Proceedings of the American Bee Research Conference

The 1991 American Bee Research Conference was held on October 7 and 8 in Tucson, Arizona. Meetings were in the Meats Auditorium at the Campus Agricultural Center of the University of Arizona. The seventh American Bee Research Conference will be held in College Station, Texas on September 21 and 22, 1992. The following are abstracts from the 1991 conference.

1. Buchmann, S.L.[a] and C.W. Shipman[a] — FORAGING DISTANCES FLOWN BY HONEY BEE COLONIES: ANALYSES USING MATHEMATICA SOFTWARE[ff] — Managed and feral honey bee colonies in southern AZ forage widely for raw materials (pollen, nectar, water, propolis) during a 10-11 month flight season. A typical colony annually collects 40-60 kg of pollen from >50 flowering plants species up to 10 km from the nest.

If 10,000 to 25,000 foragers, each making 4-8 daily trips, visit 5-10 distinct locations at various distances, then the daily pattern of forager allocation can be likened to a giant amoeba with pseudopodia exploiting ever-changing rich floral patches. An omniscient observer plotting the seasonal, annual or multiyear forager distribution, would discover a very different pattern from that detected during an experiment of only a few days duration. Now we see that few bees forage closeby or at extreme distances, whereas the great majority visit sites at intermediate distances. The figure depicts an annual or multiyear mathematical Gaussian (normal) distribution of resource sites utilized, the 3-D color graphic produced with Mathematica software (Wolfram Research, Inc.). In this cutaway view the hive is located at 0,0 and the maximum number of long-term sites used are about 2 km away. In this quantitative model we would expect to find 3.4% of the foragers within a 100 m wide band centered 1.8 km from the hive.

A large, forgotten empirical data set (D.F. Peer, unpubl. Ph. D. dissertation, 1955) was re-analyzed using Mathematica. Peer placed color mutant (Cordovan) Italian bees in apiaries of 1, 20 and 50 colonies near Madison, Wisconsin, then scored the number of Cordovan foragers seen in all directions up to 5.0 km away during the summer months over a two year period. He censused 215 locations and recorded 16,575 bees foraging on 14 plant species in this mixed pasture woodland environment. This provides an ideal data base to examine the average distribution of foragers from an apiary. The mathematical distribution closely matched our hypothetical case and indicated that most foragers accumulated at 2.0 km from these apiaries.

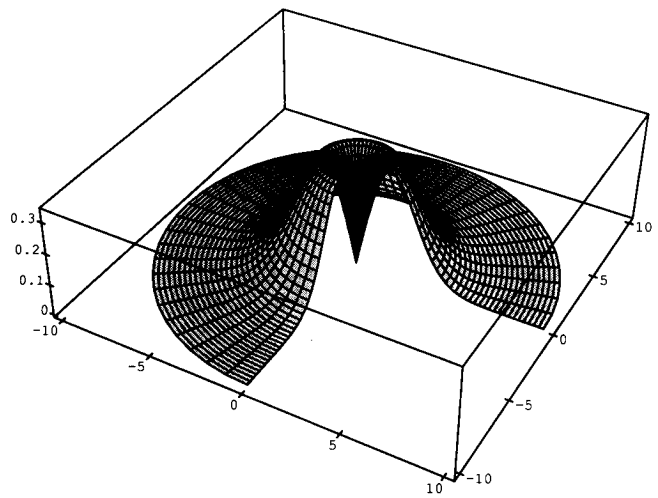


Figure - A hypothetical annual foraged sites distribution from one *Apis mellifera* colony.

This type of graphical analysis has great potential for directing the placement of apiaries for commercial pollination and to help determine risks from insecticidal applications.

2. Buchmann, S.L.[a] and C.W. Shipman[a] — SEASONALITY OF DEBRIS PRODUCTION BY HONEY BEE COLONIES IN ARIZONA — A full-sized colony of *Apis m. ligustica* housed in Langstroth equipment and given minimal management was studied from March, 1988 until June 7, 1991. The study site was located near the mouth of Pima Canyon in the Santa Catalina Mountains north of Tucson, Arizona. The colony was established on comb foundation and in deep frames above and separated from an empty lowermost deep super by a wire screen. Colony debris, but not trash carried out the normal entrance, fell into an OAC pollen trap drawer where it was collected, weighed to the nearest mg and sorted on a weekly basis. House bees could not crawl through the screen mesh to recover debris, so this method is similar to the natural debris rain that accumulates below feral colonies occupying protected rock cavities in the Sonoran desert, and can be used to estimate the age of such debris middens.

Debris consisted of dead bees (rare), dropped wax scales and reworked wax, corbicular pollen, bee parts, cocoons, larval feces, wax moth feces and other unidentified materials. The amount of debris produced varied seasonally and was not homogeneous in composition. Weekly periods of greatest production (as high as 28 g) occurred in the spring months of February and March during intense brood rearing prior to the swarming season. During 1989 the colony produced 132.9 g of total debris materials, of which 7.4 g was corbicular pollen (1502 pellets) and only 207 dead bees. The mean weekly fresh weight of dropped materials was 2.9 ± 2.2 (SD) grams. During the entire 158 week study, the colony dropped a total of 1,002.5 g, of which 45.7 g (4.6%) was dropped pollen. Thus, the colony averaged 6.3 ± 5.7 (SD) g of debris over the entire period from 1988 until 1991.

Thus, on average, a full-sized colony in the Sonoran desert produced 334.2 g of debris on an annual basis. This value can then be used to estimate the approximate ages of old blackened debris middens found below feral colonies living in the Sonoran desert of Arizona (Buchmann, unpubl. data).

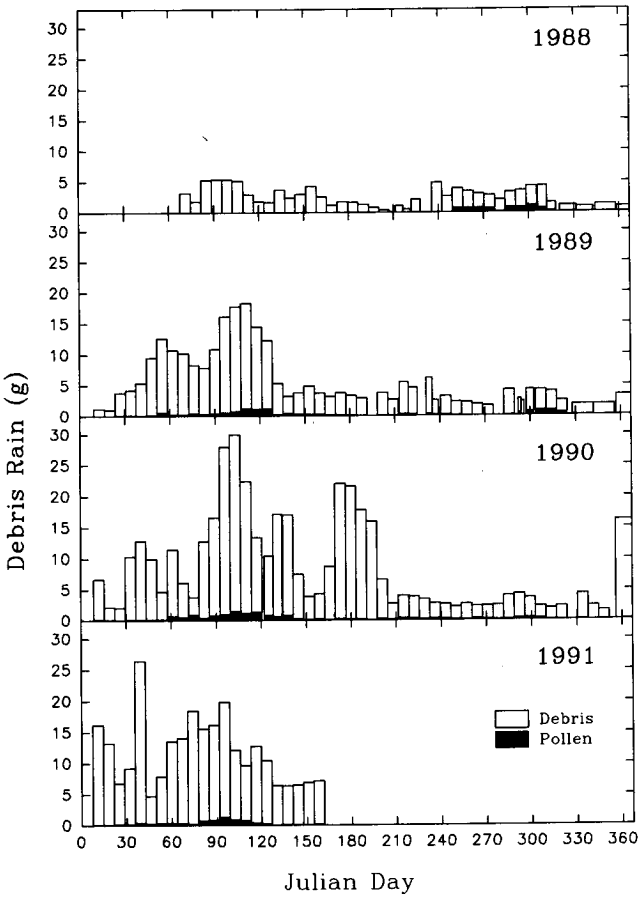


Figure - Histogram of weekly debris production (g) from March, 1988 until June, 1991. The open bars represent the total debris fresh weight, while the black bars give the weight of the corbicular pollen fraction.

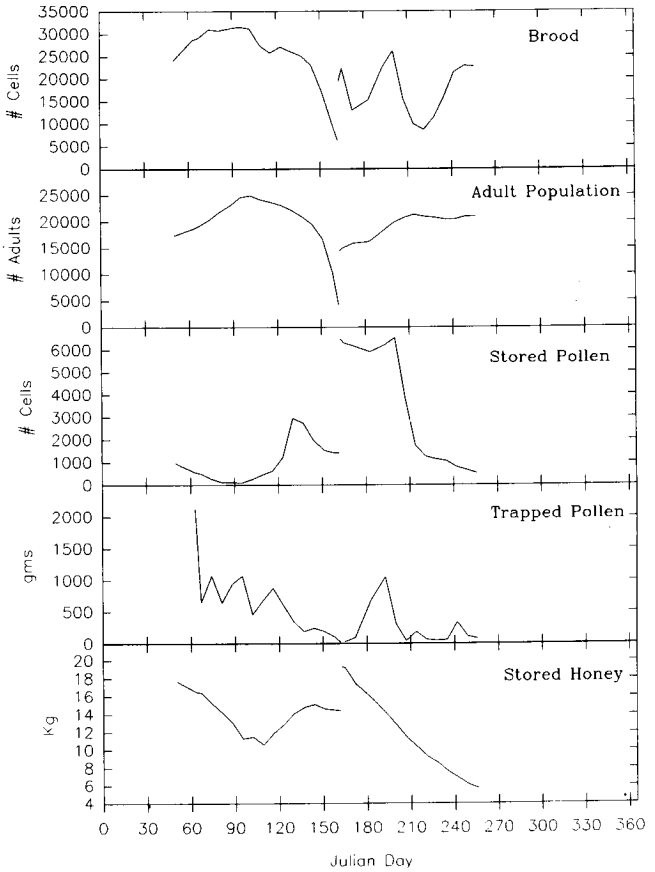


Figure - Seasonal phenology of brood, adult population, harvested and stored pollen, honey and adult population in two standard colonies of Italian honey bees in the Sonoran desert.

3. Buchmann, S.L.[a] and C.W. Shipman,[a] and S.J. Prchal — PHENOLOGY OF POLLEN STORAGE WITHIN HONEY BEE COLONIES LIVING IN THE SONORAN DESERT OF ARIZONA — A full-sized Langstroth colony of *A.m. ligustica* honey bees given minimal management was censused daily and weekly to delimit the phenology of levels of brood, adult population, stored beebread and honey and incoming trapped pollen. The colony was located at the Arthropod Discovery Center (operated by Sonoran Arthropod Studies, Inc.) on the west-facing bajada of the Tucson Mountains (Lat 32° 14' 19.9" N; ln 111° 7' 30.9" W). Our observations began on February 19, 1991 and are continuing. We report data for two colonies at this location from February 19 - June 9 and from June 11 - September 27, 1991. The colonies were located under a shade screen resting upon a Sartorius F330S electronic scale and fitted with a modified OAC pollen trap (Smith & Adie, 1963, *Can. Bee J.* 74:4, 5, 8). Pollen was collected daily and weighed fresh, while the adult population and cm^2 of brood, stored pollen and honey were censused every Friday using an overlaid grid.

No previous studies have correlated the amount of pollen harvested with that temporally stored as beebread. Honey bees do not hoard pollen as they do for honey, and we determined that stored pollen levels did fluctuate on a seasonal basis that was correlated with other colony parameters. The mean amount of trapped pollen for this 34 week period was 70.8 ± 72.6 (SD) g. Stored pollen fluctuated widely from 26 to 1607 cm (mean of 485 ± 542 (SD). The first colony failed and had to be replaced on June 7. The second colony consumed honey rapidly and local pollen availability was unusually low during the summer and fall of 1991. Unlike our expectation, that stored pollen levels might fluctuate somewhat around a "set-point", pollen stores instead seemed to fluctuate more or less inversely with brood levels and time of the year (ie. swarming or non swarming season). We plan to study the stored pollen level phenomenon further using both observation colonies and unmanipulated standard colonies on a daily basis for one year.

4. Buchmann, S.L.[a] and H.G. Spangler[a] — THERMOREGULATION BY GREATER WAX MOTH LARVAE[ff] — Greater wax moths (*Galleria mellonella* L.) are pyralid moths whose larvae live by ingesting old brood combs and other materials within managed and feral honey bee colonies. Females can oviposit 1,000 or more eggs at a time. The mature larvae are large and pass through 7-8 instars living communally inside silken tubes within dark brown honey bee combs. Some beekeepers have noted heat production by large numbers of larvae inhabiting infested bee equipment. Until now, however, there have been no published empirical studies of thermoregulation by *Galleria* larvae.

During 1991, we started wax moth cultures by inoculating an artificial diet with 1st and 2nd instar larvae, then placed the medium in an environmental chamber at 28°C, 30% R.H. and on a 12 hour photoperiod. One thermocouple probe was placed in the center of the culture while another monitored chamber air temperature. Data was automatically collected every 15 minutes using an Easy Logger datalogger (Omnidata International, Inc.). Most data resulted from our second experiment which began on June 10 and ended on July 2, 1991. A total of 850 larvae were placed in 985 g of artificial diet. By June 12 the larvae were growing rapidly and the culture temperature was already 23°C.

The figure shows the larval culture and chamber air tem-

perature for the 20 day period in the upper panel and for a 24 hour period (June 18), the highest temperature achieved, in the lower panel. The uppermost traces represent culture temperatures while the lowermost ones show environmental chamber values.

Heat production (thermoregulation) begins during the 3rd or 4th instars and remains elevated for 3-4 weeks by which time the food supply is exhausted and pupation has begun. Temperatures as high as 41.3°C were produced by these "social" pyralid larvae.

