BARD

FINAL REPORT
PROJECT NO. IS-2684-96

Lek Behavior of Mediterranean Fruit Flies: An Experimental Analysis

B. Yuval, T.E. Shelly

2000

BARD Final Scientific Report
(Cover Page)

Date of Submission of the report: July 2000

BARD Project Number: IS-2684-96R

Evaluating Panel: Plant Protection

Project Title: Lek Behavior of Mediterranean Fruit Flies: An Experimental Analysis

Address & e-mail of Investigators and Institutions:

Investigators	Institutions
Principal Investigator:	
Boaz Yuval (yuval@agri.huji.ac.il)	Hebrew University
Todd E. Shelly (tshelly@lava.net)	University of Hawaii
Cooperating Investigators:	

Continuation of (Related to) Previous BARD Project:
☐Yes xNo Number:

Keywords *not* appearing in the title and in order of importance. Avoid abbreviations.
Sterile Insect Technique, Sexual Behavior, Insect Ecology

Abbreviations used in the report, in alphabetical order:

Budget: IS: $ 151,000 US: $ 149,000 Total: $ 300,000

Signature
Principal Investigator

Dr. ERAN VARDI
DIRECTOR
AUTHORITY FOR RESEARCH AND DEVELOPMENT
Signature
Research Authority, Principal Institution

Abstract

The Mediterranean fruit fly, *Ceratitis capitata* (Diptera: Tephritidae), is a ubiquitous pest of fruit trees, causing significant economic damage both in the U.S. and in Israel. Control efforts in the future will rely heavily on the sterile insect technique (SIT). Success of such operations hinges on the competitive ability of released males. The mating system of the medfly is based on leks. These are aggregations of sexually signaling males that attract females (who then select and copulate a courting male). A major component of male competitiveness is their ability to join existing leks or establish leks that are attractive to wild females. Accordingly, we identified leks and the behaviors associated with them as critical for the success of SIT operations. The objectives of this proposal were to determine 1. what makes a good lek site, 2. what are the energetic costs of lekking, 3. how females choose leks, and finally 4. whether the copulatory success of sterile males may be manipulated by particular pre-release diets and judicious spatial dispersal.

We established that males choose lek sites according to their spatial location and phenological status, that they avoid predators, and within the lek tree choose the perch that affords a compromise between optimal signalling, micro-climatic conditions and predation risk (Kaspi & Yuval 1999a&b; Field et al 2000; Kaspi & Yuval submitted). We were able to show that leks are exclusive, and that only males with adequate protein and carbohydrate reserves can participate (Yuval et al 1998; Kaspi et al 2000; Shelly et al 2000). We determined that females prefer leks formed by protein fed, sexually experienced males (Shelly 2000). Finally, we demonstrated that adding protein to the diet of sterile males significantly enhances their probability of participating in leks and copulating wild females (Kaspi & Yuval 2000).

Publication Summary (numbers)

	Joint IS/US authorship	US Authors only	Israeli Authors only	Total
Refereed (published, in press, accepted)		3	7	10
Submitted, in review, in preparation	1	1	1	3
Invited review papers		1	1	2
Book chapters			1	1
Books				
Master theses				
Ph.D. theses			1	1
Abstracts				
Not refereed (proceedings, reports, etc.)				

Patent Summary (numbers)
(not relevant)

Cooperation Summary (numbers)

	From US to Israel	From Israel to US	Together, elsewhere	Total
Visits/Meetings		2	2	4
Sabbaticals				
Postdoctorates				

- **Cooperation**, briefly explain whether synergistic, complementary or supportive.

Working independently on different aspects of medfly lek behavior, and jointly on the same questions in different medfly habitats, we communicated at least weekly during the project. Our cooperation,both synergistic and complementary, has been extremely satisfying.

