, most notably, to sting readily make African bees difficult and less profitable to keep (Michener et al. 1972, Michener 1975, Winston et al. 1983). When African bees invaded New World countries, the beekeeping industries were devastated (Michener et al. 1972, Rinderer 1986a, Caron & Gray 1991). In some places, the industries have slowly rebuilt as management practices have adapted to the African bee (Gonçalves et al. 1991).

The African bee population has been spreading into Texas since October, 1990, and is expected soon to become well-established across the southern tier of the United States. The northward spread of the African bees may be halted by temperate climatic and ecological conditions. A hybrid zone may be formed between them and the persisting European population to the north (Taylor 1977, Taylor & Spivak 1984). Within the United States, the highest concentration of feral African bees will be in Florida. With a large beekeeping industry, many crops dependent on honeybee pollination, and an economy dependent on tourism, Florida will be the state most severely affected.

DIFFERENT VIEWS ABOUT THE AFRICAN HONEYBEE TAKEOVER IN THE NEOTROPICS

Superior adaptability to tropical ecological conditions is probably largely responsible for the amazing success of the neotropical African bees (Fletcher 1978, 1991, Winston et al. 1983, Rinderer 1988, Ruttner 1988). Despite the phenomenal nature of the African bee takeover, little is known about how it has happened. To understand the processes responsible, the extent to which African and European bees have interacted must be established. Currently, there are two major views concerning the composition and spread of the neotropical African population.

One school of thought maintains that, because African and European bees are of the same species, extensive hybridization has occurred. The neotropical African population has been described as a "hybrid swarm", meaning that it represents the entire range of hybrid genotypes (Rinderer 1986b). African drones mating with extant European

queens has been assumed to be the primary driving force responsible for African bee spread (Rinderer et al. 1985, Rinderer 1986b). As the extant bees become Africanized, they serve to perpetuate further the spread of African genes. The term "Africanized bees", commonly used to describe all African-derived bees in the Americas, not only implies that the bees are hybrids but also that the bees were originally European.

The other school of thought holds that African bee spread has been through maternal migration, that is, through queens accompanying swarms (Taylor 1977). The retention of African phenotypic characteristics suggests that neotropical African bees have largely retained an African genetic integrity (Michener 1975, Taylor 1985a, 1988, Fletcher 1991). Although mating occurs between African and European bees, strong selective processes in the tropics would favor African bees to such an extent that the presence of European-African hybrids would only be transitory. Hybrids would exist primarily in temperate-subtropical boundary regions, where both types of bees would be equally adapted (Taylor & Spivak 1984, Taylor 1985a, Lobo et al. 1989, Sheppard et al. 1991). As will be discussed, these latter views have been supported by recent studies.

DISTINGUISHING AFRICAN AND EUROPEAN BEES AND THEIR HYBRIDS

Studies of hybridization have been impeded due to a lack of genetic markers that distinguish African and European bees (reviewed by Daly 1988, 1991). Identification of African bees has been primarily through discriminant analyses of subtle morphological features. The genetic basis of the morphological characteristics has not been defined. The characteristics are certainly a consequence of complex temporal and spatial interactions among multiple gene products, and they are subject to pleiotropic and environmental effects. The characteristics exhibit a range of variability shared by both African and European bees. Different distributions within the range allow assignment of probabilities that bees belong to one group or the other. Intermediate morphometric scores in neotropical bees have been used as evidence for hybridization (Bucó et al. 1987, Sheppard et al. 1991) but intermediate probabilities mean that a colony of bees cannot be identified confidently. Such scores could be a result of hybridization but do not actually demonstrate it.

Hybridization is demonstrated more appropriately and accurately by the exchange of genetic markers. Allozymes, commonly used in population genetic studies, are few in number and lack specificity in honeybees, as in hymenopteran insects in general (reviewed by Daly 1988, 1991). Only allele frequency differences at a few enzyme loci distinguish African and European populations. Intensive parental analysis of individual colonies increases the effectiveness of allozymes in following introgression (Taylor et al. 1991). Recently, understanding of the processes of African bee spread has been greatly enhanced through the use of DNA markers seen as restriction fragment length polymorphisms (RFLPs) (reviewed by Hall 1991). DNA differences, as a whole, are not as subject to the selective forces that limit protein differences. Potentially, many DNA polymorphisms can be discovered.

