Proceedings of the Third American Bee Research Conference

The 1988 American Bee Research Conference was held at the Hoblitzelle Center of the Texas A & M Agricultural Experiment Station in Weslaco, Texas on October 12 and 13, 1988. The fourth conference will be held at the same place on October 3 and 4, 1989. Abstracts of the proceedings follow.

1. Bram, R. A.[a] – **THE USDA-ARS RESEARCH PROGRAM ON PARASITIC MITES OF HONEY BEES** – In response to new apicultural problems caused by the accidental introduction of the honey bee tracheal mite and the *Varroa* mite, the U.S. Department of Agriculture, Agricultural Research Service (ARS) has forged a substantial program of research on the biology and control of parasitic mites. This program has been achieved through the reassignment and redirection of the equivalent of 5.5 scientist years and accompanying financial resources. In addition to base funding appropriated to ARS, resources for specific projects have been provided by the Office of International Cooperation and Development, the Binational Agricultural Research and Development Fund, the Interregional Project No. 4 of the Minor Use Chemical Group, and by chemical industry cooperators. Three of the major ARS honey bee research laboratories now have component research activities devoted to the identification, biology, and control of parasitic mites. Priorities of the national ARS research program are established to address the greatest needs of the honey bee industry, the program activities of the Animal and Plant Health Inspection Service, and the interests of other state and federal action agencies.

The Beneficial Insects Laboratory, Beltsville Agricultural Research Center, Beltsville, Maryland, provides official service identifications of parasitic mites affecting honey bees and conducts biosystematic research on acarines associated with social bees of the subfamily Apinae. In addition to providing research on classical morphometric techniques for mite identification, whenever feasible identification approaches also include modern experimental methods such as enzyme-linked immuno assay tests and other current bio-technologies. The laboratory is also addressing the problem of controlling the *Varroa* mite. Utilizing a specially renovated, federally approved quarantine laboratory, research is underway to evaluate potential chemical compounds and formulations that can be used by the beekeeping industry to control or eradicate this serious pest. In order to devise regulatory treatment formulations and strategies, field investigations are conducted cooperatively in areas of the country where *Varroa* mites exist in sufficient numbers to provide convincing results. The Beneficial Insects Laboratory also conducts limited research on the control of the tracheal mite with emphasis on efficacy of toxicant compounds.

Parasitic mite research emphasis of the Honey Bee Research laboratory, Weslaco, Texas, is on the biology and control of the tracheal mite. Field activities are geared to acquiring tracheal mite life-cycle data under a variety of environmental and seasonal conditions using different honey-bee strains and management practices. This information, coupled with data on economic thresholds, is being applied to the development of practical control strategies. In addition, the laboratory continues to conduct research to discover chemotherapeutic agents for tracheal mite control.

The Honey Bee Breeding, Genetics, and Physiology Laboratory, Baton Rouge, Louisiana, has initiated research to genetically select or engineer honey bee stocks in order to produce a variety of honey bees resistant to the effects of *Varroa* mites. Selection for Varroa-tolerant honey bees is a long-term commitment that will be conducted in complete coordination with the Beneficial Insects Laboratory, Beltsville, Maryland.

2. Bromenshenk, J. J.,[b] R. C. Cronn,[b] J. J. Nugent,[b] and G. J. Olbu[b] – **BIOMONITORING FOR THE IDAHO NATIONAL ENGINEERING LABORATORY; EVALUATION OF FLUORIDE IN HONEY BEES** – During their foraging activities, honey bees accumulate contaminants from various sources, including soil, air, vegetation and water. Chemical residues in bee tissue and pollen have been used to detect and map the dispersion of trace elements, low-level radionuclides and pesticides (Bromenshenk *et. al.*, *Science* 227:632-634; Wallwork-Barber *et. al.*, *Amer. Bee J.* 122:770-782) from point and area sources over large geographic areas in the United States and Europe. Foraging in honey bees is characterized by a rapidly changing mosaic of forage patches (Visscher and Seeley, *Ecology* 63:1790-1801). With this combination of wide ranging foraging patterns and bioaccumulation of pollutants, honey bees can provide spatially and temporally integrated samples.

For the past two years, honey bees have been used as a surveillance organism for fluoride at the Idaho National Engineering Laboratory (INEL) in southeast Idaho. Industrial processes that release elemental fluoride at the INEL include a fluorinel dissolution process at the Idaho Chemical

Processing Plant (ICPP) and the operation of a fluidized bed coal combustion unit. These releases total an estimated 520g/day, as compared to several hundred kg at regional apatite-ore processing facilities (EPA-450/2-77-005).

Honey-bee mini-colonies, consisting of approximately 10,000 bees, were placed at 26 locations over a 1200 sq. mile area on the INEL during 1986 through 1988. Samples consisted of 150 forager bees aspirated into polyethylene bags from the entrance of each colony. Bees were collected from the mini-colonies at two-week intervals throughout the field seasons. Bees also were obtained from commercial colonies in the region surrounding the INEL. Fluoride was recovered from air-dried bee tissue using calcium oxide as a binding method and analyzed by ion specific electrode (Bromenshenk *et.al.*, *Science* 227: 632-634).

In 1986, fluoride concentrations in bee tissue remained relatively low (8.8-19.0 ppm in dried bee tissue) through the collecting season at off-site locations near the perimeter of the INEL. On the INEL site, early season samples displayed enriched fluoride, but only at a few locations. A value of 65.7 ppm was detected by the ICPP, with levels above 40 ppm at locations 5 km downwind of this facility. This enrichment trend was not evident in late season samples. Fluoride in bees collected on the INEL during late summer ranged from 8.9 to 23.8 ppm. These levels were similar to those observed at locations along the perimeter of the INEL.

Bees were collected from commercial colonies in late summer at sites throughout a large region around the INEL. These samples showed tissue fluoride ranging from 8.8 to 85.3 ppm. The highest tissue fluoride concentrations occurred at Pocatello, far from the INEL. In addition, bees sampled as far as 65 km downwind from Pocatello had levels of fluoride in excess of 20 ppm, indicating long-range transport.

Currently, fluoride is being examined in bee samples collected during 1987 and 1988, following startup of a calcining process at the ICPP. These studies should reveal what affect, if any, operation of the calciner has on fluoride levels on and off of the INEL. In addition, these samples provide an opportunity to assess whether the patterns of long-range transport of fluoride from sources distant from the INEL and short-range transport of fluoride from sources on the INEL persist through time. Finally, ion-chromatography determination of fluoride is being compared to the ion selective electrode as part of methods development to increase sensitivity and reduce analysis time. The objective is to increase the efficacy of using bees as monitors of fluoride. (Research supported by U.S. DOE and the University of Montana.)

3. Bromenshenk, J. J.,[b] J. Fisher,[b] S. Roth,[c] and G. DeGrandi-Hoffman[c] – MODELING THE EFFECTS OF CHEMICALS HAZARDOUS TO BEES – TOXICS is a multi-purpose computer program. It is a data base for chemicals toxic to bees. TOXICS determines anticipated bee mortalities based on probit analysis of published dose-mortality curves. It serves as an input module for BEEPOP, a computer model that simulates honey bee colony population dynamics (DeGrandi-Hoffman *et. al.*, *Ecol. Model.* in press).

The TOXICS data base is derived from the work of E. L. Atkins *et. al.* (*Univ. of Calif. Leaflets* 2883, 2286, and 2287) concerning the toxicity of pesticides and other agricultural chemicals to honey bees. The probit analysis utilizes LD50 and slope values to reconstruct the dosage-mortality curve to obtain other LD values. When an exposure concentration within the range reported to be toxic to bees is entered via the keyboard, the program provides an anticipated percent mortality of adult bees. TOXICS is being expanded to include a more comprehensive review of the literature with an emphasis on incorporating the results of field studies of chemicals hazardous to bees. The program will provide the user with the literature citation for each dose-mortality curve. It will include on-screen help for the interpretation of mortality estimates, as well as warnings concerning limitations of extrapolations to the "real world." This program will enable the user to use the probit analysis to generate LD values for specified percent mortalities.

Currently, this program, written in FORTRAN, is installed on the UM VAX mainframe computer. It can be installed and run on an IBM compatible PC. Also, it can be used as an input module for BEEPOP.

BEEPOP is an interactive model that estimates colony population size. Input parameters include initial colony population size, starting date of the simulation, the reproductive status of the queen, number of days a bee can forage before expiring, duration of the simulation, and weather conditions. In addition, BEEPOP has been modified to allow the user to predict the effects of toxic events on population size. A toxic event is simulated by entering a percent mortality either from the keyboard or from the output of TOXICS. The input menu allows the user to specify the day, duration and intensity (degree of mortality) of the toxic event. The user may specify differing levels (0-100%) of mortality for eggs, larvae, pupae, and adults of worker bees and drones.

Extensive testing and modification of BEEPOP is planned for the coming year. We are conducting field verification and validation studies using colonies of bees at Superfund Hazardous waste sites. We are most interested in obtaining field base data, especially data relating colony population size or weight to climatic conditions. (Supported by Cooperative Research Agreement CR-814456-01-0 with the EPA Corvallis Environmental Research Laboratory and by the University of MT.)

4. Bromenshenk, J. J.,[b] J. L. Gudatis,[b] R. C. Cronn,[b] G. J. Olbu[b] – UPTAKE AND IMPACT OF HEAVY METALS TO HONEY BEES – The uptake of arsenic (As), cadmium (Cd), lead (Pb), copper (Cu), magnesium (Mg), manganese (Mn) and zinc (Zn) by forager honey bees was evaluated using mini-colonies located on a fallow alfalfa field near a lead smelting complex at East Helena, Montana. Fifteen bee colonies were placed individually in flight enclosures; while three colonies were allowed to fly freely. Six flight cages were enclosed with polyethylene film, and all incoming air was filtered. In three of these units, the vegetation was removed and the soil covered. In the other three units, soil and vegetation remained accessible to the bees. Nine enclosures had open-mesh sides and a forced air system to provide a constant flow of unfiltered air. The vegetation was cut and removed from three of these cages; the soil was covered in three, and soil and vegetation were left in three. Metal concentrations in the ambient air were continuously measured by three high volume air samplers. Forager bees, alfalfa blossoms and foliage, duff, and soil surficial dust were sampled each week from July 3 to August 5, 1986.

The presence or absence of vegetation or soil had little or no effect on bee body burdens of metals. Arsenic and lead concentrations for bees exposed to ambient air in cages were not significantly different ($P > 0.05$) from those of free flying bees. Cadmium concentrations were higher in free flying bees, while copper concentrations tended to be lower. Magnesium and zinc tended to be lowest in free flying bees.

Body burdens of arsenic, cadmium, copper, and lead increased significantly over the five-week period and paralleled increases in the concentrations per cubic meter of these chemicals in the ambient air. Magnesium values for bees remained relatively constant, while manganese and, to some extent, zinc levels decreased over the time span of the experiment. Magnesium and manganese concentrations in the ambient air decreased, whereas zinc values increased.