We hypothesize that heat production in this species may function to aid metabolism of complex wax esters, promote rapid larval growth and possibly eliminate interspecific competition from other sorted product insects.

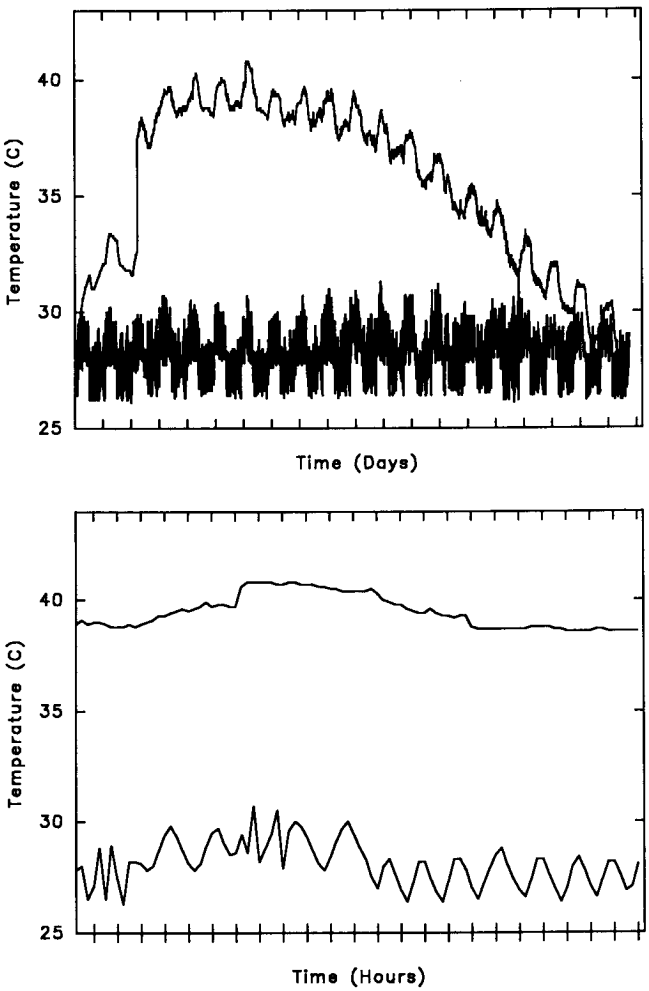
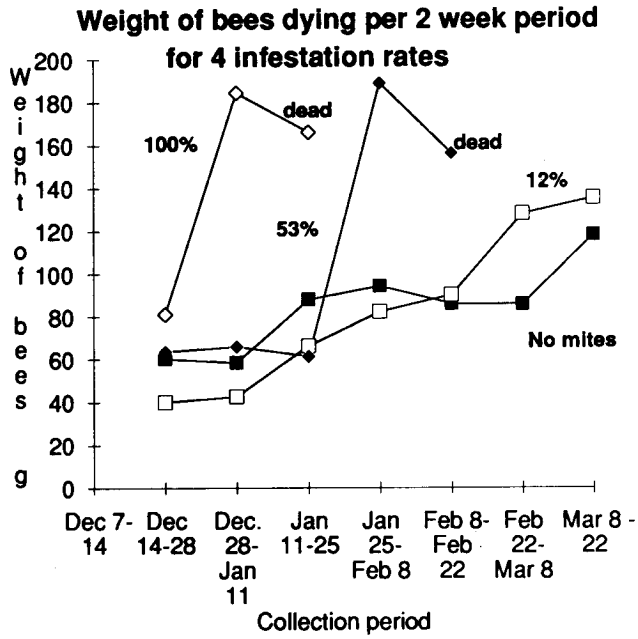


Figure - Upper panel shows the temperature of the wax moth culture (upper trace) with its rapid rise then gradual cooling above the ambient temperature (lower trace) set at 28°C. The lower panel gives an expanded record for the day (June 18) when the culture reached its maximum temperature of 41.3°C.

5. Clark, K.J.[b] – WINTER SURVIVAL TIMES OF COLONIES INFESTED AT VARIOUS LEVELS BY TRACHEAL MITES – Two-chamber colonies were housed in a dark overwintering building from early November until early April. Tracheal mite infestation levels were determined in November and 4 colonies from 0 to 100 % infested were selected for monitoring. Bees that dropped from individual colonies were collected every 2 weeks from early December to late March (Figure). From a colony with all bees infested, large amounts of bees died starting at the end of December, and the colony was dead at the end of January. From a colony infested 53 %, large amounts of bees died starting at the end of January, and the colony was dead at the third week in February. A colony infested 12 %, and another without mites, had lower but gradually increasing amounts of dead bee fall, and both survived the winter.

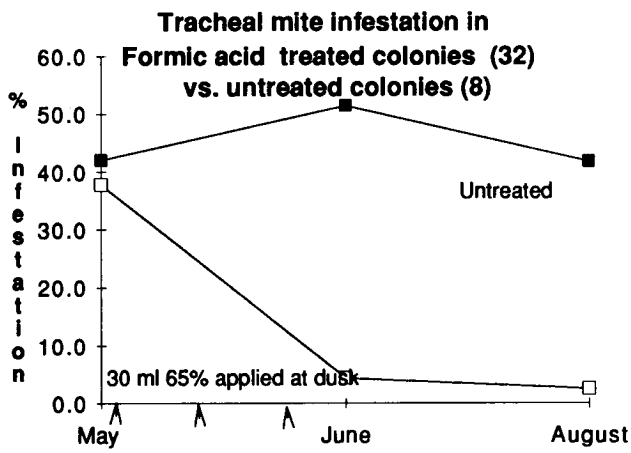


6. Clark, K.J.,[b] and J. Gates[c] – TRACHEAL MITE CONTROL TRIALS IN BRITISH COLUMBIA – Tracheal mite control trials that were started in April were partly inconclusive as a result of high variability in both treated and untreated groups. The trials were repeated in summer and it was concluded that:

1. Vegetable oil-soaked corrugated cardboard squares applied weekly, did not decrease mite infestation;

2. Formic acid (30 ml, 65%) applied 3 times at 1 week intervals, reduced mite prevalence between 50 and 90% compared to untreated colonies. Better results may have resulted from evening applications. A study in progress suggests that formic acid content of honey from spring treated colonies, may be no higher than that from untreated colonies. Fall treatment is being investigated.

3. Menthol was effective during a late May to mid-June period, but combined with brief hot periods, may have contributed to colony failure.



7. Collison, C. H.,[d] H. R. Fulton,[e] M. Tomasko[f] and J. Steinhauer[g] – EQUATING ETHER-ROLL SAMPLING RESULTS WITH TOTAL NUMBER OF VARROA MITES,

VARROA JACOBSONI, PRESENT – Honey bee colonies in varroa mite, *Varroa jacobsoni*, infested apiaries in Pennsylvania and Mississippi were sampled with the ether-roll technique to determine the extent of mite infestation. Following sampling, we initiated the following studies to determine the relationship between ether-roll sampling results and absolute female mite densities found within the colony (Pennsylvania) and within the ether-roll sampling container (Mississippi). Fifteen Pennsylvania colonies were killed by either brushing or sucking bees from individual combs into containers of 70% ethanol. Bee samples were kept separate by individual comb positions. After the combs were cleared of bees, any remaining bees in the unoccupied hive body and on the bottom board were combined into an additional sample for each hive. In the laboratory, absolute colony mite densities were determined with a washing technique (DeJong, *Glean. Bee Cult.*, 107: 639-640,644). Subsamples of 200 bees each were removed from alcohol, sorted for mites and shaken vigorously (either by hand or mechanically) for 10 minutes in a warm, weak detergent solution. The solution and bees were then poured into a funnel fitted with a screen to prevent bees from passing through. Bees within the funnel were stirred while being rinsed several times with water. Any mites washed from the bees were collected by a 0.180-mm mesh brass sieve placed below the funnel. Infested colonies contained a mean of 9,735.2 ± 1021.2 bees (range 4447-16840) and 60.4 ± 34.8 mites (range 0-529). Percent colony infestation levels ranged from 0.0 to 5.5% with an overall mean of 0.6%. Each comb had a mean of 1050.6 ± 59.5 bees (range 17-2905) and 6.5 ± 1.7 mites (range 0-152). A regression analysis of number of bees (Y) to absolute numbers of mites (X)/comb sample was significant ($df=138$, $F=14.328$, $p=.0002$, $R^2=0.1$). The resulting equation $Y=11.053X + 978.517$ indicates that at extremely low infestation levels, a minimum of approximately 1,000 bees should be sampled.

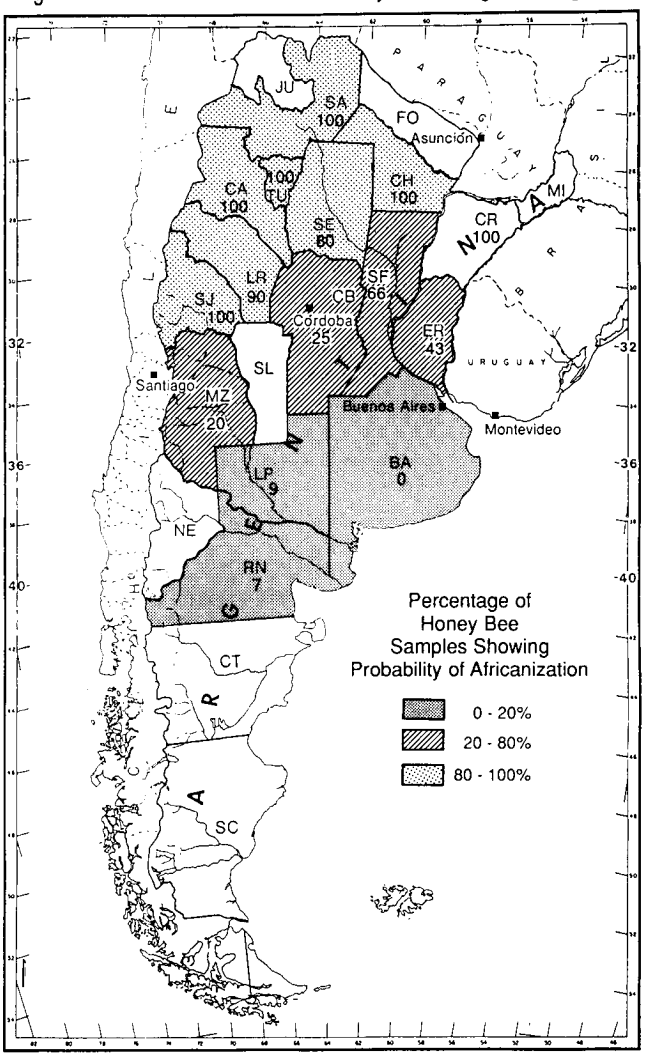
In Mississippi, 18 colonies were sampled with the ether-roll technique, number of mites counted on the inside of the jar, and then the sample was preserved by adding 70% ethanol. In the laboratory, bees were counted and any mites seen were removed. Samples were further checked for mites by the washing technique described above. More mites were extracted from the ether-roll bee samples in the laboratory than were seen on the inside of the jar at the time the sample was taken (16.9 ± 4.4 and 9.7 ± 3.0 mites, respectively). The size of the bee samples ranged from 134 to 737 bees with a mean of 326.2 ± 47.7. Regression analysis of mites sampled with the ether-roll technique (X) to absolute mite densities (Y) in the jar was significant ($df=17$, $F=52.7$, $p=.0001$, $R^2=.77$) and produced the equation $Y=1.267X + 4.624$. In all samples in which the ether-roll technique indicated 0 mites, no additional mites have been found to date in the sample after hand sorting or with the washing technique.

8. DeGrandi-Hoffman,[a] G., I. Terry,[h] and D. Li[h] – PC-REDAPOL: A POLLINATION AND FRUIT SET PREDICTION MODEL FOR APPLES – The mainframe computer version of the REDAPOL cross-pollination and fruit set prediction model (DeGrandi-Hoffman *et. al. Environ. Entomol.* 16:309-318. 1987) has been converted to run on IBM compatible personal computers. PC-REDAPOL generates predictions on the percentage of apple blossoms setting fruit hourly during bloom, and a cumulative fruit set percentage at the end of bloom. Fruit set predictions are based upon weather conditions, orchard design, blossom viability, and the size of the honey bee foraging population in the orchard. PC-REDAPOL has features that were not included in the original REDAPOL such as the ability to update sample counts of the number of honey bees per tree so that a new estimate of the foraging population can be generated at any time during the simulation, on-line HELP, and sampling schemes so that growers can enter the exact conditions of their orchard into the program. PC-REDAPOL also can simulate orchards that use pollen dispensers to enhance pollination. PC-REDAPOL is more interactive than the original REDAPOL and asks users at the beginning of each day whether they have taken a new count of the number of foraging bees per tree so that the foraging population estimate can be updated, whether pollen dispensers will be used that day, and the cultivar of pollen that will be put in the dispenser. PC-REDAPOL is more than a fruit set prediction model. The program also contains information on major bloom period insect pests of apple, how and when to sample them, and recommendations on how to control the pests if they are at economic threshold levels.

At this time the pest species are limited to those occurring in the western U.S., but in the future, information and recommendations will be made available for pests of other regions. PC-REDAPOL is public domain software and available by sending a formatted 720Kb or 1.2Mb floppy disk to Dr. DeGrandi-Hoffman.