FINDINGS WITH HONEYBEE DNA

Two significant studies, conducted independently, found that virtually all feral swarms caught in regions occupied by African bees carried African mitochondrial DNA (mtDNA) (Hall & Muralidharan 1989, Smith et al. 1989). These findings were confirmed by additional research employing a rapid method to identify the mtDNA type (Hall & Smith 1991). In regions where the African population was well established, such as in Venezuela, both managed and feral colonies had African mtDNA. Because mtDNA is maternally inherited, these studies revealed that neotropical African bees have spread

as unbroken African maternal lineages extending back to the original queens brought from Africa to Brazil. The speed and distance involved in the expansion of the African bee population and the notorious swarming tendency of African bees logically implicated maternal migration (Taylor 1977).

Analysis of both nuclear DNA and mtDNA has allowed the contributions of maternal and paternal gene flow to be distinguished. European colonies in southern Mexico, established before African bees entered the region and sampled fifteen months afterwards, carried European mtDNA but carried low levels of European nuclear DNA markers (Hall 1990). A loss of European alleles would result from daughters of the European queens mating with African drones. Such bees can be accurately called "Africanized."

African paternal introgression into apiaries has been well documented. The increase in defensive behavior in the managed colonies, shortly after African bees move into an area, is impressive. However, Africanized apiaries are not a significant factor in the spread of African genes and in the establishment of the African population. The absence of European mtDNA in the feral population shows that swarms from European colonies have not contributed to the migrating feral population, even if they have become extremely Africanized after repeated backcrossing. Apparently, European maternal lineages in tropical apiaries eventually disappear. Unless actively maintained, they are probably lost through attrition. Thus, Africanization of European matriline is a dead end process. African mtDNA in apiaries would result from beekeepers replacing dead colonies with feral swarms and, possibly, from African queen takeovers of European colonies (Michener 1975, Taylor 1985ab).

In feral African swarms from the same region of Mexico, the European nuclear DNA markers were present at low levels (Hall 1990). Thus, some paternal "Europeanization" apparently occurs at the edge of the expanding African population, where populations of managed European bees are first encountered. In colonies from Venezuela, the markers were almost completely absent (Hall 1990). Hybrids carrying European markers appear to be lost over time as the African population becomes established. However, the lower level of European markers in Venezuelan colonies may indicate that the bees had been more isolated and had encountered fewer European colonies. Samples at the same locations must be collected over time to confirm a temporal loss of European markers. African swarms collected in northern Mexico exhibited a dramatic change in allozyme frequencies in less than a year, reflecting an initial European contribution that was subsequently lost (Taylor et al. 1991). A loss of European markers would be consistent also with observations in Panama of a temporal change in feral bees towards a more African morphology (Boreham & Roubik 1987).

In the nuclear DNA studies described above, markers were used that distinguished African from east European bees but not from west European bees (Hall 1990). Neotropical African-European hybridization was investigated recently with a DNA allele present in almost all west European bees but absent in east European bees. This allele was discovered at a low level in Old World African bees but at a much higher, relatively constant, level among several New World African populations (McMichael & Hall, manuscript in preparation). It appears that, as a consequence of African-European hybridization, a west, but not an east, European contribution has persisted in the neotropical African gene pool.

The mtDNA and nuclear DNA findings have demonstrated major asymmetries in both paternal and maternal gene flow between the neotropical feral African population and the European apiaries. Despite the overwhelming numbers of European bees present in managed colonies at the time of African bee introduction, the genotype of the neotropical African descendents has remained largely African.