Concentrations of cadmium and lead were significantly correlated ($P < 0.05$) with the concentrations per cubic meter of air. Analysis of temporal patterns of toxic metals in the ambient air, highly significant treatment effects, and correlative fingerprints for these seven metals in bees and on filters from Hi-Volume air samplers indicate that exposure to ambient air is the primary source of arsenic, cadmium, lead and copper to forager bees.

These results are important to the interpretation of potential impacts on honey bees. We have investigated occasional severe bee kills at sites near smelters that have been inactive for several years. We also have observed statistically significant reductions of population size and increase of brood mortality in bee colonies near operating smelters (Bromenshenk *et. al.*, in prep.). The most severe effects seem to occur at two times: in the early Spring and mid- to late-summer. During spring buildup of populations, the colonies are under considerable stress and have a preponderance of older bees that may have had long-term exposure (*i.e.*, over the winter). Late season bees often are exposed to dry conditions, when aerial deposition of metals in dusts can be expected to be at a high. (Supported by EPA Assistance Agreement R-812525-01-1, Office of Exploratory Research, and by the University of MT.)

5. Collins, A. M.[d] and R. L. Hellmich[e] – DISRUPTION OF DEFENSIVE BEHAVIOR[u] – When a colony of Africanized honey bees is disturbed, a large number of worker bees will respond with defensive behavior including threatening and stinging, which can persist for considerable time. In response to the needs of beekeepers to have a way to disperse defending bees that may follow from the apiary and for emergency rescue or other people to disperse attacking bees, a variety of chemicals were evaluated for their ability to repel such honey bees.

Three preliminary trials were conducted using cooking oil, deodorant, sugar syrup, oil of bitter almond, eucalyptus oil, spearmint oil, insect repellent (N, N-diethyl-meta-toluamide), canned smoke, Bee-go (butyric anhydride), resmethrin, baygon, wasp and hornet spray, chlorox, and dish detergent. Some of these products were diluted in paraffin oil or water to facilitate spraying from a hand spray bottle or pump. Controls were paraffin oil alone, water, or nothing sprayed at the bees. Comparisons of numbers of stings in a suede leather patch waved by the experimenter for 20s after spraying a chemical for 15s, indicated that oil of bitter almond, insect repellent, and spearmint oil significantly ($P < .05$) reduced the number of stings.

A fourth test compared insect repellent, benzaldehyde (the major component of oil of bitter almond), wasp and hornet spray, mint flavored water, and two controls, water and nothing. The number of bees around the experimenter was counted from a video tape for 3 time periods: before spraying (10s), during spraying (15s), and during the waving of a suede leather patch (30s). The number of stings was counted from the patches. The table shows these results. Insect repellent, benzaldehyde and to a lesser extent, wasp and hornet spray, were effective in dispersing bees. Further work is planned to evaluate related compounds for effectiveness and safety for human use.

Table – Number of bees around experimenters and number of stings in leather patches for a spray treatment by various chemicals.

	No Stings	No. bees Before Spraying	No. bees With Patches
Insect repellent	1.83	64.3	9.2	6.6
Benzaldehyde	5.16	53.4	10.1	16.2
Mint in water	16.33	54.1	30.6	25.7
Water-control	19.17	54.1	38.8	47.4
Nothing-control	14.67	28.9	27.0	29.8
	$F = 3.26$ $P = 0.03$		$F = 10.2$ $P < 0.0001$	

6. Cox, R. L.,[d] J. O. Moffett,[d] M. Ellis[f] and W. T. Wilson.[d] LONG-TERM BENEFICIAL EFFECTS OF MENTHOL TREATMENT ON HONEY BEE COLONIES INFESTED WITH TRACHEAL MITES *(ACARAPIS WOODI)*[u] – Honey bee *(Apis mellifera)* colonies infested with tracheal mites *(Acarapis woodi)* were treated with solid menthol cakes in three different experiments near York, Nebraska. Colonies were treated in the spring only, spring and autumn, or autumn only.

In the first experiment there were 90% fewer live mites/bee in the menthol-treated colonies at the end of the ten-week treatment period in the spring. Beginning in April '87 the remaining 29 living colonies involved in that test were monitored for one year following treatment to determine the long-term effects of the treatment on the mite and honey-bee populations. The mite infestation level (mean percent of individual bees infested/colony) was significantly lower from May, 1987 until April, 1988 in colonies treated with menthol (*i.e.* from 90-97% mite control with menthol compared to untreated checks). In addition, 50% of the untreated colonies died during the winter, while none of the treated colonies died. The untreated colonies that survived the stress of winter had significantly smaller populations (i.e. fewer frames of adult bees and a smaller brood area) than the colonies treated with menthol (Table).

In the second experiment, 32 nucleus colonies were treated in the spring in south Texas and Nebraska, and again in the autumn in Nebraska. The mite infestation level was reduced by seven weeks of menthol treatment in April and May, 1987 (from 25.3 to 1.6%), while the infestation level in untreated colonies decreased only slightly (from 22.9 to 18.8%). Following a second treatment in the autumn, tracheal mite populations increased in the untreated colonies, while they remained low in the treated colonies. During the period of Sept. 1987 until April 1988, there were, on the average, 98% fewer infested bees/colony in the menthol-treated colonies compared to checks. In April 1988 there was no significant difference between either winter survival or bee brood populations in colonies treated with menthol the previous year and untreated colonies.

In experiment #3, colonies that were treated with menthol in autumn only, contained very few mites initially (*i.e.* the percent of bees infested with mites in 39 of 40 colonies was less than 1% on Sept. 16, 1987). In April 1988 the menthol-treated colonies had 50-79% fewer mite-infested bees than the untreated checks. However, in the untreated group the average infestation level was under 2%.

In all three experiments the menthol evaporated slowly due to relatively cool temperature conditions. Only about 22-33% of the menthol in the cakes evaporated in the hives during each 7-10 week exposure period. A more efficient method of application is needed.

TABLE – Winter survival, mean number of frames of adult bees/colony, square inches of sealed brood/colony and mean percent of bees infested with *Acarapis woodi*/colony treated with menthol one year earlier. Near York, Nebraska. April 19, 1988.

Treatment	No. of colonies Nov. 87	No. of colonies Apr. 88	Adults (frames)	Sealed brood (in^2)	Infestation (% pos.)
0 grams	6	3	4.0a*	540.3a	19.1a
25 grams	5	5	8.6ab	1005.4ab	0.6b
50 grams	4	4	10.4b	1338.1b	0.0b
75 grams	6	6	10.5b	1131.8ab	1.8b

*Means within the same column followed by a different letter are significantly different ($p < .05$) by ANOV and Duncan's new multiple range test (SAS Institute, 1985).

7. Dietz, A.,[g] C. Vergara,[g] D. S. Hurley[g] and F. A. Eischen[g] – PRODUCTION OF TRACHEAL MITE-FREE HONEY-BEE QUEENS WITH APITOL® – Twelve 3 lb. (1.5 kg) packages of honey bees were obtained from 9 colonies infested with *Acarapis woodi* (av. infestation was

64.9%) and transferred to 3 groups of small mating nuclei ("baby nuclei"). There were 24 nuclei in group A (control) and 23 mating nuclei each in groups B and C. Each nucleus, containing 2 small combs and a feeder, was established by adding roughly 250 gm of bees and a queen cell ready to emerge. The treatments used consisted of 250 ml of 50% sucrose syrup (wt/wt) in group A (control), and syrup containing either 0.3 mg/ml (group B) or 0.5 mg/ml (group C) of cymiazole (2-[2,4-dimethylphenyl-imino]-3-methyl-4-thiazole hydrochloride, Apitol (GRA. 17.5% cymiazole) Ciba-Geigy Ltd.

The newly mated queens, and approximately 20 adult worker bees, were removed every 2 weeks, placed in alcohol, and subsequently dissected to determine the levels of infestation. Fresh treatments, as well as new queen cells, were added every 14 days. Mating success rates and infestation levels of queens and worker bees were determined.

There were no statistical differences in the mating success rates of queens in treatment groups B and C during the three sampling periods. Considerable fluctuations occurred, however, in the control group A. There was a significant decline in the mating success rate of honey bee queens in the group A after the second sampling period. Statistically, there were no differences in matings between queens in groups A and B for the 3rd sampling period.

Mite control was excellent in treatment group C. Only a single queen (5.9%) was found to be infested after the first inspection period, or 14 days. No further mite infestation was found on successive inspections in group C. In group B, 46.6% of the mated queens removed from the nuclei were infested after the first inspection. The infestation rate of 16.7% for mated queens in treatment group B was identical for the 2nd and 3rd sampling period. The infestation rate of mated queens removed from the control group A was 100%. All mated queens in the control group A were found to be heavily infested with mites after the 1st and 2nd sampling period. There was a significant decline in the infestation rates of mated queens and worker bees in the control group A for the 3rd sampling period. This decline may be due to the replacement of many of the old and infested worker bees by emerging young bees.

The mite populations in worker bees did not decrease substantially in all treatment groups until the 3rd sampling period. A distinct decline in the bee populations of the control group A occurred between the 2nd and 3rd sampling period.

One of the most interesting findings was that there appears to be essentially no mite transfer between queens and bees feeding on sucrose solution with Apitol concentrations of .5mg/ml. This observation seems to indicate that Apitol will inhibit mite migration at such concentrations. The benefit of Apitol treatment was demonstrated by the production of honey-bee queens essentially free of mites in mating nuclei established with infested bees. A concentration of .5 mg/ml of sugar solution appears to be required. In order to prevent a decline in bee populations, it should not be fed continuously to bees in mating nuclei.

8. Eischen, F. A.,[s] W. T. Wilson,[d] J. S. Pettis,[d] and J. Vargas[h] — THE SPREAD OF *ACARAPIS WOODI* IN NORTHEASTERN MEXICO — Colonies of honey bees *(Apis mellifera)* in northeast Mexico were examined for honey bee tracheal mites during the period 1984-1988. Nearly all apiaries contained mite-infested colonies. This appears to be a substantial increase from previous surveys made in 1983 by Mexican investigators, and probably reflects a rapid rate of spread. On average about 60% of all colonies were parasitized. The prevalence of colonies having 30% or more of their workers infested was about 40%. This level of parasitism is associated with economic damage.

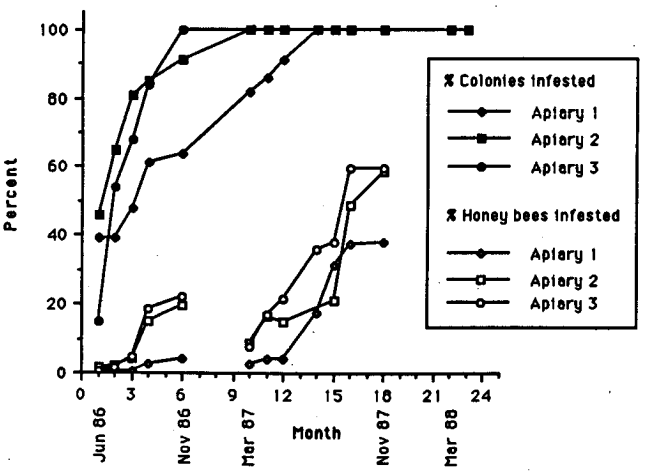
Northeastern Mexico is rather dry and the biome characterized by mesquite scrub. Rivers and their associated irrigation channels are foci of agricultural activity, including beekeeping. Citrus is the most important crop and during the spring bloom, colony density then becomes high. Unfortunately, this is when both *A. woodi* populations are highest, and colony failure is most likely due to a number of causes including *A. woodi*. These conditions lead to the robbing of weak colonies which are frequently heavily infested. Drifting by worker bees during truck transport to the citrus orchards and during the nectar flow also promotes the spread of *A. woodi*.