9. Dietz, A.,[i] C. Vergara,[j] F. A. Eischen,[j] and R. Krell[k] – PRELIMINARY INVESTIGATION ON THE DISTRIBUTION OF FORAGING HONEY BEES IN ARGENTINA – Africanized honey bees (AHB) were found in most of the northern provinces of Argentina; such as Formosa, Misiones, Corrientes, Chaco, Santiago del Estero and Santa Fe by 1968 (De Santis and Cornejo, *Rev. Fsc. Agron. La Plata*, 44:17-35, 1968) and in Cordoba by 1970 (Dietz *et al. Apidologie* 16:99-108,

Fig.1. Distribution of Africanized Honey Bee Foragers in Argentina



1985). To determine the distribution of feral populations of AHB in Argentina, we collected foraging honey bees from "roadside" patches of flowering plants. Although we collected foragers in areas where colonies were not readily seen, it is possible that some of these foragers came from managed rather than feral colonies.

The distribution of the two geographic types of foragers collected in 14 provinces of Argentina is shown in the figure. Between 80 and 100% of the samples captured in the northern provinces were AHB, identified by morphometric discriminant methods (Daly and Balling, *J. Kansas Ent. Soc.* 51:857-769, 1978). For samples collected in Entre Rios, Santa Fe, Cordoba and Mendoza, the range was 20 and 80%. In the Buenos Aires, La Pampa and Rio Negro provinces, the range was 0 and 20%. No samples were collected in San Luis.

The distribution of foraging AHB and EHB appears to be very similar to the patterns observed for managed honey bee colonies in Argentina (Dietz *et al. Apidologie* 16:99-108, 1985). That is, a heavy concentration of AHB in the northern part of the country and a high concentration of EHB in the south. This distribution is the result of differences in the survival strategies of the two geographic types of honey bees. Since AHB have evolved strategies to cope with changing resources in tropical areas, they do not store large amounts of honey essential for survival in subtropical or temperate areas such as southern Argentina. Other major differences between AHB and EHB, including the strong defensive behavior of the former, however, are not more limiting than the natural restriction in food resource availability.

Based on these results, we concluded that the distribution of foraging honey bees in Argentina is influenced by various environmental conditions, including water and food resources and nest site availability. Low temperature is not the dominant factor limiting the distribution of Africanized honey bees in Argentina.

This research was supported in part by Cooperative Agreement No. 25-21-RC293-085 (Principal Investigator: Alfred Dietz) from the Honey Bee Breeding, Genetics and Physiology Laboratory, USDA, Baton Rouge, LA 70820.

10. Eischen, F. A.[j] and B. A. Underwood[j] – CANTALOUPE POLLINATION TRIALS IN THE LOWER RIO GRANDE VALLEY – The study was conducted in three commercial cantaloupe fields located within two miles of one another about 5 miles west of the Rio Grande City, TX. All fields were direct-seeded using the cultivar Primo in single rows on 80-inch wide beds with 12-inch spacing between plants. Black polyethylene mulch was used and plants were watered as needed with drip irrigation.

Honey bee colonies belonging to a commercial beekeeper were moved to the fields about 7 days after the onset of female flowering and remained in place until 16-18 days before harvest. Three fields, designated, viz., S-2 (Control, 1.25 colonies/acre, clumped, 37 acres), Boone L. (3 colonies/acre, clumped, 12 acres), and R-4 (3 colonies/acre, dispersed, 18 acres). Yields were assessed by weighing, and sizing each melon harvested (at full slip at 1- to 2-day intervals) from five plots (each 100 meters) within each field.

Primo melons from another experiment were utilized in comparisons of individual melon weights, sugar content, and seed number and weight. Sugar contents of melons are the means of three equatorial samples measured with a hand-held refractometer. Seeds were weighed after drying them to constant weight.

The fields receiving three colonies per acre out-yielded the field receiving 1.25 colonies per acre by considerable margin. The five plots within field R-4 (3 colonies, dispersed) produced a total of 1863 melons weighing 3177 kg or 52% more melons and 97% more weight than S-2 (1.25 colonies, clumped), which yielded 1226 melons weighing 1610 kg. Boone L. (3 colonies, clumped) was intermediate in production with 1422 melons weighing 2141 kg. The differences between R-4 and S-2 translate into nearly 7 tons more per acre for R-4. Not only were more melons harvested from R-4 and S-2, but those melons included a greater percentage of large fruit.

For cantaloupes of this variety, melon size is a good indicator of quality. Sugar content increases with melon weight. Both seed number and total seed weight were also significantly correlated with melon weight and presumably reflect, at least in part, pollination effects.

11. Ferrari, T. E.,[k] B. Palmer,[k] and C. Samimy[k] – COMPONENTS OF ALMOND POLLINATION – Almond flowers are self-incompatible and must be pollinated by another variety for fertilization to occur. Unfavorable conditions in commercial orchards often limit natural pollination. Two common problems which cause inadequate nut set are failure of pollinizers to bloom synchronously with the main variety, and reduction in the number of pollinizer trees due to disease. Application of precollected pollen to honey bees (enpollination) can augment cross-pollination by natural means. However, opinions vary in regard to the efficacy of supplemental pollination.

A definitive way to prove efficacy of colony enpollination is to perform paternity tests on embryos produced following flower set. Almond varieties possess proteins with unique isozyme banding patterns that permit identification of male parentage. The contribution to yield from supplemental pollen can then be expressed as a percentage of a crop produced. We sampled nuts from six orchards where paternity testing could be performed. Proteins in embryos of nuts were evaluated by electrophoresis.

In Orchards 1-4, from 6–24% of the nut crop resulted from supplemental pollen (+P, Table). Orchard 5 contained enpollinated colonies, but levels of the protein marker were low or undetectable in nuts from Carmel (5,C) and Nonpariel (5,NP). However, the orchard has an above average yield history and a heavy crop was produced during the test. Orchards with the poorest yield history had the greatest contribution to nut production from supplemental pollen.

In five orchards we determined the natural occurrence of marker proteins to be less than 2% (−P, Table). All locations sampled were more than 100 feet from the nearest known source of pollen contamination. Background levels were not determined from orchard 2, the most isolated.

Background levels for protein markers in Orchard 6, containing Merced and Nonpariel, was 2% (n=100) and 1% (n=200), respectively, in 1990 and 1991. Of 24 plots sampled in both years, embryos with the marker protein were from the same site, suggesting contamination from a nearby "stray" Mission tree in the orchard.

In summary, paternity tests indicate that precollected almond pollen applied to colonies was transferred by foragers to

Table – Embryos with marker isozymes.

ORCHARD	−P	+P	DIST TO CONTAM	YIELD HIST
	%	%	Feet	
1	N	24	31000	light
2	<1	18	3000	light
3	0	16	600	medium
4	0	6	100	medium
5,NP	0.25	0	2600	heavy
5,C	0	0.7	2600	heavy
6,1990	2	N	>5000	medium
6,1991	1	N	>5000	medium

N = no data or no enpollination

flowers and resulted in fertilization and nut set. As a cultural practice, enpollination is most profitable in orchards when poor pollination limits crop production.

12. Gilliam, M.[a] and B. J. Lorenz[a] – ENZYMOLOGY OF MYCELIAL AND SPORULATED STRAINS OF *ASCOSPHAERA APIS*: MARKER ENZYMES AS TAXONOMIC AIDS – Identification of the honey bee chalkbrood pathogen, *Ascosphaera apis*, from bees and hive products is relatively easy when + and − mycelia have mated to form characteristic spore cysts. Since mycelia alone are often isolated and can only be positively identified as *A. apis* by performing mating tests, easier and faster methods are needed.

Therefore, we determined the enzymatic profiles of 15 strains of *A. apis* (2+, 7−, and 6 mated) using the API ZYM system. Procedures to prepare inocula for API ZYM tests were devised since methods for other molds were unsatisfactory.

All strains produced alkaline phosphatase, butyrate esterase, leucine aminopeptidase, acid phosphatase, and β-glucosidase. Alkaline phosphatase was the only enzyme produced in high concentration (≥30 nanomoles) by all strains. Myristate lipase, trypsin, α-galactosidase, β-glucuronidase, and α-fucosidase were not produced.

There are some differences in our results compared to those of isolates from Spain (Alonso *et al., 5 Congr. Nac. Apícola, Spain*, 44-46) which could be due to variations in strains, methods, or API ZYM kits. For example, only one of our strains approached the levels of phosphoamidase that were found in Spanish strains, and only one (6%) produced a trace of α-glucosidase, whereas 78.3% of Spanish strains produced detectable amounts of 10nm.

Predominantly mycelial inocula of some fungal species produce the same enzymes, although in reduced amounts, as spore suspensions. However, with *A. apis* the opposite occurred in regard to valine aminopeptidase, phosphoamidase, and N-acetyl-β-glucosaminidase. The most obvious potential marker enzyme for mycelial strains of *A. apis* is valine aminopeptidase which was produced by unmated but not by mated strains. Few molds associated with honey bees produce this enzyme. β-Galactosidase and α-mannosidase may be candidate marker enzymes for both mated and unmated strains since they were produced by most strains tested, but are rarely produced by other bee-associated molds.

Testing of additional *A. apis* strains will determine whether these enzymatic profiles are indicative of most strains and will test further the feasibility of using marker enzymes to identify unmated *A. apis* strains and to distinguish *A. apis* from other molds commonly associated with bees.

13. Harbo, J. R.[1] – EFFECT OF CELL SIZE ON QUANTITY OF BROOD AND WEIGHT OF WORKER BEES – Colonies having larger cell sizes must increase the area of their broodnest if they are to equal the brood production of colonies having smaller cells. An objective of this study was to determine whether bees regulate brood production by numbers of cells or by brood area.

Fifteen colonies were established on May 15. Each colony contained 6304 ± 171 (mean ± SD) worker bees, a queen, a feeder, and 4 frames of beeswax foundation. Six colonies had foundation with 711 cells per sq. decimeter (dm^2), and 9 had foundation with 857 cells /dm^2 (a natural size for European bees). Worker populations were made uniform by first placing all worker bees in a large, screened cage and then subdividing this population into 15 parts. A second experiment was set up on August 23 to compare an even smaller cell size (1004 /dm^2, a natural size for African bees) with the largest size (711/dm^2). This experiment was very similar to the experiment above except only 8 colonies were established and drawn combs were used instead of foundation.

Colonies compensated for the large cells by increasing their brood area to maintain about the same number of broodcells. Colonies with larger cells (711/dm^2) produced larger areas of brood than colonies with smaller cells (857/dm^2), but the two groups never differed in numbers of brood cells (see table).

Larger cells produced larger bees. The difference between worker bees from the large and medium sized cells was 6 mg (113 and 107 mg) in the first experiment ($P<0.01$). The difference between workers from the largest and smallest cells in experiment 2 was 11 mg (117 and 106 mg; $P = 0.001$).

Table - Comparisons of 6 colonies having large cells with 9 having medium sized cells. Area and number of cells are means that represent the total amount of capped brood in the colonies.

Cell size (per dm^2)	June 5		June 17		June 28	
	area	No. cells	area	No. cells	area	No. cells
711	613cm^2	4356	357cm^2	2538	504cm^2	3682
857	529cm^2	4536	283cm^2	2429	433cm^2	3714
P^1 =	0.036	0.54	0.006	0.53	0.087	0.68

[1]Probabilities that the means listed above represent the same population.

14. Harbo, J. R.[1] – LAYING WORKERS PRODUCE A DRONE POPULATION THAT GENETICALLY REPRESENTS THEIR COLONY – In bee breeding, one normally evaluates colonies and then selects some of them as breeding stock for the next generation. Queens produced from these colonies are sisters of the workers that were evaluated, so a group of queens is a good representation of a colony. In contrast, the drones from a colony represent only the genes of the queen.

The objective is to produce a population of drones with gene frequencies that represent the worker population of a colony. Drones were produced from egg-laying workers in field colonies. The technique was to remove a brood chamber, as many young worker bees as possible, and capped brood from the main colony and place them next to or on top of the main colony but with the entrance facing a different direction. The older workers soon returned to the main colony, leaving only young workers and capped brood in the queenless portion which was fed pollen and given a comb with drone-sized cells. Drones were reared in this queenless colony from eggs laid by the workers. If successful, spermatozoa from these drones would accurately represent the entire colony. Moreover, by mixing this spermatozoa (Harbo, *J. Apic. Res.* 29:151-158), the entire colony can be genetically represented in the insemination of a single queen.

To be successful, (1) drones must be produced from every subfamily of workers in the colony and (2) there should be minimal subfamily dominance for ovary development of workers or for survival of drone progeny. I tested the first requirement by inseminating 7 wild-type queens with genetically marked semen from 5 different colonies. Each colony had one of the following single gene recessive markers: cordovan body (cd), hairless (h), diminutive wing (di), chartreuse eye (ch^B), and white eye (s). Worker progeny from these matings did not express any of the recessive genes, but they were each heterozygous for one of the marks. Workers from the 7 colonies were stimulated to produce drones, as described above, and these drones were checked for mutant marks. The mark identified the subfamily parentage of the drone. One half of the drones were expected to express one of the 5 marks. About 200 drones were examined from each colony.