MECHANISMS THAT MAY PRESERVE THE
AFRICAN GENOTYPE IN THE NEOTROPICAL FERAL POPULATION

The view that the neotropical bee population is a panmictic European-African "hybrid swarm" (Rinderer 1986b) is difficult to reconcile with the persistence of African traits (Michener 1975, Taylor 1985a, Fletcher 1991), the maintenance of African allozyme frequencies (Taylor et al. 1991), and the paucity of European DNA markers (Hall 1990). The number of African alleles contributed by the introduced bees would have been a small fraction of the number contributed by the extant European colonies (Kerr 1967). In a freely interbreeding African-European population, with no hybrid limitations, alleles would be reproduced in proportion to the parental contributions (in Hardy Weinberg equilibrium). To allow the superior fitness of the African bees to be manifested and not be obliterated by hybridization, an African genetic integrity had to be largely retained. Either isolation had to prevent mixing of the genotypes or, once hybrids were formed, the proportion of European alleles had to be reduced. The alleles had to be diluted by inward migration of more pure African bees and/or eliminated through selection.

Significant isolation has probably been realized through allopatric separation. Feral African bee populations likely became established in many neotropical regions distantly separated from managed colonies (Taylor 1985a). With a higher reproductive rate, the numbers of the feral African bees would have increased in proportion to the European bees in apiaries. If European alleles had not entered the gene pool through prior hybridization, the isolated populations could have served as sources of pure African bees.

As swarms of African bees moved from the isolated regions into areas with European bees, they would have been subject to genetic dilution through hybridization. If there had been no selection against hybrids, gene frequencies would have reflected the effective size of the migrating African front and the cumulative numbers of European bees encountered enroute. As the African population spread through regions with managed European colonies, African allele frequencies would have decreased over distance as a gradient, that is, as a cline. At the outer edge of the African bee distribution, where African gene frequencies would have been very low, the rate of expansion would have been limited by short mating flight distances and by short swarm flight distances more typical of European bees (Taylor 1977). Despite a slow rate of spread, the African bee takeover could have occurred through this process. Continuous migration of African bees from the more isolated areas into areas of hybridization would have diluted the European gene pool and would have advanced the cline.

In contrast to a cline, the edge of the African population persists as a rapidly expanding front without significant mitigation of African traits (Taylor 1977, 1985a). In African swarms near the migrating front, European DNA alleles are found, but they represent a minor proportion (Hall 1990). For the expanding front to be maintained, selective processes must exist to eliminate European alleles. Long distance dispersal itself may be a primary selective factor. Hybrids may have a reduced reproductive capability and a lower propensity to swarm and to migrate long distances. Hybrids may tend to fall behind the front and could persist as a more stationary population. However, the levels of European DNA alleles are lower behind the front (Hall 1990). Lower levels could result from continued migration of African bees from isolated regions or from selective processes that continue to purge European alleles. Since European bees are considerably less fit than African bees in the tropics, it is logical that hybrids may be less fit due to a number of ecological factors (Taylor 1985a). Also, possible genetic incompatibilities have not been ruled out which would tend to be expressed in late generation and backcross hybrids.

In North America, most managed colonies carry east European mtDNA, whereas some feral populations carry a large proportion of west European mtDNA (Hall & Smith

1991). Thus, in temperate feral populations, remnants of the early European introductions seem to persist. By encountering European bees primarily as a managed population, the spreading African bees would have confronted a large proportion of the east European type. Nevertheless, as in the North American feral population, west European races may have represented a sizable proportion of the South and Central American populations prior to African bee takeover, particularly from rustic apiary colonies and from feral colonies that may have existed. As described above, an allele, that is predominantly a west European marker, is present at a significant level in neotropical African populations (McMichael & Hall, manuscript in preparation). This allele may have come from early hybridization with *A.m. mellifera* or *A.m. iberica* before the feral African population became established. The constant level of the allele among neotropical populations suggests that, after entering the African gene pool, this allele has been replicated in the same proportion and has not been selected against (that is, a neutral marker in Hardy Weinberg equilibrium). An intriguing possibility is that the higher retention of this marker reflects a closer relationship, hence greater genetic compatibility, between African and west European bees than between African and east European bees. The west European allele has not accumulated further as the African population has expanded. Thus, even if there were greater compatibility, African-west European hybrid formation and/or survival still appear to be limited in the tropics.