In addition to providing attractive resource locations for beekeepers, rivers may serve to channelize normal bee movements. Rivers and their associated plant communities often provide nest sites, food, and prominent landmarks. Swarming and absconding flights are more probable along waterways than away from them. Beekeeping practices have undoubtedly spread *A. woodi*. However, these practices may be amplified by the natural movement of the honey bee population along riparian habitats.

9. Furgala, B.,[i] S. R. Duff,[i] S. Aboulfaraj,[i] D. W. Ragsdale,[i] and R. A. Hyser[i] — SOME EFFECTS OF THE HONEY BEE TRACHEAL MITE *(ACARAPIS WOODI)* ON NON-MIGRATORY HONEY BEE COLONIES IN EAST CENTRAL MINNESOTA — Seventy-five honey-bee colonies in three commercial apiaries with an initial low incidence of honey-bee tracheal mites (HBTM) (<1%) were monitored during a 22 month period to determine the effect of infestation on non-migratory, wintering honey bee colonies. The apiaries were managed by the commercial honey producer. Samples of 100 honey bees were taken from each colony in June, July, Aug., Sept., and Nov. 1986; March, April, May, July, Aug., Sept., and Nov. 1987; and March 1988. Oxytetracycline and fumagillin were used as chemotherapeutic agents. All colonies were requeened in May 1987 with Starline queens from a single source.

Low levels of HBTM infestations were initially detected in 33% of the honey bee colonies (see figure). Thirteen months later, 100% of the surviving colonies were infested. The spread of infestation among colonies may have been facilitated by the paired arrangement of the colonies, and activities that promote drifting, such as harvesting the honey crop.

Percentage of colonies infested with HBTM, and the percentage of honey bees infested in each of 3 apiaries.



The percentage of honey bees that were found infested increased as the study progressed (see figure). During the first season the mean percentage of honey bees infested increased dramatically in apiaries 2 and 3, and modestly in

apiary 1. During the second season the mean percentage of honey bees infested in the colonies that survived the winter of 86/87 peaked at approximately 60% in apiaries 2 and 3, and 40% in apiary 1. Overall the mean percentage of honey bees infested increased by a factor of over 50.

During the 22 month period the number of colonies in the study declined from 75 to 11. Fourteen colonies failed to survive the winter of 86/87 and 37 failed to survive the winter and early spring of 87/88. Eleven colonies were very weak in late summer and therefore were not prepared for the winter of 87/88. Two colonies dwindled during other periods of the study.

These results demonstrate that under Minnesota conditions HBTM can spread among colonies, increase rapidly within colonies, and that winter survival is minimized when high late summer and early fall levels of HBTM exist in nonmigratory colonies.

10. Hall, H. G.[j] and K. Muralidharan[j] – AFRICAN HONEY BEE MIGRATION INTO MEXICO FOLLOWED WITH DNA MARKERS – Important understanding of African honey bee population dynamics is being obtained with mitochondrial DNA (mtDNA) and nuclear DNA restriction fragment polymorphisms. We have used two restriction enzymes, EcoRI and BglII, to distinguish European and African mtDNA (fragment patterns previously reported – Smith & Brown, *Experientia* 44:257-260; Moritz *et al.*, *Experientia* 42:322-324; Sheppard & Huettel, *Amer. Bee J.* 127:851). Thirteen feral swarms caught in bait hives near Tapachula, Mexico, in January 1988, all carried African mtDNA. Out of six managed colonies established prior to African bee arrival, five carried European mtDNA and one had African mtDNA. Ten colonies tested from Venezuela all had African mtDNA. Almost identical results were obtained independently by D. Smith at the University of Michigan.

MtDNA is maternally inherited. It is not transmitted by mating drones, but only transported by queens accompanying swarms. For the Mexican swarms to carry African mtDNA, they had to be progeny of unbroken maternal lineages from the African bees originally introduced in 1957; not a single intervening generation since that time could have been the result of a African drone mating to a European queen. These findings dispute the contention that the primary driving force of Africanization is through the flight of mating drones (Rinderer *et al.*, *Science* 228:1119-1121; Rinderer, *Bull Ent. Soc. Amer.* 32:222-227) and support the view that the migrating "Africanizing" force consists almost exclusively of swarms from feral populations that have largely retained an African genetic integrity (Taylor, *Bull. Ent. Soc. Amer.* 31:14-24; Taylor in Needham *et al.* eds, 1988, *Africanized Honey Bees & Bee Mites*, 29-41)

Nuclear DNA analyses (Hall, *Proc. Natl. Acad. Sci. U.S.A.* 83:4874-4877; Hall in Needham *et al.* eds, 1988, *Africanized Honey Bees & Bee Mites*, 287-293) show that the established Mexican colonies, carrying European mtDNA, have become "Africanized" by gene flow from African drones. Such "Africanized" bees, however, apparently do not contribute to the spread of the African honey bee, and, unless actively propagated by beekeepers, probably disappear through attrition. Nuclear DNA markers reveal that feral Mexican bees are "Europeanized" to a minor extent, but to a greater extent than are Venezuelan bees. European alleles that enter the African population during frontal passage seem to disappear later. This coincides with a previously reported change towards a more African morphology and may be due to an overwhelming increase in African gene frequencies as the feral population becomes established (Boreham & Roubik, *Bull. Ent. Soc. Amer.* 33:34-38).

The importance of direct takeovers of European colonies by African queens has not been clearly documented, but can be determined using mtDNA. The established Mexican colony with African mtDNA may have resulted from a takeover. A number of mechanisms may exist that select against European-African hybrids, hence against "Africanized" colonies. Such mechanisms would also tend to eliminate European alleles in feral populations because of their lower allele frequency. Metabolic differences, dictated by mitochondria, may provide African maternal lines the selective advantage over European bees in tropical climates. Perhaps, swarms with African mtDNA constitute the migrating force because they have the intense metabolic capability needed to fly long distances. Sub-optimal interactions between mitochondrial and nuclear coded metabolic enzymes could contribute one of the possible disadvantages in hybrids.

11. Harbo, J. R.[e] – SPERM COMPETITION – Sperm competition implies that there are physiological (*e.g.* age, nutrition) or genetic differences in spermatozoa which affect their ability to fertilize eggs. If sperm competition exists in honey bees (and I shall present evidence that it does), it could affect the propagation strategies in a selective breeding program.

There are numerous examples of how a queen produces significantly different proportions of genetically marked worker progeny at different times. This variation is generally attributed to non-mixing of spermatozoa either during sperm migration to the spermatheca or during storage in the spermatheca. However, if non-mixing is the cause, then a group of queens that is instrumentally inseminated with the same semen mixture should show only random changes in their proportions of marked workers. This was not the case. The changes within a group were often unidirectional.

Data from three experiments showed such unidirectional differences and also suggested that both semen dosage and time can affect the proportion of marked progeny. Each experiment involved a group of mutant queens inseminated with a mixture of mutant and wild-type spermatozoa; each mixture was diluted 1:1 with saline and stirred for 5 minutes (Harbo, *Amer. Bee J.* 127:845). The sample size was 500 workers per queen. In the first experiment the mutant frequency was about 2%. A group of 18 queens produced 1.5% mutant progeny in the fall and 2.4% the following spring. The difference was statistically significant. In 2 other experiments, a group of about 20 queens was inseminated in random sequence with either a very tiny insemination volume (0.2 μl) or a more normal volume (6 μl). One experiment had a mutant frequency of 5%, the other 50%. In both experiments, the insemination volumes had a significant effect on the proportion of mutant progeny ($P<0.01$).

In one experiment, spermatozoa were collected from the spermatheca of 10 queens that had been inseminated with 6 μl of semen and then used to inseminate 10 queens (supersisters to the donor queens). The donor queens had produced 51.5 ± 4.8% (mean ± SD) mutant progeny (about 1100 progeny were counted from each queen). The queens receiving the sperm produced a significantly higher proportion of mutant progeny than the donor queens. Four recipient queens that produced 500 or more worker progeny produced 73.7 ± 9.9% mutants. The remaining 6 queens each produced only 19 to 243 workers, but had a similarly high proportion of mutants (82.9 ± 11.1%). The mutant and wild-type drones in this experiment came from two different queens, so their sperm were genetically and environmentally different. The higher proportion of mutant progeny from queens inseminated with spermathecal sperm indicated that one of the sperm populations may have survived the transferring process better than the other. This differential survival may be a form of sperm competition that may also occur during *in vitro* sperm storage and during sperm migration to the spermatheca.

Dominance of a certain sperm type at different times does not seem to be caused by differential survival, for such

dominance is often reversible. Another form of competition must exist that shifts the advantage to a particular population of sperm at different times. This change in competitive advantage may be environmentally induced (for example when there are many spermatozoa released per egg, when a queen has very few spermatozoa in her spermatheca, when seasons change, or when a queen first begins to lay). Although simple non-mixing of spermatozoa could not be the only cause (for reasons cited earlier), imperfect mixing probably occurs and this could enhance or reduce competitive effects among spermatozoa.

12. Harris, J. W.[k] and J. R. Harbo[e] — BREEDING FROM WORKER HONEY BEES — The production of unfertilized eggs from a single worker honey bee and the rearing of adult drones to be used in instrumental inseminations of queen bees are keys to selective breeding from laying workers. Because of racial differences in ovary development (Ruttner & Hesse, *Apidologie* 12: 159-183), single Cape bee (*Apis mellifera capensis* Esch.) workers have become laying workers and 'false queens' when placed in a population of *A. m. carnica* workers and maintained in laboratory cages (Velthuis & van der Kerk in Needham *et al.* eds, 1988, *Africanized Honey Bees & Bee Mites*, 80-86; Verma & Ruttner, *Apidologie* 14:41-57).

When differences in ovary development periods between worker honey bees are small, as is the case between workers of the various European races and members of the same colony, reproductive competition increases to a point that more than one worker becomes a laying worker upon queenlessness. To circumvent this problem we describe a scheme for limiting reproductive competition within groups of 25 caged bees. The system requires the replacement of the 24 workers surrounding a selected worker at 5-day intervals. One marked worker was placed in each of 10 incubator cages that contained honey, water, pollen, and a two-sided, framed drone comb measuring 8.5 cm x 5 cm. Each cage was provided an additional 24 sister workers. All workers were young, non-flying bees shaken from brood combs of a single colony. After 5 days the 24 unmarked workers from each cage were removed and frozen until their ovaries could be dissected, and 24 new sisters (emerged in an incubator and less than 24 hrs. old) were added to each cage. Five days later, all bees were frozen. Two of the 10 marked workers died on the 6th day and were not dissected. Ovariole number and extent of ovarian development were determined for samples from both groups of fostering bee populations and the eight remaining marked workers using a ranking system developed by Velthuis (*Ent. Exp. & Appl.* 13:377-394).