Results showed (1) that drones were produced by all subfamilies of workers in all 7 colonies (similar results were obtained

when this was repeated with 5 surviving colonies the following spring) and (2) none of the five markers affected the survival of egg, larval, or pupal stages of drones. I arrived at this second point because the frequency of mutant drones from all 7 colonies was 49.1% (1362/2775), very close to the expected 50%. The 95% confidence interval for the fraction was 46-52%.

This study was designed to detect only large differences in the number of drone progeny from each subfamily of laying workers. Neither total dominance nor total absence of a subfamily was observed when scoring the drones for genetic marks. Slight differences may have existed, and large differences could occur with different stocks.

In addition to its potential value in selective breeding, the use of egg-laying workers in field colonies enables one to (1) produce drones at suboptimal times of the year, (2) produce drones from weak colonies, (3) produce drones from colonies that have become queenless or recently superseded, and (4) retrieve various combinations of genetically marked drones from a single colony.

15. Loper, G.M.[a] and R.M. Roselle.[a] — EXPERIMENTAL USE OF BEESCENT® TO INFLUENCE HONEY BEE VISITATION AND YIELDS OF WATERMELON[ff] — This paper presents results of the use of BeeScent® to watermelons (*Citrullus lanatus*) under irrigated field conditions in central Arizona, Aug. - Sept. 1991. Data on percent bee visitation and watermelon yields were subjected to statistical analysis (ANOVA) using the "minitabs" program (release 1.1, 1988) from Minitabs, Inc. A commercial planting of watermelons in 2, adjacent 14-acre fields near Coolidge, AZ was studied. Each field was planted (June 28) to two cultivars in a 2 X 4 pattern. Two rows of the cv "Picnic" provided pollen for 4 adjacent rows of a seedless cultivar (Maynard and Elmstrom, 1990). One field was used as a control while the other had alternate, 18-row strips which were either untreated or treated using an 18-row boom sprayer.

Flower and bee visitation counts were made in both cultivars within 3 treated, 3 untreated, and 3 control plots (20m/plot). On August 15, the plants were in bloom but covered only about 85% of the row. BeeScent® (2 qts/acre in 20 gal of water) was applied by a tractor mounted ground sprayer operated at 90 psi and 5 mph. Application was made between 7:25 - 07:40 A.M. MST at 29°C with overcast skies and a slight drift of spray was visible extending out to 2 - 3 rows (2 - 3m) downwind. Honey bees (2 col/acre) had been delivered in the evening of Aug. 8, 1991 and placed in groups of 4 colonies along all sides of the fields. Counts of bees and flowers were made at 0900 and 1000h on Aug 15, 16, 17, 18 and 19 and counts of watermelons (23cm or longer) were made on Sept. 16 and 17 (in 20m of row) in each of 10 randomly selected plots in each study area (treated, untreated, and control).

Percent bee visitation was high (7.6%) on the day of application, dropped on the next day and then generally remained close to 4% even as flower numbers increased. In the seedless cv, there was a significant (P = .06) difference in bee visitation between treatments only on Aug. 16. In the "Picnic" cv there was a significant increase in number of bees on Aug 15 (P = .01) and Aug. 16 (P = .14), but again the difference disappeared with time. We have no explanation for the different results on Aug 15, the day of application, but after that the trends were similar, more bees on the treated plots for the next day, but by the 3rd day after treatment there was no difference between treatments. Watermelon yields were considered "above average" by the grower so pollination had been accomplished satisfactorily, but there was no significant difference in yields between the treatments. In fact, with both cvs, there were slightly more watermelons in the control field than in the treated field.

The use of BeeScent® did increase honey bee visitation for up to 2 days, but the influence quickly disappeared. Temperatures by 10 a.m. were generally about 33 - 35°C (90 - 95°F) and increased to 36 - 38°C (97 - 100°F) daily under sunny skies. It was not possible for us to smell the applied spray even on the afternoon of the day of application. Waller (*J. Apic. Res.* 9:9-12. 1970) noticed that marked foragers returned to a scent treated plot on subsequent days. We suspect that bees that chanced upon the treated plots soon after spraying tended to return to that plot even after the actual scent was gone and that this effect temporarily increased bee visitation to the sprayed plots. Since watermelon yields were not increased it is concluded that application of BeeScent® under these condition was ineffective.

16. Loper, G.M.,[a] W.W. Wolf,[m] and O.R. Taylor[n] — RADAR DOCUMENTATION OF FLIGHT PATTERNS OF HONEY BEE (*APIS MELLIFERA* L.) WORKERS, SWARMS AND DRONES — A trailer-mounted x-band radar unit placed adjacent to a commercial apiary was used to observe the flight of honey bees (*Apis mellifera* L.). Honey bee activity was recorded on black and white film. By varying the elevation of the beam and time-of-day, foragers were distinguished from drones. Drones established flyways and congregation areas and responded to queen pheromone. Movies of this activity were converted to VHS format. A 10-minute video presentation includes both "raw" radar images and computer-enhanced images permitting the viewer to observe the bee's behavior as influenced by wind and terrain features.

17. McKenna, W. R.[o] — AFRICANIZED HONEY BEE: REACTIONS AND POTENTIAL ANTIVENOM TREATMENT — Increased life threatening allergic (IgE mediated) reactions can be expected because of more contact with the AHB's in the Southern U.S. However, current allergy testing and venom immunotherapy should be adequate since biochemical and immunochemical comparisons of AHB and domestic honey bees are very similar (Nelson & Collins, *J. Allergy Clin. Immunol.* 85:80-85, 1990.)

However, also there will be more toxic reactions from multiple stings especially when 100 and above stings occur. Death (toxic) has occurred with as few as 100 to 200 stings. The elderly and small children are most suseptible. There have been more than 40 deaths documented in Mexico in the past five years.

The LD_{50} of domestic and Africanized honey bees are similar in experiments using IV injection of venom in Swiss mice (Schumacher & Schmidt, *Am. J. Trop. Med. Hyg.* 43:79-86, 1990.).

Also (see the prior reference) studies with Swiss mice have revealed using beekeeper's serum antibody protected against toxic reactions when venom was injected simultaneously. Also, in allergic patients who had significant allergic reactions to immunotherapy, individuals were pretreated with beekeeper's hyperimmune IgG and then were able to be injected with bee venom immunotherapy without systemic reaction (Bousquet, *J. Allergy Clin. Immunol.* 79:947–54, 1987). Therefore, studies show neutralization of the toxic reaction and systemic reaction.

Therefore, with the above information and studies it would seem logical to use beekeeper's hyperimmune IgG to counteract the severe toxic reactions of individuals who receive high numbers of bee stings. I have proposed a model for this and referred to the table for details. It is certainly logical that the amount of antivenom will vary relative to the severity of the reaction (especially related to victims age, weight, and health) which is similar to the amount of antivenin used in snake envenomation. Another potential plan is establishment of a plasma cell line able to secrete monoclonal antibodies against *ie.* mellitin (and possible other venom components) which is the main toxic component of bee venom. Therefore, a specific antibody can be directed against single components and known quanity as opposed to antibodies developed to a variety of venom com-

ponents as would be in the beekeeper model.

We do need to identify more about pathophysiologic mechanisms associated with individual venom components.

Table – Proposed example of amounts of Antibody needed for protective antitoxin effect

Bee Venom Exposure	Beekeeper Ab.+
2.85 * mg. venom/Kg wt. Child of–20kg	1,400mg
5.7 mg venom/Kg wt. Child of–20kg	5,700mg
2.85 mg venom/Kg wt. Adult of–60kg	4,000mg
2.85 mg.venom/Kg wt. Adult of–80kg	5,300mg
5.7 mg. venom/Kg wt. Adult of–60kg	17,000mg
5.7 mg. venom/Kg wt. Adult of–80kg	23,000mg

* LD_{50} in Swiss mice model (which is approximately equal to 19 stings per Kg weight in humans)

+ Example of Beekeeper with high levels of antibody to honey bee venom.

18. Meade, D.E.[p] – EFFECTIVE FORAGING RANGES OF FERAL COLONIES – Surveys of honey bee foraging activity before and after colony removal on Santa Cruz Island, Santa Barbara County, California, revealed several characteristics of colony foraging patterns, including an upwind foraging bias, a failure to exploit rich sources very near a colony, and an expansion of foraging range when colony density was reduced.

Although honey bees theoretically forage an average distance in all directions from their colony (Roubik, *Ecology and Natural History of Tropical Bees*, pp 82-90. 1989), actual patterns may be influenced by topography, wind, and competition. Feral honey bees in the eastern central valley of the island appeared to forage primarily upwind from their colony, even though abundant forage existed downwind at similar distances (see figure). Removal of the target colony indicated that foragers travelled up to 1600 meters upwind of the colony; no significant difference in honey bee numbers occurred downwind after colony removal.

Differences in forager distribution caused by experimental manipulations can be detected in the field even when several other colonies forage in the same area. An experiment, that determined honey bee presence along a 1280 meter transect in the western central valley during a manipulation of colony flight activity, indicated that foragers travelled upwind no more than 1000 meters from the colony in an area where at least 7 other feral colonies also foraged.

Surveys in Sauces Canyon revealed that honey bees may not always forage on the richest nectar sources. Honey bees from one colony visited *Malva parviflora*, but ignored *Brassica* spp. in the same area. A second colony 600 meters downcanyon visited primarily *Brassica* spp. and *Raphanus sativus* and ignored *Malva parviflora*. Colonies often remain faithful to less profitable food sources if they have concentrated on a particular plant before others bloom.

An expansion of honey bee foraging range seems to occur when colonies are released from intraspecific competition. Round trip times from feeding stations to colonies obtained during beehunting allowed comparisons of foraging range in high colony density (2.4 per square km) and low colony density (1.2 per square km) at the same site. Foraging range expanded significantly (round trip time from x = 7.4 min. to x = 12.2 min., $p < 0.001$) after reduction of feral colony density.

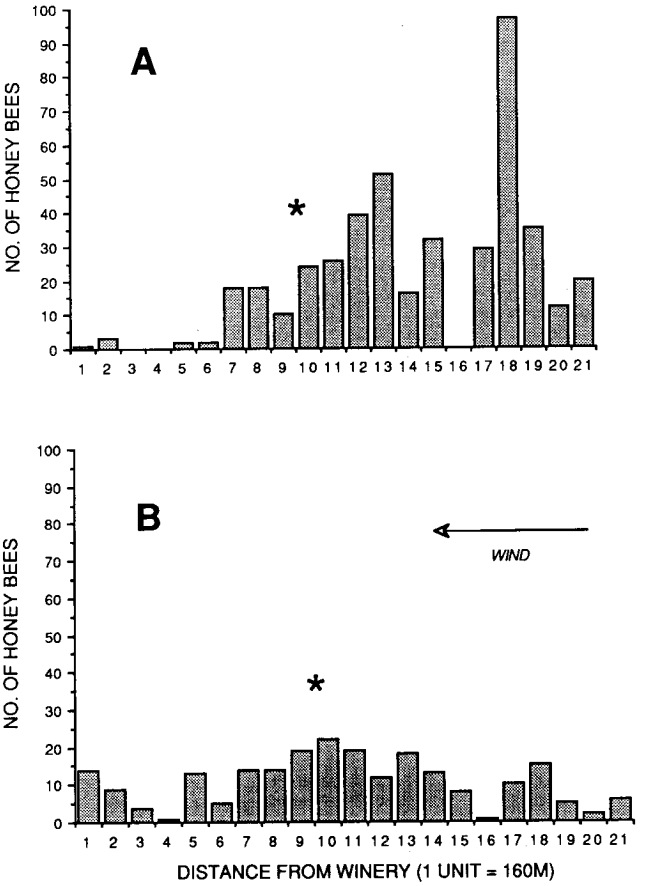


Figure - Honey bees appeared to forage primarily upwind from their colony on July 24, 1991 (A). Colony removal (asterisk marks place along transect) and recounting honey bee presence nine days later (August 2, 1991), resulted in marked decline of upwind visitation (B). The remaining foragers shown in B were from other colonies. Wind was steady from the east during the experiment. Bees were foraging on Sweet Fennel (*Foeniculum vulgare*), that maintained a consistent bloom during the study period; abundant forage existed both east and west of the colony throughout the survey area.

19. Nation, J.L.,[q] M.T. Sanford,[q] and K. Milne[q] – COMPARISON OF CUTICULAR HYDROCARBONS FROM VARROA MITES AND HONEY BEES – Gas chromatography (GC) was used to analyze the cuticular hydrocarbons of both mites and honey bees. This is the first report of the hydrocarbon pattern of *Varroa* mites.

The hydrocarbons from adult worker and drone honey bees have been identified and described by others (Smith, *Bee Science* 1:23-32; Carlson, *Africanized Honey Bees and Bee Mites*, Needham *et. al.* (eds.) pp. 264-274). The major components are the odd carbon, straight-chain molecules with 23, 25,27,29 and 31 carbon atoms, respectively (see figure). Additional small peaks represent unsaturated and branched chain homologs of those molecules. Three unsaturated hydrocarbons are also major components: two with 31 and one with 33 carbon atoms.