Acheivements:

The following significant findings have emerged during this project:

1. males choose lek sites according to their spatial location and phenological status, that they avoid predators, and within the lek tree choose the perch that affords a compromise between optimal signalling, micro-climatic conditions and predation risk near female oviposition sites
2. Male diet, in particular protein feeding, significantly enhances the probability that males will a. join a lek; b. emit pheromone in a lek; c. copulate; d. transfer sperm and e. inhibit the female from remating.
3. Females in the field are attracted to males that had previously copulated in the laboratory.
4. Sterile males fed on protein or exposed to trimedlure prior to release are more likely to copulate females in leks than sugar fed or olfactorily naive males.

a. Significance of main scientific achievements or innovations:

On the level of basic science, our research has provided the first comprehensive account of lekking in an insect. Spatially, we analysed leks on 2 scales, taking into consideration hosts, conspecifics, microclimate and predators. We also analysed energetic aspects of lek behavior, showing a significant link between male foraging ability and reproductive success. In addition, we developed novel tools to study yhe attraction of females to leks in the field, thus being able to manipulate the composition of leks and monitor female preferences.

On the applied level, we have introduced a new approach to increasing the effectiveness of the sterile male technique. We have shown that manipulation of sterile males (primarily via their diet and olfactory environment), during the post teneral period has the potential to make this technique more effective.

b. Potential for application and benefits:

Our results have immediate to mid-range application potential, though more research is required to balance the positive effects of protein diet on copulatory success, with the negative effects of increased mortality .

Tens of millions of US$ are spent rearing and purchasing sterile flies each year, by control programs in California, Florida, Central America and Israel. As the mating efficiency of sterile flies increases, it will be possible to reduce the numbers released. As medfly invasions continue, we anticipate that similar numbers may be released, over greater areas, with increased efficiency and reduced cost..

c. Other benefits:

Our work has been very well received by the community of applied biologists working in the field of medfly control by SIT. The importance of post teneral diet and behavior is being incorporated into quality control measures instituted by sterile fly growers and end users.

d. Details of cooperation:

Cooperation has been complimentary and synergistic. Cooperation has consisted of frequent and detailed consultations via e-mail. In addition a complementary experiment examining female attraction to leks was implemented, with very favorable results. P.I.'s were both invited to 2 week long international symposia on medfly sexual behavior (in Guatemala and Vienna) and were able to discuss the projects progress and future at length. In year three Yuval & Kaspi spent 2 weeks performing field work with Shelly in Hawaii. As a result, we were able to compare the behavior of the medfly in Israel and Hawaii, and produce a synthesis of lek behavior and recommendations for improving its control via the sterile insect technique.

Section 2: Body of the Report

Background, scientific and agricultural relevance of the project

Many species of fruit flies (Diptera: Tephritidae) are agricultural pests that exert a significant economic burden on fruit growers The most notorious member of this family is the widely distributed Mediterranean fruit fly, *Ceratitis capitata* (medfly), that can attack over 200 species of fruit. Various approaches are adopted to curb the populations of this ubiquitous and invasive pest. These vary from the rather crude spraying of insecticides to sophisticated programs of integrated pest management, which may combine lures, cultural practices and the release of mass reared sterile males.

The Sterile Insect Technique (henceforward SIT) is probably the most promising control approach for the future. It is axiomatic to the SIT approach that success depends on the ability of released males to locate and inseminate wild females. Currently, applications of SIT rely on massive rearing of males and saturating the treated area with sterile individuals. This overflooding, though expensive, is necessary because of the relatively low competitive ability of the sterile males. Consequently, improving the competitive ability of released sterile males will greatly enhance the effectiveness of control programs. Sterile insect release programs make considerable efforts to control the quality of male performance but these efforts are limited by the lack of information on factors regulating male success. This project caters to this need by focusing on one critical component of the medfly mating system- the male aggregations (leks) where most copulations take place. We identified lek sites- and the behavior of males and females associated with them- as the critical parameters in understanding the mating system of the medfly as a significant link between lekking and male reproductive success has been demonstrated repeatedly. Thus, males released in SIT programs must be able to find and join existing leks (or establish such sites themselves), attract females that approach the lek, and finally copulate them. Accordingly, our objectives, in a coordinated series of comparative and complementary observations and experiments were:

1. To establish empirically the properties of lek sites in nature,
2. To determine quantitatively the energetic costs of lekking and the effects of post-teneral nutrition on copulatory success of lekking males,
3. To experimentally analyse how females choose lek sites.