The results in the table show that three of the remaining eight workers became sole layers. No eggs were laid by the attendant workers, for the first group of attendant bees was removed before eggs were laid, and we found no bees with highly developed ovaries in the second group.

Table — Ovary development of marked workers and attendants. The marked workers were in the cage for 10 days; the first and second sets of attendants were in the cage from days 1-5 and 6-10, respectively.

	Marked worker			% attendants having stage II or III ovary development	
Cage No.	Stage of ovary devel.*	Onset of egg laying	No. of eggs	1st set of attendant bees (no. dissected)	2nd set of attendant bees (no. dissected)**
1	I	---	0	70% (10)	20% (10)
2	III	day 9	8	50% (10)	33% (21)
3	III	---	0	50% (10)	30% (10)
4	II	---	0	10% (10)	20% (10)
5	III	day 10	4	40% (10)	27% (22)
6	III	---	0	40% (10)	10% (10)
7	III	day 9	10	60% (10)	4% (24)
8	II	---	0	40% (10)	10% (10)

*I = resting or inactive, II = early stages of development when eggs are rounded to bean-shaped, and III = presence of sausage-shaped eggs.

**No class III ovaries found in samples from 2nd set of attendants.

13. Hellmich, R. L.[e] and A. M. Collins[d] — FACTORS THAT INFLUENCE DRONE FLIGHT TIMES IN AFRICANIZED AND EUROPEAN COLONIES — The objective of this research was to identify colony factors that influence drone flight times. Possibilities addressed in this experiment included: Workers influencing drones, or drones influencing each other. Such an analysis was possible because there are distinct differences in departure times of Africanized and European drones. European drones fly 10-30 minutes earlier than Africanized drones; and flight times of European drones are 50-90% more variable (Hellmich, *Amer. Bee J.* 127:846). We observed drones leaving composite colonies (various combinations of workers and drones) to determine which colony factors were important.

The experiment was conducted in an apiary near Acarigua, Venezuela. Africanized and European drones were demarcated with enamel paint, then put into colonies (*ca.* 100/colony). Twelve observers made counts (2 min) on 36 colonies by rotating among colonies. There were six treatment combinations: Africanized colonies with only European drones, with only Africanized drones, or with a mixture of Africanized and European drones; and European colonies with the same three combinations.

Mean Differences. European drones flew significantly earlier than Africanized drones in both non-mixed and mixed drone conditions (table). (This confirms previous results.) Effect of colony type, though, in both cases, was not significant which suggests that drone flight times are not influenced appreciably by workers. Africanized drones flew at similar times, whether they were flying from colonies with just Africanized drones or from colonies with both drone types. These results were similar for European drones flying from the two types of colonies. This outcome suggests that drone flight times are not influenced appreciably by the presence of other drones.

Variance Differences. Within-colony variance for European drones was significantly higher than that of Africanized drones in both non-mixed and mixed colonies (table). (This also confirms previous results.) However, colony effect on this variance appears to be the opposite. Drones flying from Africanized colonies had more variable flight times than those flying from European colonies. This was significant for non-mixed colonies and suggests that workers influence the variability of drone flight times. Flight-time variances of both Africanized and European drones changed only slightly when the drones were flying from non-mixed colonies and when they were flying from mixed colonies. These results suggest that the flight-time variability of drones is not influenced appreciably by the presence of other drones.

Table. Flight-time means (± SE in min) of European (E) and Africanized (A) drones flying from non-mixed (A or E drones) and mixed (A + E drones) colonies, flight-time means of drones flying from non-mixed and mixed European and Africanized colonies, and within-colony variances (min²; no. of drone flights in parenthesis) for drone flight times for same combinations.

	Flight-time means		Within-colony variances for drone flight times	
Drone Effects	Eur. drones	Afr. drones	Eur. drones	Afr. drones
Non-mixed Colonies	3:56 ± 2.6'	4:09 ± 2.7' *	1881 (1545)	1491 (1709) **
Mixed Colonies	3:50 ± 5.6'	4:16 ± 3.7' *	1899 (419)	1385 (917) **
Colony Effects	**Eur. cols.**	**Afr. cols.**	**Eur. cols.**	**Afr. cols.**
Non-mixed Colonies	3:56 ± 2.6'	4:09 ± 2.7' ns	1567 (2083)	1862 (1204) *
Mixed Colonies	3:50 ± 5.6'	4:16 ± 3.7' ns	1499 (864)	1632 (472) ns

(* $P < 0.005$; ** $P < 0.0001$; ns $P > 0.05$)

14. Lebougle, J. M.,[l] J. A. Gutierrez,[l] E. Yarce,[l] and M. Mendez[l] — EVALUATION OF PHEROMONES AND

BAIT HIVES FOR AFRICANIZED HONEY BEES – The objective of our study is to develop a new type of bait hive for capturing Africanized honeybee (AHB) swarms to replace the cardboard bait hives widely used by the Agricultural Ministry of Mexico (SARH). For this purpose we established two trap lines of 250 bait hives in Las Choapas, Ver., and Tapanatepec, Oax. We selected these two places because (a) they have a high rate of AHB swarms, (b) they probably represent two distinct migratory fronts, (c) they have different vegetation and climate, and (d) they have a similar number of managed hives and technical level of beekeeping.

Each line has 80 cardboard bait hives (SARH-I), 10 pressboard bait hives (SK-I), 80 cardboard bait hives with pheromone (SARH-II), and 80 pressboard bait hives with pheromone (SK-II). The lure we are using is Taylor's African formulae which is placed at the entrance of each bait hive. The cardboard bait hive is exactly the same as SARH is using in Central and Southern Mexico and includes a blue plastic bag and a round entrance at the bottom. Pressboard bait hives are the same as those used by Taylor in Kansas, Schmidt in Tucson, and Roubink in Cd. Victoria, Tamps., Mexico. Bait hives were randomly placed and all were at a height of 2-3 m.

Our work will continue until April of 1989 in order to cover the migratory and reproductive seasons. In a second batch of bait hives we are also testing different kinds of lures: (a) Taylor's European formula; (b) Taylor's African formula; (c) three different mixtures of citral-geraniol-mineral oil; (d) three different concentrations of lemon grass water; and (e) queen substance pheromone. Both studies also includes morphometrics (forewing length, femur length and III sternite), and electrophoresis of MdH and HK.

Field activities in Las Choapas and Tapanatepec area started in the second half of July. By the end of August, in the Tapanatepec area we found 25 swarms, 16 of them in SK-II and 9 in SARH-II. In the Las Choapas area from the second half of August to the end of September, we also captured 25 swarms: 13 in SK-II, 10 in SARH-II and only 2 in SARH-I. These last two swarms were captured very recently. Our statistical analysis using SPSS PC + shows a significant difference in the number of swarm captures between SK-II and SARH-II, with the SK bait hives being more attractive overall and more efficient by capturing both the very small and the very large swarms.

Some other results were: average weight for the entire population (50 swarms) was 1.37 kg (SD = 1.26); average weight for Las Choapas area (25 swarms) 2.14 kg (1.31); for Tapanatepec area (25 swarms) 0.61 kg (0.54); average weight for SK-II traps (29 swarms) 1.46 kg (1.43); for SARH-II (19 swarms) 1.15 kg (0.91); and for SARH-I (2 swarms) 2.25 kg (1.77). Since all our results are preliminary, we think more data are needed before reaching any firm conclusions.

15. Loper, G. M.,[c] W. W. Wolf,[t] and O. R. Taylor, Jr.[o] – 1988 UPDATE ON THE USE OF A GROUND-BASED X-BAND RADAR TO STUDY THE FLIGHT OF HONEY-BEE DRONES – Our studies of mating biology require knowledge of drone flight patterns and locations of drone congregation areas (DCA's). A desert area was selected which is basically flat except where small washes (0.5-1.0 m deep) cross, generally from South to North. Small (4-5 m) mesquite *(Prosopis)* trees line the washes and occur in scattered small groups; generally the area has scattered, small (1-1.5 m) creosote *(Larrea tridentata)* and catclaw *(Acacia greggii)* bushes, but some areas (100 by 150 m) are barren. One apiary of 60 colonies was near the center of this area, but a 12-colony abandoned apiary was discovered (in 1988) which also contributed drones.

The USDA-ARS Entomological radar unit was used in March and April of 1987 and 1988 at a total of 36 separate positions. By triangulating on the areas of intense drone activity and referring to an aerial photo, we have been able to get a "bird's-eye" picture of the flight patterns and DCA locations in this area. In 1987, we concentrated on a smaller (1.8 x 1.2 km) area adjacent to the major apiary. In 1988, the study area was extended in all directions to encompass an area 4.2 by 2.5 km. This included a fallow cotton field (1 by 1 km) close to the apiary.

DCA's and flyways between them were found to be in the same places on consecutive days and in both years. Our definition of a DCA now is based on the radar documentation of drone flight in small (80-100 m diameter) areas which extend upward as high as 30 m, whereas drone flight in the flyways was generally below 15 m. The major flyways formed parallel to and about 80 m from the edge of the strongest tree lines in the area. Many of the DCA's formed where the flyway branched. That is, as the drones flew away from the apiary, they encountered strong visual cues (tree line along major wash), turned to fly alongside it (both N and S) and at intervals some branched off at angles (*ca.* 100-120°) to form a "minor" flyway some of which were also observed to branch again. At these branch points, large numbers of drones "accumulated" and flew much higher in these generally restricted areas. Some of the DCA's were essentially circular in cross-section; some were elongated in the flyway, but became more circular at higher elevations.

We hypothesize that the physical limitations and arrangement of at least some of the drones' omatidia constrain their visual discrimination and flight behavior resulting in their formation of flyways at least 60 m from the tree line and possibly also in the location of DCA's (branch points). Some of the DCA's were located where drones were approaching within 100-250 m of some of the scattered "clumps" of larger trees not associated with the tree line. It was also obvious that the drones avoided barren areas when the flyways approached them. No drone flyways or DCA's were ever found over the fallow, "featureless" cotton field (a 12 minute VHS video was presented to elucidate the techniques and results).

16. Moffett, J. O.,[d] W. T. Wilson,[d] R. L. Cox,[d] and M. Ellis[f] – FOUR FORMULATIONS OF AMITRAZ REDUCED TRACHEAL MITE, *ACARAPIS WOODI*, POPULATIONS IN HONEY BEES[u] – Preliminary studies have indicated that Amitraz may be of value in controlling both tracheal and *Varroa* mites. Therefore, in April, 1988, 4 formulations of Amitraz (N-methyl-bis (2,4-xylyl-iminomethyl) amine) were applied to colonies infested with tracheal mites in northeast Nebraska.

Two experimental apiaries of 25 colonies each were established from three apiaries heavily infested with tracheal mites. Each treatment was applied to 5 colonies/apiary. Each treatment group of five was placed at least 30 meters from any other group to reduce the drifting of bees among treatments.