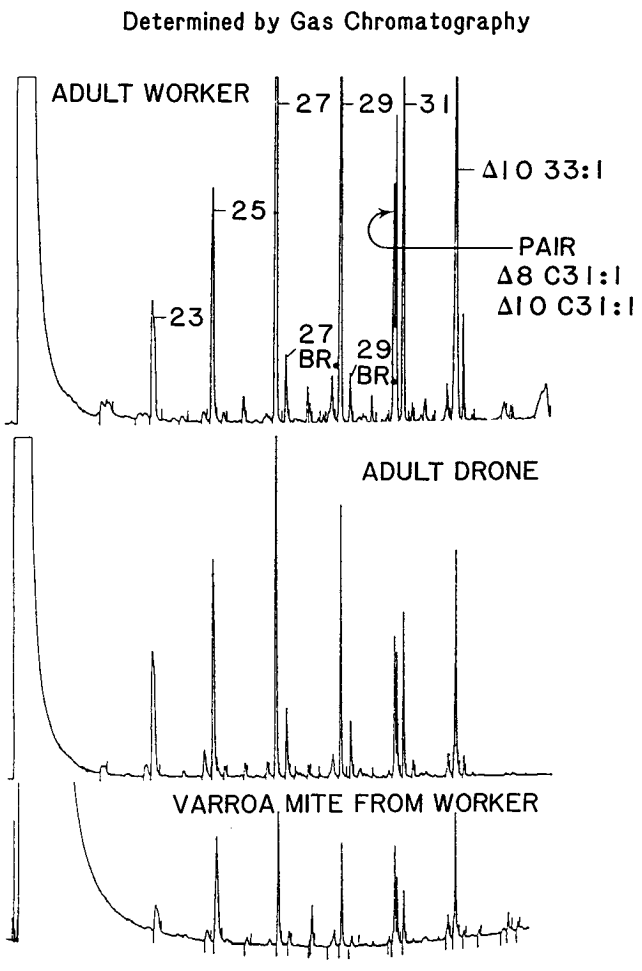
The figure shows the typical hydrocarbon patterns of an adult worker, drone and a *Varroa* mite from Gainesville, Florida. The bees show hydrocarbon patterns similar to what has been published elsewhere. The record in the figure is from a single mite taken from an adult worker bee. Its hydrocarbon profile is similar to both adult workers and drones.

GC tracings of hydrocarbons from the cuticle of pupal workers or drones (purple eye stage) are recognizably different from the patterns shown by the adults of both castes. Specifically, the large peaks for unsaturated hydrocarbons having 31 and 33 carbon atoms in the chain are not detectable or present only in very low quantity. The same is true for mites associated

with pupae.

To further document the similarity of mite and adult bee hydrocarbon profiles, we determined the location of the double bond in the C31 and C33 unsaturated hydrocarbons of both worker bees and mites by GC Mass Spectrometry techniques (details to be published elsewhere). The results were the same for both species. One of the two C31 hydrocarbons had a double bond at position 8, while the other C31 and the C33 molecules' were both located at position 10.

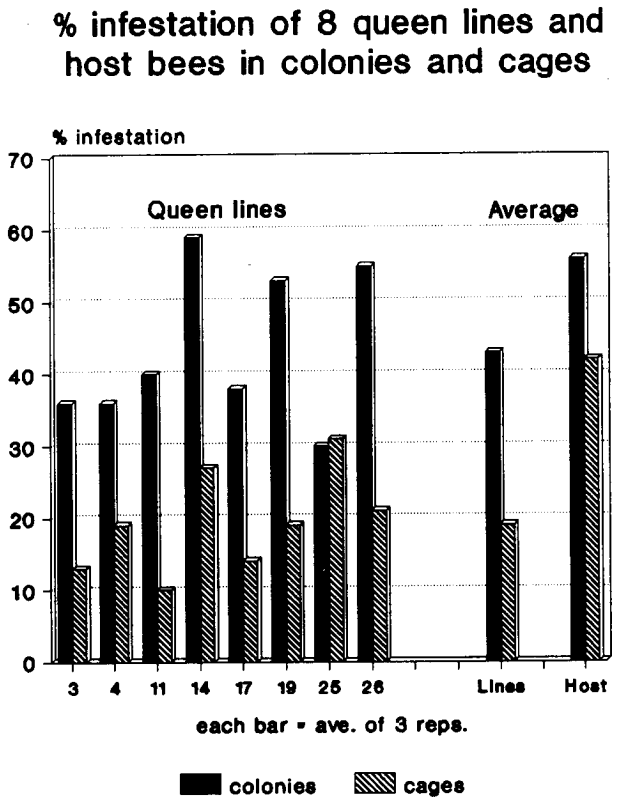
In conclusion, the pupal and adult stages of worker and drone honey bees have easily discernable differences in their cuticular hydrocarbon patterns. *Varroa* mites mimic very closely the pattern of their host stage. It is thus plausible to hypothesize that failure of *Apis mellifera* to clean mites from their own body or those of nest mates may be due to difficulty in recognizing the mites as different from their host. Data are too limited at present to justify further conclusions on the significance of this mimicry or the taxonomic use of hydrocarbon patterns.



20. Nelson,[r] D.L., G. Grant,[r] and D. McKenna[s] — COMPARISON OF COLONY WITH CAGE TRIALS FOR EVALUATING QUEEN LINES FOR RESISTANCE TO HONEY BEE TRACHEAL MITES — Newly emerged bees from eight queen lines were marked with different colors (target bees) and placed in colonies or cages with tracheal mite infested bees. Approximately 40 bees of each queen line were placed in each of the three infested colonies (single chamber) and approximately 20 bees of each queen line were placed in each of the three cages each containing approximately 100 host bees. After 14 days the target bees and a sample of 25 unmarked host bees were retrieved from the combs of each colony or cage and frozen. Later these bees were analyzed for tracheal mites by microscopic examination of KOH-cleared thoracic slices.

The target bees in colonies had a mean infestation level of 43% (range 30-59%), while the host bees in colonies had a mean infestation of 56% (range 37-83%). The target bees in cages had a mean infestation level of 19% (range 10-31%), while the host bees in cages had a mean infestation of 42% (range 25-70%) (Fig.).

The average infestation level of target bees in colonies was 43% and in cages was 19%. This difference is likely due to the difference in the ratio of infested host bees to target bees, which was estimated at 19:1 in colonies, whereas in cages was estimated at 1:4. This difference may be very important when determining whether to use colonies or cages in stock selecting procedures.



21. Royce, L. A.,[u] P. A. Rossignol[u] and B. A. Stringer[u] — AN APPARATUS FOR CONFINED MATING OF HONEY BEE QUEENS — We hypothesized that vision was a crucial parameter in honey bee mating. We describe a small chamber that provides indirect light within its upper half. When introduced into the chamber, drones will hover for long periods of time. If a tethered queen is also introduced, a behavioral sequence is often initiated that leads to copulation.

22. Schmidt, J. O.[a] and H. G. Spangler[a] — CAN THE ATTACK BY AFRICANIZED BEES BE STOPPED BY CHEMICALS?[ff] — An ideal solution to the problem of mass attack by Africanized bees would be a chemical spray that could stop the attack.To investigate this, we elicited attacks by Africanized bees from a ca. 40 hive apiary in Lomas Barbudal National Park, Costa Rica by stimulating each hive with a kick. The ensuing bee attack was measured with a modified temper testing device (Spangler *et al.* 1990. *Amer. Bee J.* 130:731-33), which records the number of stinging attempts/10 seconds directed toward a small black target. Baseline control values were recorded on the untreated target; treatment values were obtained by recording the stinging attempts during a time when

the test chemical was being sprayed in the air around and on the target. We tested a variety of detergents, surfactants, oils, DEET, and insecticides, most formulated as 1% solutions in water. As recorded in the table, most of the 11 detergents had no perceptible effect on the bee attack; Red Wing boot oil (tested because bees seemed not to attack boots so treated) was no different from the control, and balsam tar oil actually statistically increased the attack! DEET as a 1% suspension in water caused a minor reduction in the attack, whereas the Deep Woods OFF aerosol (28.5% DEET) produced a larger, but not statistically significant decrease. By far, the most effective treatments were the two aerosol insecticides containing tetramethrin or allethrin plus synergist.

Based on the tabulated data, it would appear that aerosol insecticides might offer promise for stopping a mass bee attack. Observations, however, indicate this is not so. Even in the best of treatments, including the insecticides and the DEET treatments, the attackers were repelled only from the immediate air space of the spray. In all cases, as soon as the spray stopped, the attack returned at essentially the same level as before the spray. These observations indicate that none of these treatments can affect meaningful control, or be of actual benefit under field conditions. Indeed, this conclusion is not surprising — as the individual well-being of attacking bees is not likely to be of importance in a species that readily "commits suicide" by stinging.

Table — Effect of chemical sprays on numbers of attacks on a target by Africanized bees

Test material	No. of target strikes/ 10 sec ± SD: No material	Sprayed material	% Change[1]
DETERGENTS/SURFACTANTS			
Palmolive Lemon/ Lime	46.7 ± 7.6	49.5 ± 7.8	+6.1
Brij 35 (wetting agent)	40.0 ± 4.2	38.0 ± 1.4	−5.0
Kodak Photo 200	61.0 ± 14.9	57.0 ± 4.0	−6.6
BioRad Cleaning Concentrate	68.5 ± 8.3	64.0 ± 4.6	−6.6
White Magic Glass Cleaner with Ammonia	68.3 ± 14.4	63.7 ± 4.2	−6.7
Clout Laundry Detergent	63.5 ± 16.1	58.7 ± 11.8	−7.6
Spic & Span Pine Cleaner	63.0 ± 11.0	57.0 ± 2.7	−9.5
Tween 60	61.8 ± 8.0	52.0 ± 5.0	−15.9
Triton X-100	58.8 ± 12.6	42.0 ± 5.0	−28.6
Micro Lab Cleaning Solution	70.5 ± 6.4	48.3 ± 7.8	−31.5*
Dawn Original Scent	59.5 ± 21.6	39.3 ± 5.1	−33.9
OILS			
Balsam Tar Oil	51.1 ± 7.1	69.3 ± 1.5	+35.4**
Red Wing Boot Oil	65.0 ± 13.9	63.7 ± 1.5	−2.0
INSECT REPELLENTS			
DEET liquid	66.2 ± 16.4	39.7 ± 2.5	−40.1*
Deep Woods OFF aerosol (28.5% DEET)	55.2 ± 23.5	24.0 ± 1.0	−56.5
INSECTICIDES			
Black Flag Insect Killer	52.2 ± 30.0	43.3 ± 13.3	−17.0
Ace Flying Insect Killer	45.0 ± 17.0	9.0	−80.0
Raid Flying Insect Killer	58.7 ± 12.5	10.0 ± 1.7	−83.0***

[1] Means within a row differ significantly by t-test comparision: * = P<.05, ** = P<.01, *** = P<.001

23. Schmidt, J. O.[a] and S. C. Thoenes.[a] — SWARM TRAPS: 3- VERSUS 2-COMPONENT PHEROMONE BLENDS AND ROUND VERSUS RECTANGULAR DESIGNS — Although the ARS swarm trap we designed (Schmidt and Thoenes, 1987, *Bull. Ent. Soc. Amer.* 33(3):155-58; Schmidt *et al.* 1989. *Amer. Bee J.* 129:468-710) is very effective and is being used widely, there are still a few variables that need testing. The original pheromone lure consisted of an equal proportional blend of citral, geraniol, and nerolic acid. Since the nerolic acid is difficult to obtain, this 3-component blend has sometimes been simplified for large scale field use to a 2-component blend of citral and geraniol. The original trap was round, a shape that is inconvenient for use if the attracted swarms are to be transferred to standard hive equipment. To ease this problem we (Schmidt, 1990, *Amer. Bee J.* 130:333-34) designed a rectangularly shaped trap about the size of a 5-frame nucleus hive that can hold standard frames.

We established 18 stations scattered throughout the Tucson, Arizona basin during the spring swarming season, 1991. Each station contained four traps to include one representative of each possible combination of trap shape and attractant. The traps were checked once or twice a week during the season, captured swarms removed, and replacement traps added. An additional test of the 3- versus 2-component blend was conducted with round traps in Guanacaste, Prov., Costa Rica during March 1991. The results of the blend comparison tests in Tucson were: 2-component blend — 1 swarm; 3-component blend — 17 swarms; and two stations which had a swarm in each lure type. The 3-component attractant was clearly preferred over the 2-component attractant ($P < .0001$, binomial test). The data with Africanized bees was similar with the 3-component blend attracting 7 swarms, the 2-component blend 1 swarm, and the control (no pheromone) 0 swarms. The 3-component attractant was preferred over the 2-component attractant by swarms of this Africanized bee population ($P < .035$, binomial test).

Tests of the round versus rectangular traps were conducted only in Tucson. The results were: round — 13 swarms; rectangular — 7 swarms; both in one station — 2 swarms. These differences are not significant.

The results of these tests contain practical information of potential economic benefit. The original 3-component blend of pheromone components is much more attractive and effective than the simplified 2-component blend. Since the added cost per trap of including the nerolic acid component is trivial compared to the cost of the trap itself and, especially, to the labor to service traps, it seems wise to include nerolic acid in the lure. If the swarms are to be utilized for addition to an apiary and if the traps cannot be checked at least weekly during the swarm season, it might be advisable to use the rectangular traps with a few added frames of foundation. If the traps are for survey and the swarms likely to be destroyed, then the round shape is preferable for three reasons: round traps are cheaper to purchase and to ship, they are easier to deploy, and they might be more attractive.