After intensive backcrossing with feral African drones, European matrilineages should have become more adapted to tropical conditions, and swarms from such Africanized colonies should have contributed significantly to the feral population. However, European mtDNA is absent in the feral population (Hall & Muralidharan 1989, Smith et al. 1989, Hall & Smith 1991). A persisting disadvantage from a European maternal component could be responsible. Perhaps European mtDNA limits the intense metabolic activity required for long distance dispersal (Hall & Muralidharan 1989). Mitochondrial enzymes are comprised of mitochondrial and nuclear DNA encoded subunits (Moritz et al. 1987). Repeated backcrossing would create heterologous African-European subunit combinations that may have reduced enzymatic activity. Suboptimal activity may be detrimental primarily at times of metabolic stress and may not necessarily have an obvious effect on all hybrid bee colonies. Migratory colonies would be affected more than would stationary colonies.

HYBRID ZONES

Selective factors will influence the distribution and character of the bees in the United States and must be considered in control strategies. Hybridization to dilute the African bees was attempted in Mexico (Rinderer et al. 1987, Tew et al. 1988). It would not be feasible to introduce maladaptive genes into the feral population and expect them to persist against strong selection. As African bees approach temperate climates, natural factors will begin to work in favor of European bees. Where selective pressures are not so intense, human intervention to modify the feral population may be able to overcome weak selective forces, although such efforts would have to be continuous (Taylor 1985a, 1988).

Where selective forces are equal, a zone of hybridization will be formed naturally (Taylor 1977, Taylor & Spivak 1984, Taylor 1985a, Lobo et al. 1989, Sheppard et al. 1991). Genotype distributions within the hybrid zone may reveal the nature of selective processes (Barton & Hewitt 1989). If only ecological factors are involved, the genotypes of the bees may represent a "hybrid swarm" but as reciprocal clines of African and European alleles (Taylor 1985a). If genetic incompatibilities exist between African and

European bees, a paucity of heterozygotes may be seen within the hybrid zone. A loss of hybrids as a result of selection would exist in equilibrium with hybrids formed as the parental types migrate inward.

For African bees to introgress beyond the hybrid zone and to survive northern winters, multiple co-adapted factors would have to be inherited from European bees. African-European hybridization within the zone would segregate critical epistatic relationships among physiological, behavioral, and ecological factors. Reestablishment of the relationships through random genetic assortment would occur at a low frequency, especially if it must occur independently among a number of the separate patrilineages that comprise a colony. As a consequence, northern introgression of African bees would be greatly limited (Taylor 1985a). However, unlinked neutral alleles may exhibit more independent introgressive behavior. DNA markers under different selective influences can potentially be found: neutral, non-neutral, and linked to non-neutral genes. An enlarged collection of such markers will greatly enhance studies of gene flow and hybrid zone dynamics.

CONCLUDING REMARKS

The nature of the neotropical African honeybee population continues to be debated. Is it the result of asymmetric gene flow and limited hybrid fitness favoring the African genotype, or is it a product of unlimited hybrid formation and survival of all hybrid genotypes as a "hybrid swarm"? Two recent reports continue to argue that there are no limits to hybridization. This conclusion comes from finding hybrid bees in the South American temperate hybrid zone (Sheppard et al. 1991) and from managed colonies in Mexico shortly after African bee invasion (in effect the leading edge of the migrating front) (Rinderer et al. 1991). Hybrids were identified on the basis of morphometric probabilities. In the same or similar situations, hybridization has been recognized using genetic markers (Lobo et al. 1989, Hall 1990, Taylor et al. 1991). Hybridization in subtropical-temperate transition regions does not reflect the situation in tropical regions. The hybridization in Mexico was not investigated as a transitory step in the dynamic and evolving process of African bee colonization of new territory. The study in Mexico examined only managed colonies yet concluded that feral African colonies were not favored by asymmetric gene flow. Both studies (Sheppard et al. 1991, Rinderer et al. 1991) claimed that, because hybrids were found, no incompatibilities existed between African and European bees. The mere existence of hybrids does not rule out the possible existence of incompatibilities (note the mule).