Four formulations of Amitraz were tested: 1. aerosol containing 2.5% active ingredient (AI); (2) ampules containing 12.5% AI (EC); (3) cattle bands (red plastic strips) containing 10.0% AI; (4) EVA plastic strips (white) containing 10.0% AI; and (5) no acaricide treatment (control). The treatments were applied to the first apiary on April 20 and to the second apiary on April 21. The aerosol treatment was applied to both apiaries again on May 4. The aerosol was sprayed over the top bars for 10 seconds/colony. One ampule was broken on each bottom board in treatment 2. In both treatments, 3 & 4, two of the 20.3 x 2.5 x 0.2 cm strips were suspended by a nail between the brood combs of each treated colony. The strips were removed after 6 weeks (June 3rd).

Approximately 120 bees per colony were examined by the KOH method to determine the pre-treatment level of mite infestation. About three weeks after the initial treat-

ments, 20 live bees/colony were examined to determine the number of dead and live mites and population density of the 3 life stages of the mite. On both June 3 and 27, samples of 120 bees/colony were collected, processed by the KOH method and examined for tracheal mites.

After the first 3 weeks, the aerosol spray gave the best control of tracheal mites (93%, see table). At the same time, the red and white strips were less effective, providing 69% and 71% control, respectively. The ampule provided the poorest control, reducing the mite population by only 48%.

The aerosol spray also provided the best control both 44 and 68 days after the treatments were started. The infestation in untreated colonies dropped sharply from April 19 to June 27 (from 26.29% of the bees on April 19 to 21.33% on June 3 and 5.48% on June 27). During the same dates, the infestation in the aerosol spray group dropped sharply (from 21.25% to 2.92% and then to 0.67%). The white strips provided the second best control with relative infestations of 20.09%, 6.33%, and 0.83%. The percentages of bees infested in the red strip group dropped from 17.23% to 7.24% and finally to 1.54%. The ampule treatment was not very effective, as the infestations percentage dropped only about ⅓ more than the controls (from 20.95 to 18.53 and then to 3.75%).

Table – Effect of 4 formulations of Amitraz on populations of tracheal mites 3 weeks posttreatment in 2 apiaries near Norfolk, NE. May, 1988.

Treatment	No. of live mites/20 bees				% Control
	Eggs	Larvae	Adults	Total	
Aerosol	1.8a	2.1a	5.6a	9.5a	92.9
EVA Strip	7.2ab	12.5b	18.5b	38.2b	71.4
Cattle Band	10.3ab	12.7b	18.8b	41.8b	68.7
Ampule	19.9c	19.8b	30.3b	70.0c	47.6
Untreated	40.8d	40.6c	52.1c	133.5d	----

*Means followed by the same letter are not significantly different by ANOV and Duncan's new multiple range test.

17. Pettis, J. S.,[d] R. L. Cox,[d] and W. T. Wilson[d] – EFFICACY OF FLUVALINATE AGAINST THE HONEY BEE TRACHEAL MITE, *ACARAPIS WOODI*, UNDER LABORATORY CONDITIONS[u] – Fluvalinate is effective against *Varroa* as a contact acaricide, but has shown no control over the internal tracheal mite. Nevertheless we believed that fluvalinate would be effective externally against the dispersing female mites in search of young host bees.

In one experiment, random-aged adult tracheal mites were placed on either parafilm or fluvalinate impregnated plastic strips (Apistan®) (2.5% or 10%) in petri dishes held at 30°C. Four replications of 20 mites per treatment group were used. Mites were observed every half hour for mortality and the data analyzed by probit analysis. The LT_{50} of mites on parafilm (untreated) was 194 minutes (n = 56), while the LT_{50} of mites on the 2.5% and 10% strips was 77 (n = 64) and 68 (n = 67) minutes, respectively. The mites exposed to fluvalinate exhibited a significant decrease in LT_{50} ($P < 0.05$).

In a second experiment a fluvalinate strip (2.5% or 5%, 25 by 14 by 1 mm) was suspended in an 8 by 8 by 13 cm screen cage containing 96 ± 6 worker "host" bees (Gary & Page, *Exp. & Appl. Acarology* 3:291-305) infested with tracheal mites (100% of bees infested). Cages were held at 30 ± 1°C for 9 days and daily mortality of the "host" bees recorded. Newly emerged bees were marked on the abdomen with paint and introduced into cages on days 4 and 7 of the experiment, to serve as "targets" for the dispersing female *A. woodi*. On the tenth day live "target" bees were dissected and life stages of *A. woodi* recorded.

Controls and both treatments allowed *A. woodi* females to migrate to new hosts and lay eggs. However, no mites were found in the bees introduced on day 7 in the 5.0% cages (see table). Additionally, in the 2.5% and 5% cages a significant drop in "target" bees infested was observed between the 4 & 7 day introductions ($P < 0.05$, t-test). Henderson in Needham *et al.* eds., 1988, *Africanized Honey Bees & Bee Mites*, 380-386) showed that fluvalinate exhibited increasing toxicity to younger bees. We observed that the treated bees were more aggressive than the controls towards the introduced "target" bees. These two factors account for the lower recovery of "target" bees in the treatment groups.

Our test with live bees showed that exposure to fluvalinate strips did not prevent the transfer and reproduction of *A. woodi*. However partial control was demonstrated with the bees introduced on day 7 in both the 2.5% and 5% cages. A test of fluvalinate against dispersing *A. woodi* females in bee colonies is needed.

Table – honey bees ("target") were exposed to "host" bees (100% infested with tracheal mites) in cages containing fluvalinate strips (2.5% or 5%). "Host" bees were treated for ten days. "Target" bees (n = 10 bees/cage) were introduced on days 4 and 7 and dissected on day 10. Ten cages per treatment group (96 ± 6 "host" bees/cage) were used; all cages held at 30° C.

		Untreated	FLUVALINATE 2.5%	FLUVALINATE 5.0%
% mortality of "host" bees		5	19*	39*
% mortality of "target" bees	DAY 4	12	51*	75*
	DAY 7	14	53*	83*
% of "target" bees infested with *A. woodi*	DAY 4	16	22	16
	DAY 7	10	2*	0

*significantly different from preceding value ($P < 0.05$, t-test)

18. Reséndez B., J. J.,[m] W. L. Rubink,[d] G. Garza H.[m] – RUSTIC APICULTURAL PRACTICES IN THE MEXICAN STATE OF TAMAULIPAS – The state of Tamaulipas, especially the less agricultural central and southern regions, offers favorable conditions for apiculture. Approximately 73,000 managed honey-bee colonies exist. Of these, about 15,000 are "rustic," *i.e.* made from locally available materials and lacking any type of moveable frames. In at least one area, the central region between the cities of Ciudad Victoria and Soto La Marina, rustic colonies actually outnumber modern hives by a ratio greater than 2:1. The prevalence of this form of apicultural management in Tamaulipas, and the drastic changes likely to occur as the Africanized bee invades the state, led us to prepare this summary of rustic apiculture. We also report preliminary morphometric data from rustic bee colonies in central Tamaulipas.

Rustic hives are variously prepared from trunks of several species of palms or soft-wooded trees, wooden fruit crates, baskets, pottery, or other suitable materials. Locally, the rustic hives are termed "trabucos" or "bútanos." A hollowed log (ca. 1.2 by 0.3m), a common hive type, is fitted with carved wooden plates which are cemented with mud and manure onto one or both ends of the hollowed log. A small entrance hole is made in the hive cover. The hives are then covered with a cylindrical section of bark or sections of metal roofing to provide additional insulation and protection from rain. These hollowed logs are generally placed about 1m above the ground on a wooden framework and are generally located near human habitation (within 30m).

Initially all hives are populated from locally collected swarms, but after an apiary is established, the population is built from swarms issuing from the same apiary. A loose cloth covering is generally placed over the open entrance of the newly established colonies for a period of several days.

Most rustic apiculturists pursue beekeeping through family tradition, lack any formal training or concept of honeybee biology, and are primarily senior citizens living under

poor economic conditions. This has led to a great deal of folklore being associated with rustic apicultural practices.

Data from preliminary morphometric analyses demonstrated that the morphometrics of honey bees commonly cultivated in rustic hives are within the ranges reported by Daly and Balling *(J. Kans. Entomol. Soc.* 51:857-69) for bees of European ancestry. Mean forewing lengths of 10 samples from 5 apiaries ranged from 8.78 to 9.29 mm. The bees are easily managed; however, under certain weather conditions the bees have been known to become very defensive. Most rustic beekeepers use little or no protective gear in their operations, although they do use smoke.

19. Rinderer, T. E.[e] – THE REDISCOVERY OF *APIS KOSCHEVNIKOVI* – A fifth species of honey bee, *Apis koschevnikovi*, is found in northeast Borneo. This bee has a rufus body color that clearly provides field identification in Borneo. Morphologically, *A. koschevnikovi* is distinguished by a very high cubital index, a hairy fringe on the hind tibia of workers, and a unique endophallus. Behaviorally the drone flight time is separate from other species.

The defensive behavior of *A. koschevnikovi* is minimal. They can be handled without protection or smoke. Guard-bees inspect returning foragers and escort them into the nest.

Variations in enzymes exist between *A. koschevnikovi*, *A. cerana* and *A. dorsata*.

This newly understood species is a valuable germplasm resource. It may be a bridge for transferring genes between *A. cerana* and *A. mellifera*. It may also be a source of unique genes having scientific or commercial value.

20. Rivera, R.,[d] R. L. Cox,[d] and J. O. Moffett[d] – LONG-TERM MENTHOL RESIDUES IN HONEY[u] – This study was conducted to monitor menthol (5-methyl-2-(1-methyl-ethyl) cyclohexanol) residues in honey and wax in honey bee *(Apis mellifera)* colonies treated with menthol for control of the tracheal mite *(Acarapis woodi)* over a one year period. A petri dish containing 25 grams solid menthol was applied to the top bars of the frames of 32 of 48 tracheal-mite infested colonies in South Texas during the spring of 1987. Menthol was applied from April 18 to May 26 in South Texas. Colonies were then transported to Nebraska in May. Menthol was then reapplied to 16 of the 32 treated colonies. The menthol treatments were removed in July prior to the summer honey production season. Samples were taken on September 16, before menthol was again applied. The menthol treatments for the autumn were 0, 25, 50 grams per colony. The treatments were applied on September 17 and removed on November 12, 1987. Residues of menthol in honey and wax from the honey supers were determined for six sampling dates during the year. The honey and wax were separated, extracted with acetone and the extract analyzed by gas chromatography. Menthol residues in the honey are shown in the table. The menthol did not sublimate readily from the solid cakes in the autumn in Nebraska, even when placed above the warm winter cluster. Consequently, residues were low. In some cases menthol repelled the bees. There was less than 1 part per million residual menthol in the honey after one year of spring and autumn exposure. Menthol was absorbed into the wax more readily than into the honey and dissipated slowly from the wax. At the present time menthol has not been registered as an acaricide, therefore no minimum residue tolerance has been established.