24. Shen, L. G.[b] and J. O. Schmidt.[a] — ARE STING AUTOTOMIZED BEES SUICIDAL? — In worker honey bees individual reproductive fitness is expressed mainly by helping behavior in which workers pass their genes to future generations via increasing the reproductive capacity of their relative, the queen. Since their personal reproductive potential is nil, they can best transmit their genes by caring for and defending their colony. Accordingly, their own survival is of secondary importance to that of their colony. Honey bee sting autotomy, the loss of the sting apparatus upon stinging, represents the ultimate both in the individual's ability to defend the colony

and in self sacrifice. Once a bee has autotomized its sting, it will shortly die, and its value to the colony in terms of traditional work and foraging is essentially zero. For an autotomized bee, the best evolutionary strategy to increase its fitness, is to use what little is left of its life vigorously defending its colony. This it can do by attacking any suspected potential predator within sight and continuing the attack as long as possible. This strategy would be effective despite the lack of a sting because a potential predator cannot determine that the autotomized bee is essentially harmless.

To test the hypothesis that sting autotomized bees will behave more suicidally by persistently attacking potential predators longer and more vigorously than sting-bearing bees, we stimulated three separate apiaries of Africanized bees and enticed many individual bees to lose their stings. We then sampled the population of bees defending individual colonies or flying around our heads. The distance samples were taken at increasing distances, up to 510 m from the apiary. The sampled bees were chilled on ice, then analyzed for presence or absence of a sting. The results of these analyses supported the hypothesis that the percentage of attacking bees that were autotomized increases with distance from the apiary. In one apiary the percent of autotomized attackers increased from about 5% near a colony to 15-30% at distances over 400 m. In the other apiaries the respective percentages increased from about 3 and 12 to about 5.5 and 28.

A secondary hypothesis is that the stimuli for attack should be very high when the colony is directly assaulted and that the difference in the attack vigor between sting-bearing and autotomized bees should be small. To test this hypothesis we netted all the attackers emanating from two isolated colonies of Africanized bees and then captured all the bees remaining inside. The results of samples from the various collections revealed that there were no differences in the percentages of autotomy in early versus late attackers. These data suggest that in Africanized bees "suicidal" behavior is exhibited essentially equally among all attackers — sting-bearing or stingless — when the threatening predator is attacking the colony.

Preliminary evidence from European bees suggests that the differences between the behaviors of sting bearing and autotomized bees is greater than in Africanized bees. Overall, these investigations indicate that sting autotomized bees tend to be suicidal and that attacking bees do behave in a fashion that maximizes their inclusive fitness.

25. Smith, R.-K.,[v] M. Spivak,[a] and O. R. Taylor, Jr.[n] — CHEMICAL DIFFERENCES BETWEEN NATURALLY MATED AND INSTRUMENTALLY INSEMINATED QUEENS — During the past year a part of our research has been concentrated upon answering an often voiced criticism against using extracted hydrocarbons for identification of Africanization and certification of European queens. This criticism keys upon a suspected but never investigated variation in hydrocarbon patterns as the bee ages. We designed and carried out a series of aging experiments which conclusively show that there is variation in unsaturated hydrocarbon patterns from time of emergence from the queen cell to 36 hrs. post-emergence.

A side result from these experiments concerned observations on the development of tergal gland alkenes, compounds found in mature queens which are concentrated on the dorsal side of the abdomen in the area of the tergal glands (Smith and Taylor, *J. Kans. Entomol. Soc.* 1990. 63(3):369-374). These compounds are suspected pheromones.

Our first hypothesis for biological activity of the tergal gland alkenes postulated that they act as close range sex pheromones. This was shown to be false by a conspicuous lack of the compounds on queens up to the normal time for mating (7 - 10 days).

We developed a second hypothesis proposing a role for the compounds which communicated to the workers that the queen was mated and capable of producing eggs and that the operative trigger for the substance production was accumulation of carbon dioxide in the bloodstream resulting from the mating flight. To produce information which would test this hypothesis a group of queens of the same age was needed. One-day-old eggs were grafted into queen cups, placed in a cell building colony, then moved to an incubator where the time of emergence could be noted. After emergence the queens were moved to holding cages in a queen bank. At seven days post-emergence half the queens were treated with carbon dioxide and again at 10 days. The other half of the queens served as the control group. The queens were returned to the queen bank and all were analyzed at day 23. Of the carbon dioxide group 5 out of 8 had produced the tergal gland alkenes, while the virgin controls had 3 out of 5 with the compounds, which indicated that carbon dioxide had no stimulatory effect on the initiation of synthesis ($p > 0.75$). Another experiment compared a group of instrumentally inseminated queens with sister virgins and found no difference in the onset of production of the tergal gland alkenes. We also set-up an experiment where half of a group of sister queens received drone semen and the remaining queens were subjected to the instrumental insemination procedure with insect saline. Again no difference between the two groups was noted.

We had looked at a few naturally mated queens and all less than 23 days post-emergence had the tergal gland alkenes present. An experiment was performed where a group of sister queens was confined to their mating nuclei for 7 days post-emergence, then allowed to free mate. Queens were collected for analysis at 24 hr intervals beginning 48 hrs from the time of mating. All the naturally mated queens exhibited the TG alkenes. Statistical analysis of the data using the Mann-Whitney procedure indicated the instrumentally inseminated queens were not different from the virgin queens ($p > 0.66$), while both groups were significantly different ($p = 0.001$) from the naturally mated queens.

26. Spangler, H.G.,[a] S.L. Buchmann,[a] and S.C. Thoenes[a] — DOES SOUND LAUNCH HONEY BEE SWARMS? — We monitored sounds of swarming bees using an improved detector to study the occurrence of the buzz run (Schwirrlauf). By subjecting bees to loud airborne sound, we attempted to simulate the sound of the buzz run and study its effects. Esch (*Z. vergl. Physiol.* 56:408-411) described the sounds of the buzz run. He found that the sound frequency went from 180 - 250 Hz to 400 - 500 Hz when the buzzing bee contacted another worker. Spangler *et al.* (*Amer. Bee J.* 130:813-814) reported on the occurrence of the buzz run just prior to swarms taking flight. They used an optical detector 1 m away to detect buzz runs occurring within a diameter of about 5.5 cm. They found that a mean of 4.5 buzzes occurred during the period 90-60 sec before flight, 7 between 60 and 30 sec, and 14.5 between 30 and 0 sec. Once a colony started to leave, all detectable bees were airborne within 35.5 sec.

Most of the equipment used for the present study was similar or identical to that used by Spangler, *et al.* The swarm was placed on a bee-tight 24 cm long cone-shaped screen container. The larger end was attached to a 17 cm diameter wood disk. This swarm holder was suspended from an electronic scale with an analog output. The queen and a few workers were placed inside the holder.

The optical detector detected buzz runs occurring over a 5 cm diameter from 5 cm away with greater sensitivity and less interference from flying bees. This allowed detection, not only of twice as many buzz runs prior to flight but also runs occurring after take off began. The data were collected on 2 natural swarms, which repeatedly flew off without their queen and returned. Ten take offs were recorded. A mean of 14.8 buzz runs was produced during the 120-90 sec before the bees be-

gan to take flight, 17.7 between 90 and 60 sec, 19.7 between 60 and 30 sec, and 29.3 from 30 to 0 sec. Weight data showed that once they began to leave, all detectable bees were airborne within 40.7 sec. Bees could be detected producing buzz runs for 25 sec into the take off; a mean of 19.6 buzz runs was detected during that interval.

Next a loud sound was produced that might simulate the signals of the buzz run closely enough to cause the bees to respond, perhaps to take flight. A 7 cm diameter automobile security system horn was attached to the bottom of the screen container with the highest sound intensity directed upward. This horn produced 121 dB (SPL) at 25 cm and considerable vibration with a fundamental frequency of 430 Hz, within the frequency range of buzz runs.

Chart recordings of colony weight variations showed that during each of the five times the horn sounded, one or more "chunks" of bees fell from the swarm cluster. Three times the sound was turned off soon after the bees fell. During the remaining two times, the bees took flight when the sound was left on. During one episode the colony lost a chunk, then after 13 min 29 sec of sound it took flight in 38 sec; and the horn was turned off. In 5 min 36 sec the bees began to return; all had returned after 11 min 28 sec. Spangler (*Amer. Bee J.* 111:92-93) noted that bees subjected to substrate vibration had an increased tendency to swarm (abscond). He attributed this to effects of bees becoming motionless when standing on a vibrating surface. However, it now seems possible that sound plays a more direct role in causing swarms of honey bees to take flight.

27. Stringer, B. A.,[u] L. A. Royce,[u] and P. A. Rossignol[u] — MATING BEHAVIOR OF DRONES IN A CONFINED MATING CHAMBER — Confined mating has been achieved with a limited number of queens in a small domed chamber. Our observations indicate that drones tend to hover vertically a few inches from the sides of the chamber wall and dome. To increase the unit's efficiency, up to 5 queens are suspended simultaneously from a wire carousel 2-6 inches from the top center of the dome.

Drone behavior leading to mating follows set, sequential events. First, drones track or see the queen from a few cm below her. Then they approach the queen and begin to antennate her from the rear or laterally moving to the rear. Following antennation, some drones attempt to mount. At this point, provided the drone continues to beat his wings, he can pull himself into position where the tip of his abdomen meets the tip of the queen's abdomen. If the queen's sting chamber is open, the drone everts his genitalia into the queen and inseminates her.

Studies indicate that pheromones may play an important role in drone attraction to virgin queens. Chloroform extracts of tergites III-V and mandibular glands from 5, 10 and 17 day old virgin queens were applied to lures and compared with extracts of sternites when hung in the unit. Tergite and mandibular extracts were attractive to drones, eliciting similar behavior as that seen toward live virgin queens.

28. Spivak, M.,[w] and M. Gilliam[a] — NEW IDEAS ON THE ROLE OF HYGIENIC BEHAVIOR IN DISEASE RESISTANCE IN HONEY BEES — Hygienic (nest cleaning) behavior in honey bees has been previously correlated with resistance to American foulbrood (Rothenbuhler, 1964, *Anim. Behav.* 12:578-583) and to chalkbrood (Gilliam *et al.*, 1983, *Apidologie* 14: 29-39). Most colonies, however, display intermediate levels of hygienic behavior. Preliminary testing of 50 colonies in Tucson indicated that only seven were highly hygienic (the bees uncapped and removed freeze-killed brood from 200 sealed cells in a comb insert within 48 hours), 27 were intermediate (two to seven days required to clean out the insert), and 16 colonies were non-hygienic (over seven days required).

If rapid removal of dead or diseased brood confers resistance, why does this trait occur in such a low frequency in honey bees? One hypothesis is that hygienic behavior may not be adaptive in another context. Previous research has indicated that behavioral and physiological mechanisms of resistance are not genetically linked (Rothenbuhler, 1956, *J. Econ. Entomol.* 49: 470-475). Thus, the actual expression of the disease would depend primarily on the degree of physiological resistance or susceptibility of the adults and/or larvae in the colony and secondarily on the degree of hygienic behavior displayed by the colony. For example, if bees have no physiological mechanism of resistance to the disease, but uncap and remove diseased brood from cells, they may subsequently infect healthy larvae when feeding them and thus spread the disease. In this case, there may be selection against hygienic behavior.

Experiments are underway to answer the following questions related to hygienic behavior and its correlation with resistance to the chalkbrood pathogen, *Ascosphaera apis*: 1) does the addition of hygienic nurse bees to non-hygienic colonies increase the level of hygienic behavior of the colony; 2) do non-hygienic colonies tend to cap dead and diseased larvae rather than removing them from the cells; 3) are there non-hygienic colonies that are resistant or hygienic colonies that are susceptible to chalkbrood, and what are the mechanisms of physiological resistance to chalkbrood in these colonies?

Queens were reared from selected hygienic and non-hygienic colonies and inseminated with semen from hygienic or non-hygienic lines. Progeny from the two lines (eight hygienic and eight non-hygienic colonies) were tested in observation hives containing approximately 3000 bees to investigate the first two questions. The colonies were later challenged in standard 10 frame hives with the chalkbrood pathogen via contaminated pollen supplements.

Preliminary results indicated that addition of 25-35% hygienic bees to non-hygienic colonies decreased rather than increased the level of hygienic behavior of the colony. When two inserts, one containing live brood and the other freeze-killed brood, with holes poked in the cappings of each insert, simultaneously were presented to hygienic and non-hygienic colonies, the bees recapped 85.6% ± 6.48 (s.d.) and 90.3% ± 7.33 of the live brood, respectively. Hygienic colonies recapped only 2.0% ± 2.85 of the freeze-killed brood, but non-hygienic colonies recapped 48.0% ± 16.7 of it. In general, the degree of hygienic behavior in the assays using freeze-killed brood corresponded closely with the degree of infection after challenge with the pathogen. However, three of the eight non-hygienic colonies had very low infection levels (3, 5, and 7 mummies observed in the center comb during the time of peak infection), and three of the hygienic colonies had high infection levels (16, 22, and 27 mummies in the center comb). These colonies are currently being investigated for differences in their modes of physiological resistance to the disease.