New World African and European honeybee hybridization is multifaceted. Generalizations cannot be made about the entire process if only limited aspects are studied. The different components must be defined and evaluated: tropical or temperate climates, feral or managed colonies, African or European matrilineages, established or transient populations. It is even possible that hybrids between different European and African subspecies differ in their fitness. The complex dynamics among the components must be recognized to appreciate fully and to understand clearly New World African and European honeybee interactions.

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REFERENCES CITED

- BARTON, N. H., AND G. M. HEWITT. 1989. Adaption, speciation and hybrid zones. *Nature* 341: 497-503.
- BOREHAM, M. M., AND D. W. ROUBIK. 1987. Population change and control of Africanized honey bees (Hymenoptera: Apidae) in the Panama canal area. *Bull. Ent. Soc. America* 33: 34-38.
- BUCO, S. M., T. E. RINDERER, H. A. SYLVESTER, A. M. COLLINS, V. A. LANCASTER, AND R. M. CREWE. 1987. Morphometric differences between South American Africanized and South African (*Apis mellifera scutellata*) honey bees. *Apidologie* 18: 217-222.
- CARON, D. M., AND B. GRAY. 1991. The impact of the Africanized bee on beekeeping in Panama. *Bee Science* 1: 139-143.
- DALY, H. V. 1988. Overview of the identification of Africanized honey bees, p. 245-249. *in* G. R. Needham, R. E. Page, M. Delfinado-Baker, and C. E. Bowman [eds.]. Africanized honey bees and bee mites. Ellis Horwood Limited, Chichester, United Kingdom.
- DALY, H. V. 1991. Systematics and identification of Africanized honey bees, p. 13-44. *In* M. Spivak, M. Breed, and D. J. C. Fletcher [eds.]. The "African" honey bee. Westview Press, Boulder, Colorado.
- FLETCHER, D. J. C. 1978. The African bee, *Apis mellifera adansonii*, in Africa. *Annu. Rev. Entomol.* 23: 151-171.
- FLETCHER, D. J. C. 1991. Interdependence of genetics and ecology in a solution to the African bee problem, p. 77-94. *in* M. Spivak, M. Breed, and D. J. C. Fletcher [eds.]. The "African" honey bee. Westview Press, Boulder, Colorado.
- GONÇALVES, L. S., A. C. STORT, AND D. DE JONG. 1991. Beekeeping in Brazil, p. 359-372. *in* M. Spivak, M. Breed, and D. J. C. Fletcher [eds.]. The "African" honey bee. Westview Press, Boulder, Colorado.
- HALL, H. G. 1990. Parental analysis of introgressive hybridization between African and European honeybees using nuclear DNA RFLPs. *Genetics* 125: 611-621.
- HALL, H. G. 1991. Genetic characterization of honey bees through DNA analysis, p. 45-73. *in* M. Spivak, M. Breed, and D. J. C. Fletcher [eds.]. The "African" honey bee. Westview Press, Boulder, Colorado.
- HALL, H. G., AND K. MURALIDHARAN. 1989. Evidence from mitochondrial DNA that African honey bees spread as continuous maternal lineages. *Nature* 339: 211-213.
- HALL, H. G., AND D. R. SMITH. 1991. Distinguishing African and European honey bee matrilineages with amplified mitochondrial DNA. *Proc. Natl. Acad. Sci. U.S.A.* 88: 4874-4877.
- KERR, W. E. 1967. The history of the introduction of African bees to Brazil. *South African Bee J.* 39: 3-5.
- LOBO, J. A., M. A. LAMA, AND M. A. MESTRINER. 1989. Population differentiation and racial admixture in the Africanized honeybee (*Apis mellifera* L.). *Evolution* 43: 794-802.
- MICHENER, C. D. 1975. The Brazilian bee problem. *Annu. Rev. Entomol.* 20: 399-416.
- MICHENER, C. D., J. ALLRED, H. E. ESCH, N. E. GARY, S. P. HUBBELL, W. C. ROTHENBUHLER, M. V. SMITH, G. F. TOWNSEND, AND J. A. ZOZAYA. 1972. Final Report: Committee on the African honey bee. *Natl. Acad. Sci. USA*, Washington, District of Columbia.
- MORITZ, C., T. E. DOWLING, AND W. M. BROWN. 1987. Evolution of animal mitochondrial DNA: relevance for population biology and systematics. *Annu. Rev. Ecol. Syst.* 18: 269-292.
- OERTEL, E. 1976. Bicentennial bees. Early records of honey bees in the eastern United States. *American Bee J.* 116: 70, 71, 114, 128, 156, 157, 214, 215, 260, 261, 290.
- PELLETT, F. C. 1938. History of American Beekeeping. Collegiate Press, Ames, Iowa. pp. 1-5, 69-67.
- RINDERER, T. E. 1986a. Africanized bees: an overview. *American Bee J.* 126: 98-100.
- RINDERER, T. E. 1986b. Africanized bees: The Africanization process and potential range in the United States. *Bull. Ent. Soc. America* 32: 222-227.