MENTHOL RESIDUES (PPM) IN HONEY

TREATMENT	MAY	JULY	SEPT.	NOV.	FEB.	APR.
0	0.19	0.70	<0.10	<0.10	0.45	0.54
*LOW	0.55	2.27	0.31	<0.10	0.30	0.58
**HIGH	0.39	5.31	1.53	0.80	0.77	0.95

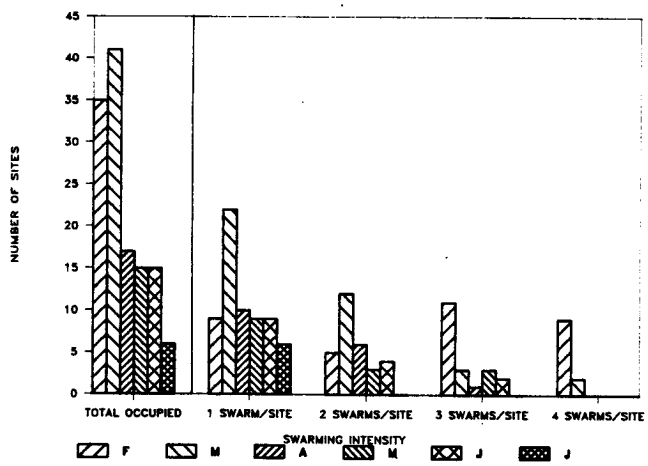
*Low treatment spring 25 grams menthol Apr. 18-May 26
Low treatment autumn 25 grams menthol Sep. 17-Nov. 12

**High treatment spring 25 grams menthol Apr. 18-Jul. 8
High treatment autumn 50 grams menthol Sep. 17-Nov. 12

21. Rubink, W. L.,[d] W. T. Wilson,[d] J. J. Resendez B.[m] and D. L. Maki[d] – BIOLOGICAL CHARACTERIZATION OF PRE-AFRICANIZED HONEY-BEE SWARMS IN NORTHEAST MEXICO AND SOUTH TEXAS – This report covers an ongoing study begun in February, 1988 to characterize honey bee swarming biology in subtropical areas of northeast Mexico and south Texas. Two transects of pulp-pot bait hives (Schmidt and Thoenes, *Amer. Bee J.* 127:435-38) were established: (1) a 200 km long Mexico transect, located in the state of Tamaulipas and extending from 25 km west of Cd. Victoria to the Gulf of Mexico, 50 km east of Soto La Marina; and (2) a 110 km Texas transect, located along the north side of the Rio Grande westward from Brownsville to 30 km west of McAllen. The Mexico transect is composed of 63 equidistantly (3 km) spaced stations each comprised of a cluster of 4 bait-hives. The Texas transect is made up of 23 equidistantly spaced (6 km) bait-hive stations, and 13 additional stations in the confines of Santa Ana National Wildlife Refuge and Bentsen-Rio Grande State Park. Bait hives were clustered at each station to provide a measure of local swarming intensity. A cluster consists of 2 sets of 2 back-to-back traps spaced approximately 50 meters apart, for a total of 4 traps per site.

Traps were baited with 1:1 citral:geraniol mixture in closed polyethylene (0.4 ml) centrifuge tubes and hung on shaded tree limbs about 3-4 m from the ground at the periphery of wooded areas, or in lone trees in open fields. Samples were collected monthly at all locations on both transects. All swarms were sacrificed; those not preserved in their entirety for further analysis were subsampled and discarded. On the Mexico transect, traps were also monitored for swarming activity midway between the monthly sampling periods.

Swarming activity was surprisingly high from the onset of the study. The only areas noticeably devoid of swarming activity were located at higher elevations (> 500 m) along the Mexico transect. In Mexico the total number of sites occupied by swarms (at least one swarm present) reached a peak in March and thereafter decreased steadily through July (see figure, "Total Occupied"). Local swarming intensity, measured by the fraction of traps filled per month at a given site, however, was highest in the February sampling (see figure), at 1.4 swarms per site (93 total swarms), and decreased to 0.1 swarms (6 total) per site in July, when there were no multiple catches at any trap site. A total of 233 swarms were captured in the initial 6 month period of the study. The Texas study, initiated one month later,



Relative swarming intensity in central Tamaulipas state, Mexico, during the six month period from February through July, 1988 as measured by bait-hive catches.

showed similar patterns, ranging from 1.9 swarms/site (68 total swarms) in March to 0.03 (1 swarm) in July. A total of 127 swarms were collected on the Texas transect in a 5 month period.

Swarm population size and drone fraction of 101 of the collected swarms were similar in the Texas and Mexico samples. Thirteen of 30 Texas swarms contained drones at levels as high as 1.5%, while 32 of 71 Mexican swarms contained drones at a maximum level of 2.0%. Swarm size distributions were similar, but much lower than that expected: Texas swarms ranged from 1,600-11,500 (mean = 4700) bees/swarm and Mexican swarms from 1,000-11,000 (mean = 4500) bees per swarm.

Preliminary analyses showed no indications of *Varroa jacobsoni* in alcohol washes of 1000 bee aliquots from each swarm. *Acarapis woodi* infestations were found in 33% of 52 swarms analyzed. *A. woodi* infestations generally decreased into the summer and ranged from 2 to 90% of the worker bees per swarm. Drones were found to be infested with *A. woodi* at the same level as the worker bees.

Field analyses of defensive behavior by bees in Mexico bait-hives gave mean stings per target (two 5 by 5 cm suede leather patches) of 15.9 ± 9.4, not significantly different ($p > 0.06$) from that found for Texas traps, 4.9 ± 7.2, and not suggestive of any degree of Africanization. Cell measurements from newly constructed comb ranged from 5.1-5.5 mm. Morphometric analyses gave measurements within the ranges reported for European bees by Daly and Balling (*J. Kans. Entomol. Soc.* 51:857-69) and gave exclusively European identifications using 25-character discriminant analysis.

The present study points to wide variability in the swarming behavior of European origin *Apis mellifera* in subtropical climates, especially with regard to swarm frequency and average swarm size. Evidently environmental factors play a greater role than has been previously shown in this aspect of their biology. It is hoped that this study will help to allay some misconceptions about comparative European and Africanized bee biology and contribute to a better understanding of the Africanization process.

22. Schmidt, J. O.[c] — SWARM TRAPPING — For millennia people have been trapping honey bee swarms with various cavity-containing devices. Nevertheless, as recently as the mid 1980's no substantial improvements over the primitive methods of practical swarm trapping had occurred. The increased problems of bee diseases in North America, the presence of northward moving Africanized honey bees in Central America, and the general high cost to the beekeeper of purchasing package bees and queens indicated need for a practical, durable, inexpensive, and effective swarm trap. This report describes the results of our recent development and scientific testing of a practical swarm trap.

In earlier reports (Schmidt and Thoenes, *Amer. Bee J.* 127:435-38 and *Bull. Entomol. Soc. Amer.* 33:155-58) we described the first economical and durable swarm traps that were truly effective in attracting and capturing swarms. Nevertheless, because they were labor intensive to assemble and contained expensive and potentially degradable plywood, these first swarm trap models were not as practical or economical as we desired. Both of those shortcomings have been eliminated in the swarm traps developed and used in this study. These new traps, made entirely of reinforced wood pulp, consist of two pieces that fit together without glue, wires or caulking to form an easily opened and inspected swarm trap of 31 l (approximately 9 gallons) internal volume. A crucial feature of the trap is the addition of a synthetic Nasonov pheromone attractant consisting of an equal mixture of citral, geraniol, and nerolic + geranic acids in a slow-release lure (details of trap in Schmidt and Thoenes, *Amer. Bee J.*, in press).

The effectiveness of these pulp-based devices in trapping swarms was compared directly in choice tests with the "pinata" traps that are used extensively in Mexico which consist of a thin-walled cardboard box covered with a colorful plastic bag. In addition to the two types of swarm traps, the competitive value of the pheromone, of beeswax, and of reinforced entrances were tested. The tests consisted of replicating, in 22 separate locations within the Tucson, AZ basin, sets of five swarm traps all placed in the one tree at the same height (3 m). One trap consisted of the piñata trap to which we added a pheromone lure; the other four were pulp traps with various modifications: one had a pheromone lure, beeswax applied to the inside, and a reinforced entrance hole; one had a pheromone lure and applied beeswax; one had a pheromone lure only; and the last was an unmodified trap without a pheromone lure. The swarms captured by each of these models were: piñata trap — 0 swarms (0% trap catch); pulp trap without modification or pheromone — 0 swarms (0% trap catch); pulp trap with pheromone — 14 swarms (64% trap catch); pulp trap with pheromone and beeswax — 11 swarms (50% trap catch); pulp traps with pheromone and beeswax and reinforced entrance — 11 swarms (50% trap catch). When the results are combined, we observe that all swarms selected traps with pheromone lures and traps of the pulp construction. These results clearly indicate: 1) that the pheromone formulation is an effective swarm attractant; 2) that beeswax and/or reinforced entrances do not enhance the effectiveness of the pheromone; and 3) that piñata traps, even with pheromone, are considerably inferior to pulp traps in their ability to attract swarms.

These results, reinforced by our other experiments, and those of Dr. William Rubink (personal communication), indicate that swarm traps constructed of reinforced wood pulp and containing slow-release pheromone lures are the best choice for effectively attracting and capturing honey bee swarms.

23. Smith, R. K.[n] and O. R. Taylor, Jr.[o] — DETERMINATION OF AFRICANIZATION IN HONEY-BEE QUEENS — Analysis of the extracted unsaturated hydrocarbons from worker honey bees has been demonstrated to be a sensitive and accurate method for detecting the presence of Africanization (Smith in Needham *et. al.* eds, 1988, *Afr. Honey Bees & Bee Mites*, 275-280; *Amer. Bee J.* 128:329-30; *Amer. Bee J.* 128: in press, Smith & Lavine, *Amer. Bee J.* 127:851). Extension of the method to the analysis of queens is complicated by the presence of additional unsaturated compounds. These queen-specific hydrocarbons are the N-15 odd homologous mono-olefins (C23 to C37 and above) and two additional series of diunsaturated compounds. In European worker bees, the unsaturated hydrocarbons are evenly distributed over the whole bee, however European queens display a distribution pattern depending on the olefin. The worker compounds, in queens, are also distributed evenly over the bee. Extraction of the isolated head, thorax and dorso/ventral abdomenal sectionings from fresh European queens show the queen specific hydrocarbons to be concentrated on the dorsal part of the abdomen, with only trace amounts to be found on the head and thorax. Another complicating factor is the seeming absence of the queen hydrocarbons in African queens and the non-predictable presence of the compounds in Africanized queens.

Average olefin abundances for European and Africanized queens were prepared which were normalized on the entire unsaturated extract. Fourteen compounds were identified which displayed adequately separated normalized abundances at the one standard deviation width. Using these 14 key compounds, a cluster separation map was prepared for the whole database (66 Africanized and 214 Europeans). An incomplete separation was obtained with only

94% correct classification.

An abundance profile for the unsaturated compounds indicated the worker compounds are identifically present (r = 0.998) in European queens and workers, while the presence of the queen hydrocarbons is under independent control.

Re-analysis of the queen database using only worker compounds for normalization indicated 10 compounds to have satisfactory separation of the means at the one standard deviation width. A cluster separations map of the database using the 10 compounds now resulted in complete separation of the Africanized and European queens, with 99.3% correct classification (Smith *et al., Amer. Bee J.* 128:676-678). A blind test with 100 queens was conducted using this analysis, resulting in 100% correct classification.