29. Taylor, O. R.,[n] A. Delgado[n] and F. Brizuela.[n] — CORRELATIONS BETWEEN WORKER CELL SIZES AND ALLOZYME FREQUENCIES IN HYBRIDIZING FERAL POPULATIONS OF NEOTROPICAL AFRICAN-EUROPEAN BEES IN NORTHERN MEXICO — In northern Mexico (Linares, Nuevo Leon) we are using genetic markers to follow changes which occur as invading feral African bee populations encounter managed and feral European bees. As part of this study samples were obtained from 200 swarms and analyzed for malate dehydrogenase (MDH) and hexokinase (Hk). In swarms with combs three measures for 10 linear worker cells were recorded. The relationships of allozyme frequencies to mean cell sizes are shown in the table. The frequencies for MDH1 and HK2 were highest for the smallest cell sizes and are similar to those obtained from feral African populations in Costa Rica and Honduras (Taylor *et al. Am. Bee J.* 131). Allelic frequencies for MDH obtained from swarms with the largest

cell sizes (5.3) were similar to those of European bees before the arrival of AHBs. However, the presence of .06 HK2 indicates some African paternity in this class.

A month by month analysis shows a decline in the proportion of swarms producing intermediate cell sizes. This decline is accompanied by large increases (from .36 to .81) in the proportion of colonies with cell sizes of 4.9 and 5.0. Most of these colonies had queens with HK2 and all showed HK2 paternity and are therefore African. A paternity analysis shows a rapid decline in European paternity in these swarms.

Cell size can be used for field identification with some restrictions. No European colonies were found which produced cells measuring 4.9 and only .08 (N = 39) had cells of 5.0. Thus, if all colonies with 4.9 and 5.0 were designated as African the number of false positives would be low. However, in areas with hybridization, because low rates of African paternity do not result in smaller cells, it is not appropriate to consider colonies with large worker cells as European.

Table - Allelic frequencies for MDH and Hk of workers from swarms with combs of different cell sizes

Cell Size	Allelic frequencies MDH			HK		N Col.	Ind.
	1	2	3	1	2		
4.9	.82	.15	.07	.57	.43	29	1262
5.0	.65	.17	.18	.66	.34	24	1044
5.1	.58	.24	.18	.65	.35	37	1606
5.2	.42	.32	.26	.81	.19	21	924
5.3	.23	.35	.42	.94	.06	29	1242

30. Taylor, O. R.," A. Delgado" and F. Brizuela." — IDENTIFICATION OF NEOTROPICAL AFRICAN, HYBRID, AND EUROPEAN HONEY BEES WITH THE USE OF ALLOZYMES — Two polymorphic isozyme systems, malate dehydrogenase (MDH) and hexokinase (HK) differ sufficiently in frequencies between African and European honey bee populations to be useful in identification. The different allelomorphs (forms) for these enzymes can easily be identified with starch gel or cellulose acetate electrophoresis. The procedure for use in identification is to 1) establish the allelic frequencies for managed and feral European bee populations in advance of Africanization and 2) establish allelic frequencies for feral AHBs. Feral AHBs can be identified independently of allozymes by the small size of worker cells (Taylor *et al. Am. Bee J.* 131). 3) Expected frequencies for queens and workers of each genotype for MDH and HK are obtained by expanding the allelic frequencies to give genotypic frequencies for queens and workers (Table). Tests show a close correspondence between observed and expected frequencies in most populations. The Table shows a "worst case" scenario with HK2 of .05 for European bees when, in fact, HK2 is <.01 in all preAfrican populations in northern Mexico and the United States. Also, MDH1 is unrealistically low for an African population.

Table - Expected Worker and Queen Frequencies for EUROPEAN and Neotropical AFRICAN BEE populations*

MDH	HK 1/1		1/2		2/2		Totals	
	E	A	E	A	E	A	E	A
1/1	.036	.106	.004	.205	<.001	.098	.040	.409
1/2	.083	.080	.008	.153	<.001	.074	.091	.307
1/3	.206	.040	.022	.077	.001	.037	.229	.154
2/2	.048	.015	.005	.029	<.001	.014	.053	.058
2/3	.236	.015	.025	.029	.001	.014	.262	.058
3/3	.293	.004	.031	.007	.001	.003	.325	.014
Totals	.902	.260	.095	.500	.005	.240	1.00	1.00

*Based on these frequencies:

	MDH 1	2	3	HK 1	2
European	.20	.23	.57	.95	.05
African	.64	.24	.12	.51	.49

Expected frequencies are sufficiently different for many queen genotypes to allow identification. A simple paternity analysis of 24 workers reveals whether a queen has mated with drones of the most distinctive African genotype (MDH1 + HK2). This method has been tested on over 500 colonies. Misidentifications are <.01. HK2 is present in .58 to .74 of the African queens and in >.99 of the African colonies. Because HK2 is <.01 in European bee populations, the appearance of MDH1 + HK2 paternity is indicative of hybridization.

This identification method is rapid, accurate and inexpensive. It can be used to detect low levels of hybridization and should prove valuable in validating the purity of breeding stocks.

31. Taylor, O. R., A. Delgado and F. Brizuela — RAPID LOSS OF EUROPEAN TRAITS FROM FERAL NEOTROPICAL AFRICAN HONEY BEE POPULATIONS IN MEXICO — A recent analysis of hybridization between European and African bees adds to the confusion concerning the genetic attributes of Neotropical African bees in Mexico (Rinderer *et al. Science* 253:309-311). In this study, samples were obtained only from colonies in apiaries two years after (Sept. 1989) the establishment of African bees in the area (April-October 1987) and before this population reached its peak abundance. Although hybridization was widespread in the apiaries, particular emphasis was given to a number of colonies which originated from feral African swarms.

Many of these showed evidence of backcrossing with European drones. This is the pattern seen in apiaries at the onset of hybridization, but it is a temporary condition. Nevertheless, the results were interpreted by the authors and widely cited in the media as indicative that African bee populations in Mexico are becoming "Europeanized". This is not true. Even in northeastern Mexico (Table) where European bees have a substantial feral population (Rubink *et al. J. Kans. Entomol. Soc.* 63:288-297) the feral African population shows little Europeanization.

We are using genetic markers to track the processes and consequences of hybridization in both feral and managed populations of hybridizing African and European bees.

The following trends are evident: (1) Apiary populations become similar genetically to the feral African populations. The rates of convergence are slowest in populations at the highest elevations, those with the largest populations of managed Eu-

Table - Paternal allelic frequencies for MDH and HK from feral honey bee populations in Mexico and Central America

Location	Date	Months Since Detection	N's Col.	Ind.	Allelic Frequencies MDH 1	2	3	HK 1	2	Est. N. Drones*
Merida, Yuc.**	Jan. '87	-9	15	353	.26	.17	.56	.93	.07	158
Linares, N.L.	Mar. '91	9	57	1246	.72	.14	.14	.66	.34	599
Merida, Yuc.	July '90	33	19***	424	.74	.14	.12	.64	.36	200
Tapachula, Chiap.	Jan. '91	40	49	1034	.79	.13	.08	.61	.39	515
Honduras	June '91	76	18	427	.84	.11	.05	.66	.34	189
Costa Rica	June '91	90	29	684	.86	.10	.04	.55	.45	305

*Assumes 10.5 matings/queen.

**Managed European bees before detection of AHB's. HK2 is <.01 in EHB populations in northern Mexico and the United States.

***7/19 (.37) samples lacked the most common type of European drone paternity (MDH3 + HK1).

ropean bees and those where beekeepers practiced requeening. (2) At the front (first six months) of the invading feral African bee populations, bees in swarms are larger, produce a wide range of worker cell sizes, and show greater European paternity than is found in all subsequent sampling periods. (3) Long-term trends in the changes in allelic frequencies following detection are shown for several locations in Mexico and Central America (Table). The frequencies for the allelomorphs (forms) of malic dehydrogenase (MDH) and hexokinase (HK) are similar to those from many populations in Brazil (Del Lama *et al. Apidologie* 21:271-280). (4) The sizes of worker bees and of worker cells in combs declines in concert with these genetic changes (Boreham & Roubik, *Bull. Entomol. Soc. Am.* 33:34-38, Taylor *et al. Am. Bee J.* 131).

32. Thoenes, S. C.[h] – BUMBLE BEE (*BOMBUS SONORUS*) INVASION OF HONEY BEE COLONIES – Little information is available on how honey bee colonies affect populations of other native species of bees. Most of the studies concentrate on the aspect of competition, but this study shows the physical presence of honey bee colonies moved into a native environment can also affect native bee populations.

In spring 1991, 40 colonies of honey bees were moved to a location 57 km north of Tucson, AZ. Thirty-six of the colonies were equipped with a Todd dead bee trap. An active bumble colony was noticed in mid-June ca. 100 meters from the apiary.

From July 5 through August 2, a total of 147 bumble bee workers were found in the dead bee traps on the front of the honey bee colonies (see Table). In addition five *Xylocopa californica* were found. On two separate instances a solitary bumble bee was seen flying to forage on the mesquite tree in the middle of the apiary. Each bumble bee then moved to the front of a nearby honey bee colony, back to the mesquite, back to the colony, and then out of the area.

All of the bumble bees were found during the period when the honey bee colonies were evaporating large amounts of water off newly collected honey. Apparently the foraging bumble bees were attracted to the smell venting from the colonies and made the mistake of landing on or entering the colony. Thus the physical presence of honey bee colonies resulted in the destruction of many bumble bee workers and a dead bumble bee colony by September 1.

Table – Chronological listing of bumble bees found in honey bee colonies at a site 57 km north of Tucson, AZ.

Date	# Honey Bee Colonies Involved	# Dead Bumble Bees
July 5	1	3
July 9	21	70
July 18	10	48
August 2	9	26

33. Thoenes, S. C.,[h] J. O. Schmidt,[a] and S. L. Buchmann[a] – HOW DIET AFFECTS HONEY BEE REPRODUCTIVE SWARMING – Almost no research exists on how dietary factors influence the production and timing of reproductive swarming in honey bees. The main difficulty in conducting such studies with honey bees is that free flight is essential to the successful production of a reproductive swarm, and thus any dietary manipulation is difficult because pollen and nectar are available from the surrounding environment of the study site.

Several long-term studies from the Tucson, Arizona area have been completed which compare swarming phenology, pollen quantity, pollen quality, and nectar collection. Swarm phenology was positively correlated to both pollen factors and negatively related to nectar collection.

A site 57 km northwest of Tucson, Arizona was selected as ideal for the pollen dietary manipulations. The site is characterized as natural desert and isolated from human habitation. Previous pollen trapping studies at this site revealed nonexistent or sparse pollen availability from January through April. All colonies consisted of one deep and one shallow Langstroth boxes. Four colonies were dedicated solely to trap pollen. Thirty colonies with equal amounts of brood, stored honey, and adult bees were divided into three treatment groups of 10 colonies each. Treatment consisted of high protein diet, low protein diet, and no added diet.

Results show no difference in the number of colonies that produced reproductive swarms, but considerable differences occurred in the timing of when each treatment swarmed. The high protein treatment swarmed first, followed by the low protein treatment, and the controls swarmed last. We conclude that protein content in spring time diets controls when honey bees can produce reproductive swarms.

34. Webster, T. C.[x] and D. J. Callaway[x] – MEASURING SMALL POPULATIONS OF VARROA IN HONEY BEE COLONIES – Three techniques for measuring small populations of *Varroa jacobsoni* in standard size honey bee hives were compared. Two are widely adopted by beekeepers and apiary inspectors: the "ether roll" in which a random sample of adult bees are agitated inside a glass jar with ether from an aerosol can; and the use of fluvalinate strips suspended inside the hive, with a sticky board to collect mites on the bottom board.

A third technique, a test for mites on newly emerged bees, may also have value. A frame of capped brood is placed, without adult bees, in a plastic bag over night. The following day, all emerged bees are placed in a glass jar with a small amount of vegetable oil, and agitated. Ether, as used for the ether roll test, may be used instead of vegetable oil. Mites on the bees stick to the sides of the glass jar and are easily seen.

The test of mites on emerging bees is considerably more sensitive than the ether roll test (see table), although it is less sensitive than the fluvalinate strip method. It is less time consuming, less disruptive to the hive, and less expensive than the test with fluvalinate strips. Thus the test for Varroa mites on emerging bees may be preferable under some circumstances.

Table – Number of mites/100 bees from Varroa-infested hives.

Method	Hive					
	1	2	3	4	5	6
Ether roll	0.8	0.2	1.2	0.5	9.9	0
Emerging bee	2.0	5.8	4.2	10.6	49.0	5.3

35. Wenner, A. M.,[p] D. E. Meade,[p] and J. E. Alcock[y] – FERAL BEE COLONY REMOVAL ON SANTA CRUZ ISLAND: TECHNIQUES AND PROGRESS – To help *The Nature Conservancy* restore ecological balance in the previously overgrazed *Channel Islands National Park* (Wenner, *Am. Bee J.* 129:808-809), feral honey bees initially introduced before 1880 are being eliminated on Santa Cruz Island. We have now located 125 colonies. The eastern half of the island, in which nearly 80 colonies have been found, is now mostly clear of honey bees. This setting provides a rare opportunity to study the foraging ecology and recruitment of honey bees in an undisturbed situation, as well as permitting studies of pollination by native bees versus introduced bees (the latter by Robbin Thorp, University of California, Davis).