- RINDERER, T. E. 1988. Evolutionary aspects of the Africanization of honey-bee populations in the Americas, p. 13-28. *in* G. R. Needham, R. E. Page, M. Delfinado-Baker, and C. E. Bowman [eds.]. Africanized honey bees and bee mites. Ellis Horwood Limited, Chichester, United Kingdom.
- RINDERER, T. E., R. L. HELLMICH, R. G. DANKA, AND A. M. COLLINS. 1985. Male reproductive parasitism: A factor in the Africanization of European honey-bee populations. *Science* 228: 1119-1121.
- RINDERER, T. E., J. E. WRIGHT, H. SHIMANUKI, F. PARKER, E. ERICKSON, AND W. T. WILSON. 1987. The proposed honey-bee regulated zone in Mexico. *American Bee J.* 127: 160-164.
- RINDERER, T. E., J. A. STELZER, B. P. OLDROYD, S. M. BUCO, AND W. L. RUBINK. 1991. Hybridization between European and Africanized honey bees in the neotropical Yucatan peninsula. *Science* 253: 309-311.
- RUTTNER, F. 1988. Biogeography and taxonomy of honey bees. Springer, Berlin, Germany. pp. 165-257.
- SHEPPARD, W. S. 1989. A history of the introduction of honey bee races into the United States. *American Bee J.* 129: 617-619, 664-667.
- SHEPPARD, W. S., T. E. RINDERER, J. A. MAZZOLI, J. A. STELZER, AND H. SHIMANUKI. 1991. Gene flow between African- and European-derived honey bee populations in Argentina. *Nature* 349: 782-784.
- SMITH, D. R., O. R. TAYLOR, AND W. W. BROWN. 1989. Neotropical Africanized honey bees have African mitochondrial DNA. *Nature* 339: 213-215.
- TAYLOR, O. R. 1977. The past and possible future spread of Africanized honeybees in the Americas. *Bee World* 58: 19-30.
- TAYLOR, O. R. 1985a. African bees: Potential impact in the United States. *Bull. Ent. Soc. America* 31: 14-24.
- TAYLOR, O. R. 1985b. Let's keep our facts straight about African bees! *American Bee J.* 125: 586-587.
- TAYLOR, O. R. 1988. Ecology and economic impact of African and Africanized honey bees, p. 29-41. *in* G. R. Needham, R. E. Page, M. Delfinado-Baker, and C. E. Bowman [eds.]. Africanized honey bees and bee mites. Ellis Horwood Limited, Chichester, United Kingdom.
- TAYLOR, O. R., AND M. SPIVAK. 1984. Climatic limits of tropical African honeybees in the Americas. *Bee World* 65: 38-47.
- TAYLOR, O. R., A. DELGADO, AND F. BRIZUELA. 1991. Rapid loss of European traits from feral neotropical African honey bee populations. *American Bee J.* 131: 783-784.
- TEW, J. E., C. H. BARE, AND J. D. VILLA. 1988. The bee regulated zone in Mexico. *American Bee J.* 128: 673-675.
- WINSTON, M. L., O. R. TAYLOR, AND G. W. OTIS. 1983. Some differences between temperate European and tropical African and South American honeybees. *Bee World* 64: 12-21.