The missed classifications (2) in the best computer model developed to date, are correctly classified on visual evaluation of the chromatogram. This phenomenon of the eye/brain forming a better discriminating instrument than any computerized evaluation, has been noted time and again throughout the over three-year course of this research. In most cases, a classification of the bee (worker or queen) is possible prior to any data being entered in the computer.

The finding that the worker unsaturated hydrocarbons hold the key to Africanization detection in queens, coupled with the even distribution of these compounds over the body of the queen and the roughly doubled amount of the compounds extracted from queens over that extracted from workers, has been successfully exploited. Captured queens are killed, the abdomens removed and dried for hydrocarbons assay, while the head and thorax are preserved on Dry Ice for subsequent enzyme assay. This procedure results in a dual analysis on the same specimen using the two most accurate Africanization detection methods currently available.

24. Taylor, O. R. Jr.,° M. Long° and G. A. Rowell° — GENETIC DIFFERENCES BETWEEN EUROPEAN AND AFRICAN BEES IN MEXICO — As part of an effort to track the spread of the African bee genome through Mexico and to characterize and quantify factors leading to genetic changes in managed apiaries, we established allele frequencies for two marker allozyme loci, malate dehydrogenase and hexokinase, for both feral and managed bee populations. A literature review of allele frequencies, as well as new data for these loci, show that as African (feral) populations are isolated in space and time from managed (European) populations, the frequency for the 1 (fast) allele for MDH increases from a low of .6 to approximately .9, while the 2 (slow) allele for HK increases from <.3 to .6. Due to matings by European queens with African drones, the frequencies of these alleles also increase in managed apiaries. Comparisons of observed and expected genotypic frequencies of queens show that European bee populations in Mexico are at or near Hardy-Weinberg equilibrium before the arrival of African bees, while invading feral (African) populations show an excess of the 1 (fast) allele for MDH and the 2 (slow) allele for HK. This excess is most easily explained by differential reproduction (swarming) and dispersal (long distance movement by swarms) among feral colonies representing differing degrees of hybridization with European bees. These studies, and those of G. Hall (see abstract No. 10) and D. Smith (manuscript), lead to the following conclusions concerning the genetic attributes of the feral (African) and managed (European) honey bee populations in Mexico.

1. In a broad sense of the term, feral African bees are hybrids.

2. However, as a population, they retain little if any European mtDNA, and nuclear DNA, acquired through matings with European drones, is rapidly lost through selection and dilution. Therefore, emphasis on a hybrid origin and current hybrid nature of these bees is misleading since it implies that the population contains substantial components of the European genome.

3. The feral population does not represent a "hybrid swarm" containing both parental types and a continuum of intermediates. Rather, it is a rapidly reproducing and self-sustaining population which retains and/or returns to phenotypic and genotypic frequencies that are similar to those of bees from Africa.

4. In contrast, managed European populations and the "Africanized" European mother lines contribute little to the feral population and are ultimately lost through attrition.

5. Therefore, the managed population is not a base for the buildup of the feral population and apiaries are not stepping stones for the spread of the African population.

6. The lack of mtDNA from European bees and the rapid loss of European nuclear DNA from the feral population means that African bees cannot be permanently Europeanized, except under conditions of extremely low density and low vagility where they are numerically inferior to a hyperdispersed managed and feral European population. Such conditions are most likely to be found close to the geographic limits of the feral African populations. It follows that, in spite of some hybridization with European populations, African bees will arrive in the United States essentially unchanged.

7. Because of the tendency for the African population to maintain its genetic integrity, it seems appropriate to recognize this population as a distinct biological unit.

8. Continued use of the term "Africanized" to embrace both the feral population and the "Africanized" European mother lines of apiaries obscures the fact that "Europeanization" of the feral population and "Africanization" of the European population are two different processes with strikingly different outcomes.

9. Therefore, it seems appropriate to refer to the feral population as "African derived" or "neotropical African," albeit with some European influence, and to restrict the use of the term "Africanized" to cases where European mother lines have mated with African drones.

10. Such a distinction will help us focus on the dynamics of both processes and, in particular, draw attention to the genetic basis of adaptations which give the neotropical African bee such clear advantages over their European counterparts in the neotropics.

25. Taylor, O. R., Jr.,° G. A. Rowell° and M. Spivak° — RATE OF SPREAD AND RELATIVE ABUNDANCE OF AFRICAN HONEY BEES IN MEXICO — In cooperation with the Mexican Programa Para Control de Abejas Africanas, we are developing models based on climate and honey bee reproductive biology to predict the rate of spread and relative density of the feral African bee populations throughout Mexico. The models are being assessed by extensive sampling behind and in advance of the known front of the African bee population. African bees are identified in the field by the size of worker cells, behavior and acoustical signals. In the laboratory morphometrics (FABIS, etc.), hydrocarbons (K. Smith), electrophoresis (O. Taylor), mitochondrial DNA (D. Smith, W. Sheppard), DNA fragment analysis (G. Hall), are being used to analyze critical samples.

Presently we are developing a computerized data base for the climate, vegetation, and distribution and abundance of managed European bees throughout Mexico. This information will be supplemented with data on the seasons of swarming, honey production and dearth associated with particular climates, seasons and vegetation types.

Two models are being tested. The Swarming/Absconding Model is based on the number of months per year in each

area that are favorable for swarming or absconding. In the past, African bees have often advanced 30 (50 km) miles per month in favorable (swarming) months and 15 (25 km) miles or less per month in unfavorable periods. Climate Model I integrates information on the distribution of rainfall throughout the year, total rainfall, and monthly and yearly mean high temperatures. (The temperature data accounts for changes in elevation.)

These models are being tested in two ways. First, they are being compared with past rates of spread and relative densities. The degree of success in predicting past events should indicate the utility of a model in predicting future rates of spread and density. However, since past events are not always indicative of future developments, the models are also being tested by extensive sampling behind and in front of the known African bee populations.

A preliminary assessment of the models, and the pattern of movement of African bees in Mexico to date, suggests that feral African bees will reach the Brownsville-McAllen area of Texas between November 1989 and March 1990, with the later date being more likely. Less information is available on the movement of African bees along the Pacific coast, but overall it appears that the habitats in this region are less favorable for the development of high density populations and rapid spread of the feral bees. Because of lower quality habitats and a longer route, it should take African bees at least three years longer (1993) to reach Arizona.

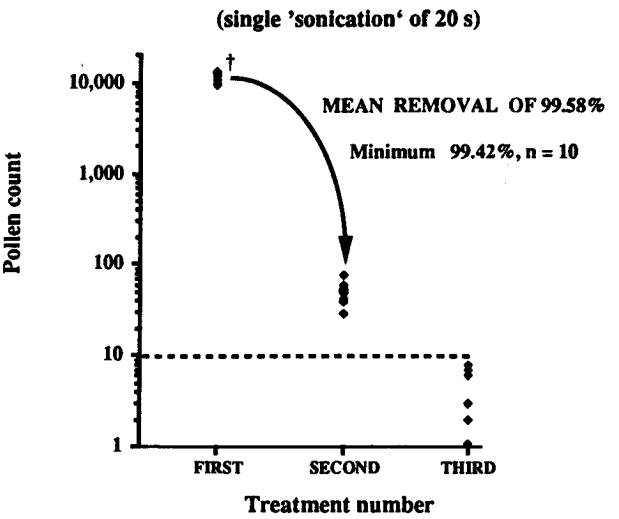
In contrast to our previous experience, African bees in Mexico appear to be moving more rapidly in the wet (April-October) than the dry season (November-March). This may be due to the strong southeasterly winds that sweep along the Atlantic coast during the wet season. African bees advanced approximately 300 miles through the Atlantic lowlands during the 1987 wet season (>40 miles/month). However, the rate of spread diminished somewhat (<20 miles/month) from October 1987 to March 1988, perhaps due to the strong northeasterly winds along the coast. Again, as in 1987, African bees have spread rapidly along both coasts during the 1988 wet season. At this time, it appears that a swarming and absconding model together with an assessment of habitat quality (*i.e.*, vegetation type) will be more useful than a model based on physical factors alone.

26. Vaissiere, B. E.[p] — A NOVEL METHOD FOR QUANTIFICATION OF POLLEN PRODUCTION AND POLLEN LOADS ON BEES AND STIGMAS USING A COULTER® COUNTER — Numerous approaches have been used in the past to evaluate the number of pollen grains on various substrates, but these lacked general applicability, and their accuracy and reliability were seldom measured. This abstract reports the development of a rapid and accurate technique to quantify the pollen production of male-fertile flowers, the pollen carry-over of pollinators, and the stigmatic pollen loads of male-sterile flowers. Such a technique will greatly enhance the study of crop pollination for hybrid seed production.

Upland cotton, *Gossypium hirsutum* L., produces an abundance of large echinate pollen grains which adhere readily to insect visitors and the large stigma. Honey bees are important pollinators of this crop for hybrid seed production, and male-sterile lines are readily available. Using Upland cotton, a two-step method was developed to (1) remove pollen grains from an anther, bee, or stigma substrate and (2) obtain fast and reliable pollen counts.

Pollen removal was achieved by cavitation in a 1% saline using a 200 W Sonicator® with a 12.7 mm horn vibrating at 20 kHz. This ultrasonic treatment also served to (1) achieve a uniform suspension of monodispersed pollen grains with a narrow size distribution, (2) stop pollen germination, and (3) disintegrate background particles, especially stigmatic papillae, below the size range of pollen grains.

Efficiency of ultrasonic treatment for cotton pollen removal from honey bees



† maximum carrying capacity (honey bees spun for 5 minutes at 150 rpm in an excess of dry Upland cotton pollen)

Pollen grains were counted with an electronic sensing-zone particle counter (ZM Coulter® counter) in which particle volume is the basis of measurement. Pure hand-harvested cotton pollen in a 1% NaCl electrolyte solution was used to calibrate size thresholds. By focusing the suspension of particles to be counted into the aperture probe of the counter (focused-flow method), high count accuracy was achieved. Blind tests conducted with 0 to 50 cotton pollen grains added to clean electrolyte repeatedly gave a very close fit between visual pollen counts and the ZM counts ($r^2 = 0.978, n = 23$, maximum deviation = 5).

By modifying the ultrasonic treatment, efficient pollen removal was achieved for male-fertile flowers, pollen carry-over of pollinators (see figure), and stigmatic pollen loads. The linearity of the counts was established up to 120,000 pollen grains, the largest amount encountered, by the addition of a known weight of cotton pollen to a clean specimen (e.g., for honey bees with 0.5 to 7.5 mg of cotton pollen added, $r^2 = 0.996, n = 6$). The calibration curve also provided an easy way to estimate the mass of pollen produced by an individual flower.

Overall, an accuracy of ± 5 pollen grains per male-sterile stigma, ± 10 grains per bee, and ± 50 grains per flower pollen production was achieved in 4 to 8 minutes. Also this technique can be extended to other plant species regardless of their pollen morphology because volume of pollen grains, rather than their projected surface area, is the basis for measurement.