To find many colonies in a short time, we have relied on a 1937 von Frisch odor-search model of recruitment (e.g., von Frisch, *Annual Report of the Smithsonian Institution for the Year Ended June 30, 1938*, pp. 423-431). Friesen clarified that process in 1973 (*Biol. Bull.* 144:107-131). Wenner and Wells updated the model in 1990 (Chap. 5 and Excursus OS in *Anat-*

omy of a Controversy: The Question of a "Language" Among Bees, Columbia Univ. Press).

Early on in our search we abandoned use of the centuries old "bee box" technique (*e.g.*, Visscher and Seeley, *Am. Bee J.* 129:536-539). Instead, our success has been due to exploitation of wind directions. We gather bees from blossoms, massively expose them to the odor of our food source, and establish bee lines to gain bearings. By multiplying 150 times the minutes required for an average complete round trip and subtracting 500 from the result, we estimate the distance to each feral colony in meters (Wenner, *Am. Bee J.* 129:808-809). We then study wind patterns in the area and set up an auxilliary station closer to and upwind from the approximate location of each colony.

If we have placed our auxillary station in an appropriate location, recruitment is immediate and rapid, most often by contrast with recruitment to the primary station. If we initially capture only a few bees for a primary station located more than 300m downwind from their colony, recruitment of others to that station fails for reasons outlined in Friesen (*Biol. Bull.* 144:107-131). We also note that recruits arrive in a zigzag flight only from far downwind, as elaborated upon more recently by Rosin (*Am. Bee J.* 131:525-526).

36. Wenner, A. M.,[p] D. E. Meade,[p] and L. J. Friesen[z] — DISTANCES HONEY BEES RANGE FROM THEIR COLONIES: A UNIFYING CONCEPT — Whereas individual bees may visit particular flower patches, the average distance a colony ranges often fits a mathematical pattern, based on the logarithm of the distances involved. Four such lognormal distributions have been identified to date: 1) distances foragers travel to food sources, 2) distances bees travel to water, 3) distribution of searching bees at test stations in field experiments, and 4) distances swarms relocate from their parent colonies.

Two techniques used to estimate the distances that foragers travel to food sources are: 1) interpreting forager waggle dances in the colony (see esp. Spangler, *Am. Bee J.*, 130:813) and 2) magnetically retrieving metal tags from returning foragers (Gary, *et al., Env. Ent.* 7:233-240). By studying waggle dances, Vergara found that African bees in Panama foraged an average of 1300 m from their colony (see figure). Mid-July results from a 1981 study at Cornell (Visscher and Seeley, *Ecology*. 63:1790-1801) also yielded a lognormal distribution with an average distance of 1300m. By contrast, the hybrid bees in the Gary *et al.* study travelled an average of only 75m within that study area.

On Santa Cruz Island (Wenner, *et al., Am. Bee J.*, 130:818), about 80% of 53 of the colonies studied collected water at an average distance of only 160m, with the other 20% travelling further during the severe drought at that time.

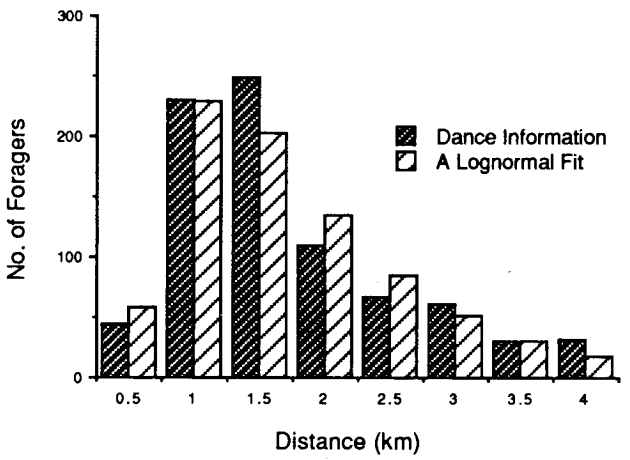


Figure - Tallies of waggle dance maneuvers in African bee colonies in Panama yielded lognormal distributions (data courtesy of Carlos Vergara, as in Fig. 2.24 of Roubik, *Ecology and Natural History of Tropical Bees*, 1989).

Experiments that tested recruited bees' ability to find stations placed at various distances from their colonies also yielded lognormal patterns whenever those stations were made more nearly equal to one another (Wenner, Meade, and Friesen, *Am. Zool.*, submitted), including results obtained in the mechanical bee experiments (Michelsen, *et al. Naturwissenschaften*, 76:277-280).

Swarms collectively likewise moved a lognormal distance from their parent colonies. Lindauer in 1955 (*Z. Vergl. Physiol.* 37:263-324), Seeley and Morse in 1977 (*Psyche*, 84:199-209), and Schmidt and Thoenes in 1990 (*Am. Bee J.* 130:811) all obtained results with that pattern. For data combined in the first two cases, swarms moved an average of 830m.

37. Wilson, W.T.[aa] and A.M. Collins[aa] — SPRING APPLICATIONS OF FORMIC ACID FOR CONTROL OF *ACARAPIS WOODI*[ee] — Hoppe *et al.* (*Am. Bee J.* 129:739-742) described the treatment of *Apis mellifera* adults with fumes of formic acid (FA) for control of *Acarapis woodi*. Earlier studies in India by Garg *et al.* (*Am. Bee J.* 124:736-738) demonstrated that FA killed parasitic bee mites. Recently, Garza *et al.* (Am. Bee J. 130: 801) reported excellent control of tracheal mites following four 20-ml applications of FA to colonies in Mexico. In samples from some colonies, 100% of adult mites were killed. The impact of FA fumes on adult bees and brood was minimal even though ambient temperatures were high (ca.30°C). However, some beekeepers in the U.S. have experienced serious adult bee and brood mortality when FA was applied during hot weather. Many of the treatment programs followed in Europe require FA applications at 4-day intervals. Such intense programs are not cost effective for U.S. beekeepers. Therefore, our goals were to increase dosage, decrease the number of applications and increase treatment interval.

In southern Oklahoma during February and March 1991, weekly 10- or 20-ml and biweekly 40-ml doses of FA (65%) were given on paper toweling in the top of most hives (n = 15, 15 & 9, respectively) for durations of 1 to 5 weeks. A few colonies received FA on paper towels administered in the broodnest (n = 3), on the bottom board (n = 3) or in all 3 positions (n = 3). Similar FA tests were conducted during April & May in Texas (n = 40) and in May & June in Iowa (n = 70) with 40 ml in the top of each colony on a weekly or biweekly schedule for 3 applications.

Data from tests in the 3 states showed that 3 applications of 40 ml of FA at weekly or biweekly intervals gave dependable mite control. Following the 3rd treatment, >90% of the adult mites were dead, while in untreated controls <17% had died. Twenty-ml treatments gave satisfactory control of the adult mite, but 4 or 5 weekly applications were necessary to achieve the 90% level. Treatment with 10 ml yielded inadequate mite control. There was no apparent damage to adult or immature bees following the FA treatments. Unfortunately, immature mites, and especially eggs, did not appear to be affected by FA.

Formic acid vaporized readily during cool spring weather (10 - 20°C) and a 40-ml dose gave excellent tracheal mite control. The mite was not eliminated, but populations were reduced to non-economic levels. Another benefit from using FA is the low cost (<$0.50) per treatment. However, the applicator should exercise caution during colony treatment. FA is caustic and may cause serious bodily injury unless adequate safety precautions are taken.

38. Wilson, W.T.,[aa] W.J. Sames IV,[bb] A.M. Collins,[aa] and M. Ellis[cc] — DEPOPULATION OF HONEY BEE COLONIES USING ETOC, A SYNTHETIC PYRETHROID[ee,ff] — For many years, the beekeeping industry has tried to protect honey bees (*Apis mellifera*) from damaging exposure to pesticides.

However, there are situations that require the killing of honey bees. Sames, *et al.* (*Am. Bee J.*, 130:810) pointed out the need to control Africanized bee swarms in the U.S. He demonstrated that detergent sprays and pyrethroid aerosols were highly effective in killing bees (Sames, *et al., Southwest. Entomol.* 16:19-24). In the current study, field colonies of honey bees were treated with ETOC or resmethrin aerosols. ETOC is an experimental compound produced by McLaughlin, Gormley, King Co. (MGK). MGK donated the ETOC.

Research during autumn 1990 at the USDA-ARS Honey Bee unit in Weslaco, TX studied efficacy, knockdown and time to kill >90% of the adult worker bees in 4-frame nucleus colonies with a 1-5 second aerosol spray of ETOC or resmethrin. Ambient day temperatures were 26 to 32°C. In 2 series, a total of 10 nucs were treated with 1% ETOC, 3 nucs with 1% resmethrin and 5 controls. The aerosols were introduced either in the top of the hive or in the entrance and the entrance was plugged with burlap. Each colony contained ca. 680 gm of adult bees. The adult population was checked 3 to 5 min. following chemical exposure, and again after 5 and 24 hrs.

Both ETOC and resmethrin killed adult bees quickly. Regardless of whether the nuclei were treated for 1, 3 or 5 sec. in the top or bottom, both chemicals knocked down 90 to 99% of the bees within 5 min. and all were dead within 5 hrs. Exposed bees "buzzed" for several seconds, but did not attempt to abandon the hives. Overall, the knockdown time for ETOC appeared to be slightly faster than for resmethrin, however they were similar in efficacy. When examined 24 hrs. later, often there were a few (ca. 50) newly emerged bees on the combs with no sign of poisoning. This indicated that the 2 pyrethroids degraded rapidly with little or no toxic residue remaining on the combs. To substantiate low residual toxicity, 1 comb from each treated nuc was placed in an untreated nuc on the 2nd day with no bee mortality after 24 hrs. Bees utilized the treated combs in a normal manner. At least for ETOC, after killing a colony, the combs are probably reusable.

When ETOC aerosol was applied to 5 standard-size colonies (each with 2 deep hive bodies) during warm weather, the results were basically the same as with the nuclei. A spray of 5 sec. in the top of a hive knocked the bee population down in <5 min. and 95% were dead after 10 min. However, when ETOC was applied to 3 standard-size colonies in Nebraska during cold (ca. 0-4°C) weather, knockdown was nearly instantaneous for bees contacted by aerosol on the outer layer of the cluster, but there was inadequate penetration. Thus, repeated treatments were needed to kill all bees in winter clusters.

39. Yakobson, B. A.,[dd] D. Elad,[dd] K. Rosental,[dd] I. Kamer,[dd] I. Slovecky,[dd] and H. Efrat[dd] – A RECENT CHALKBROOD OUTBREAK IN ISRAEL: ATTEMPTS AT THERAPEUTIC INTERVENTION – Chalkbrood (Ascophaeromycosis) was reported for the first time in Israel in 1984 (Yakobson *et al, Israel J. Vet. Med.* 43(1): 28-33). Between 1984-1990, the disease had a very low incidence. In some apiaries, the infection rate suddenly rose to a high level in the summer and autumn of 1990, and there was no apparent link between the highly infected apiaries. In the following spring, signs of the disease were detected in nearly every apiary in the country. The negative effect of the disease is not only the weakness of the infected bee colonies, but also the inability of beekeepers to divide their "healthy" or strong colonies to renew their stock. This splitting of bee colonies causes the latent infection to change into the diseases's active form. The result was an overall decline of 10-15% in honey production.

In a single apiary with 168 colonies, 59 hives with clinical signs of chalkbrood produced 37% less honey (23.3 kg/hive) than did 109 hives that showed no clinical signs of chalkbrood (36.8 kg/hive).

This outbreak could have been the result of one or, more probably, the interaction of several environmental factors, such as: (1) the possible appearance of a novel, more virulent strain of *Ascosphaera apis*. However our examination, mainly morphological, does not support this suggestion. (2) the consequences of a widespread *Nosema apis* infestation and the increased use of fumagillin (Parbucki & Gorski. 1987, *Proc. Apimondia*, 305-310). (3) a possible change in the genetic makeup of local bee populations due to large scale importation of queens from Australia. Australian stock may have had a greater susceptibility to *A. apis* infection (Gilliam *et al.*, 1983, *Apidology* 14: 29-39). A similar incidence of chalkbrood in apiaries which did not import any queens from abroad seems to exclude this possibility. (4) the effect of the stress caused by Varroa mite epidemiology and the acaricides used to control it. We did not consider this possibility very likely as no chalkbrood was observed during the height of Varroa infestation in 1986-1987. (5) a result of mixed infection (Mehr *et al.* 1976, *Am. Bee J.* 116:266-268) of *A. apis* and sacbrood virus which is known to be spread by the Varroa mite (Ball, 1986, Starnberg, W. Germany) and acts as a co-factor. This may increase the severity of the disease.

Nystatin and trichlorinated isocyanutic acid (Tawars, 1986, *Proc. Apimondia* 274-278) were used in experimental trials to control infection, but no therapeutic effect was observed. Another fungicide, enilconasol (Janssen), was tested by several methods of application but also without any marked effect.

An as yet unclassified bacterium was found which was capable, at least in vitro, of suppressing *A. apis* growth, so biological control of chalkbrood may become possible.

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