27. Villa, J. D.[e] and J. M. Labougle[l] — RANGE EXPANSION OF AFRICANIZED BEES IN MEXICO — Pheromone-baited cardboard boxes and morphometric identification techniques have been used to monitor the range expansion of Africanized bees in southern Mexico since 1986. Contrary to previous expectations, the average yearly rate of spread along the drier Pacific coast has been slower (350 kilometers) than along the Atlantic coast (420 kilometers). Movement of the "advancing front" per 3-month period suggests that nectar flows can reduce range expansion to 30 km per period, while dearths can increase it to 210 km per period. Indices of capture rate (swarms/[bait hives x months]) show that the "advancing front" is dilute and may be difficult to detect. If movement along the Gulf coast con-

tinues at the same rate, the first swarms of Africanized bees should be detected in Brownsville in March of 1990.

Table – Rates of movement of Africanized bees in Mexico along the Pacific and Atlantic coasts. Advance per 3-month period was estimated by measuring the distance between the most advanced detections of AHB at the beginning and end of the period. Yearly rates were calculated by averaging all combinations of 4 consecutive quarters (n = number of combinations).

RANGE EXPANSION (kilometers)					
	QUARTERLY RATE				AVERAGE
	Dec-Feb	Mar-May	Jun-Aug	Sep-Nov	YEARLY RATE
PACIFIC COAST					352 (n = 4)
1987.....	90	210	70	30	
1988.....	100	120	30	?	
ATLANTIC COAST					420 (n = 2)
1987.....	–	–	140	150	
1988.....	60	110	60	?	

28. Waller, G. D.,[c] R. A. Hoopingarner,[q] J. H. Martin,[c] G. M. Loper,[c] and M. M. Fierro[r] – CONTROLLED NATURAL MATING OF HONEY-BEE QUEENS DURING WINTER IN SOUTHERN ARIZONA – Honey-bee queens naturally mate while flying, encountering drones from colonies located up to 10 miles away (Peer, *J. Econ. Entomol.* 49:254-255). Controlled natural matings have been difficult to obtain. This study was conducted to determine whether controlled natural matings could be accomplished by moving drone-rearing colonies and mating nucs to a desert area during the winter. Colonies in such areas lack drone populations during the winter dearth of pollen and nectar.

This study was conducted in the Altar Valley about 25 miles southwest of Tucson during September 1987 through March 1988. The rare cordovan mutant was utilized as a genetic marker for both the queen-mother colony and the drone-mother colonies. Drone-mother colonies with naturally mated cordovan queens were moved to Tucson in late November, then to the Altar Valley in late January. Pollen cakes, placed in the colonies during December and January, supplemented incoming floral pollen to stimulate drone rearing.

After a 6-week period of no mating during late December and all of January, we began our controlled mating tests. The presence and color of drones flying in the experimental area were determined periodically during February and March by catching flying drones in a Taylor drone trap suspended from a helium-filled weather balloon (Taylor, *J. Apic. Res.* 23:18-20).

A single cordovan queen that had naturally mated with about 50% cordovan drones served as the queen mother for cordovan virgins. Groups of 5-frame, droneless nucleus colonies, each with a caged virgin cordovan queen, were moved to the Altar Valley every two weeks from late January through March. After a two-week mating period, these colonies were returned to Tucson for worker progeny testing of percent cordovan. Drone populations in the drone-rearing colonies were determined in early March from photographs of both sides of each frame.

Drone trapping in the area during February showed 98% cordovan drones flying mostly between 1:00-3:00 p.m. During March this percentage dropped to 60% and then to 56%, even though we counted more than 1300 cordovan drones in the drone-rearing colonies in early March. Virgin queens that emerged on January 18 or February 1 mated 100% to cordovan drones. Those from February 15 mated 78% to cordovan, after which the percentage dropped to 13% (February 29) and 4% (March 14). The disparity between these relatively high values for cordovan drone percentages in flight and the low percentage of cordovan progeny resulting from our March matings may have been the result of a mating advantage for non-cordovan, wild-type drones or because the virgin cordovan queens flew farther from the experimental apiary before mating than did the cordovan drones (Taylor and Rowell, in Needham *et al.* eds., 1988, *Afr. Honey Bees & Bee Mites*, 173-183.).

These results provide encouragement for beekeepers who fear the advent of the Africanized bees. Survival of a viable bee industry may depend on development of a system of controlled natural mating that excludes all or most Africanized drones from mating. Our results provide a method for defining and testing the time when controlled natural matings can occur in a given area. Off-season mating may allow beekeepers to selectively mate enough queens for their entire operation prior to the early spring build-up of colony populations each year.

29. Williams, J. L.,[e] R. G. Danka,[e] and T. E. Rinderer[e] – SELECTIVE BAIT SYSTEM FOR POTENTIAL ABATEMENT OF FERAL AFRICANIZED HONEY BEES[u] – The efficacy of acephate collected by foragers was assessed as a selective means of eradicating undesirable honey-bee populations. Test colonies consisted of single story 10-frame Langstroth hives (16.6-cm body), ca. 20,000 adult bees, a laying queen, all stages of brood, and stored honey and pollen. From each of the isolated colonies, foragers were trained to a 50% (vol:vol) sucrose syrup plus 10% honey (vol:vol) bait placed 10-m from the hive. Bees collected syrup *ad libitum* from 50 1-mm holes near the rim of an inverted feeder can. Untreated syrup was replaced with syrup containing 250 ppm (mg/liter) acephate during active foraging; bees continued to collect treated syrup for ca. 30 min. Bait stations were carefully monitored to verify that foragers came only from target colonies.

Colonies that collected an average of ca. 70 ml of 250 ppm acephate-syrup bait lost most of their adult workers and all larvae within one week. Queens in 13 of 19 colonies died without replacement within 3 days of treatment (see table). Two queens died, but were successfully replaced, and 4 queens remained alive and laying. Subsequent tests using 10-m or 100-m foraging distances have shown that colonies treated with 500 ppm or 750 ppm acephate-syrup often lost their queen within 1 or 2 days and undergo more rapid worker mortality. The selective bait system has recently been used to kill 3 managed colonies of Africanized honey bees (AHB). Typical acute responses were observed after the 3 AHB colonies collected acephate-syrup (95 ml of 250 ppm, 106 ml of 500 ppm, or 75 ml of 1000 ppm acephate-syrup/colony) from baits at 10 meters.

The baiting system shows potential for use in control programs for Africanized honey bees because it is efficacious, relatively safe, and due to the brief, managed exposure, is selective for honey bees.

Response of honey bee colonies which collected syrup-honey bait containing 250 ppm acephate. Tests conducted at Baton Rouge and St. Gabriel, Louisiana; April 1988. Data are $\bar{x} \pm$ sd (n).

Queen fate	n[a]	Max. no. of foragers on one feeder during tmt	Syrup collected (ml)	24-h worker mortality
Dead.....	13	147 ± 67	74 ± 32 (10)	1827 ± 1090 (12)
Replaced.....	2	127 ± 68	30 (1)	1105 ± 1370 (2)
Normal.....	4	34 ± 20	48 ± 43 (3)	586 ± 532 (4)
Control.....	2	129 ± 100	550 ± 637 (2)	34 ± 47 (2)

[a]Nineteen colonies were treated with acephate; two served as controls.

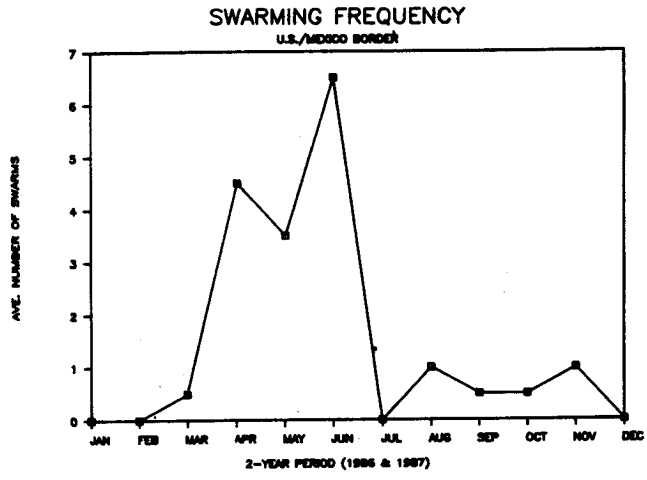
30. Wilson, W. T.,[d] D. L. Maki,[d] J. Vargas C.,[h] R. L. Cox,[d] and W. L. Rubink[d] – HONEY BEE SWARM FREQUENCY AND TRACHEAL MITE INFESTATION LEVELS IN BAIT HIVES ALONG THE US/MEXICO BORDER – A transect of 25 honey-bee bait hives (swarm traps) was established along the south side of the US/Mexico border in close proximity to the Rio Grande River in November of 1985. The purpose was to determine the prevalence

and frequency of honey-bee *(Apis mellifera)* swarms along the border, and further, to assess the tracheal-mite *(Acarapis woodi)* load in these bees. The Rio Grande presents no obstacle to swarm movement since it averages less than *ca.* 100 meters in width in the area of the transect. The transect extended from Nuevo Progreso to Matamoros, (Tamaulipas) Mexico, a distance of approximately 60 km. The wooden bait hives (48 by 16 by 24 cm.) were painted yellow and contained a pheromone attractant (Citral: Geraniol, 1:1). Each trap was placed 1.5 meters above the ground on steel fence posts approximately every 2 to 3 km. along the transect, and checked for swarms each month. When a bait hive became occupied, an empty bait hive was added in close proximity to assure bait hive availability.

During the period from January 1986 through December of 1987, 36 swarms were captured (see figure for yearly average). From January to March 1988, three additional swarms were collected. Eighty percent of the total number of swarms captured occurred from April through June (April 25%, May 19%, and June 36%) which coincides with the period of greatest brood rearing and nectar/pollen availability. No swarms were captured in July, a period of environmental stress when plants dry up from high ambient temperatures and a lack of soil moisture. From August through November, autumn plants bloomed and swarm frequency increased to 17%, but declined to 3% in the period from December to March.

Subsamples of honey-bee swarms were examined for *A. woodi* infestation by removing and examining the prothoracic tracheae of individual bees. Tracheal mite infestation rates varied greatly in the subsamples (total of 700 worker bees) examined. Overall, mites were present in 11% of the bees examined. Of the 78 infested bees, an average of 1.75 tracheae per bee was infested, 67% of which were heavy infestations (more than 30 mites per infested trachea), 22% moderate (10 to 20 mites), and 11% light infestations (fewer than 10 mites). Contrasting sharply with the 11% bee-infestation rate were samples taken from 7 rustic colonies at one location near the transect. In these samples 43% of the total number of bees (106) were infested, averaging 1.9 tracheae infested per bee. Eighty-nine percent of the infested tracheae had heavy infestations. Dispersing swarms have the faculty to spread the tracheal mite into new geographic areas.

Discriminant analysis of 25 morphological characters were employed on 4 swarm samples of 10 bees each sampled from the transect using the methods of Daly and Balling (*J. Kan. Entomol.* 51:857-869). All morphometric measurements fell within the range for bees of European ancestry.



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