Finally, integrating our results, to establish

4. Whether copulatory success of lekking sterile male flies may be enhanced by specific pre-release diets and judicious dispersal strategies.

Methodologies and Materials

Working with wild flies from Israel and Hawaii, long established laboratory strains and "wildish" flies we reared by a novel method (Kaspi et al. submitted), our experiments combined the following components: Field observations, caged-host studies, field experiments and biochemical analyses. Full details are provided in our published papers.

Results

a. Objective 1 - To establish empirically the properties of lek sites in nature

a.1. Lek site selection (Kaspi & Yuval 1999a)

In field cage experiments we examined lek site selection as influenced by presence of fruit and presence of conspecific flies. Males were allowed to choose between artificial trees containing fruit and trees without fruit. Males showed a significant preference for the tree baited with fruit. To determine which fruit- related stimuli were important, males were allowed to choose between visual and/or olfactory fruit-derived stimuli. Males preferred to lek on trees presenting both stimuli. To determine the influence of conspecifics on lek site selection, males were allowed to choose between trees containing male and/or female conspecific flies. The presence of pheromone-emitting males was preferred. In addition, both in the field and on artificial trees, we examined the effect of leaf size on lek site selection. In the field, males preferred to lek on leaves of a particular size. Moreover, leaf integrity was important, as males tended to select undamaged leaves as lek sites. In field cage experiments, males consistently chose to lek on trees containing the largest leaves. We conclude that choice of lek site is influenced by the presence of fruit and of other lekking males, while leaf size and integrity determine male location within a lek.

a.2. Male location within a lek (Kaspi & Yuval 1999b)

In this study we tested the following hypotheses: a. Fly location depends on microclimate and illumination. b. Larger and heavier males occupy preferred locations in leks. Accordingly, systematic quantitative observations of diel three-dimensional (3-D) locations of lekking *C. capitata* males were performed in field and field cage studies. We found that: Fly locations varied significantly during the activity hours. Medflies were generally found calling from the highest and most exterior locations during early morning and late afternoon hours. During the hottest hours (12 a.m. -3 p.m.) flies moved progressively towards lower locations within the tree canopy, and moved

to interior locations from 10 a.m. - 3 p.m. Fly location (from 10 a.m. to 5 p.m.), was correlated with the azimuth of the sun. However, the mean azimuth range of fly location was limited to 85^{O} (59.45^{O}-143.94^{O}). Both in the field and in the field cage the temperature, relative humidity and light intensity beneath the leaves on which the males perched, were closer to the microclimate beneath fully shaded leaves than to microclimates beneath leaves exposed to direct sunlight. We conclude that male medflies occupy locations which confer suitable microclimates for calling and copulating, and suitable locations within the canopy that provide protection from predators, wind, direct sunlight, and water loss.

a.3. Patterns of male and female settlement within canopies (Shelly, unpublished)

In a field cage we put males in cups (artificial leks) on a tree, released females, and scored female landings on cups in 1) high vs. low sites, 2) edge vs. center sites, and 3) sparse leaf vs. dense leaf sites. In all cases, we tried to keep all other factors the same except the one under consideration. Then, we did a separate set of experiments where we released males only and scored their presence on samples of leaves in sites where cups were. Results show congruence between male and female preferences.

1) Height above ground - we found male calling and female sightings were same at low-high sites; also, male settlement was similar between high-low leaves.
2) Sparse-dense - male calling was unaffected by leaf density, but female sightings and male settlement were higher in dense areas.
3) Edge-center - male calling unaffected by edge-center but female sightings and male settlement were higher in center.

a.4. Predators affect lek site selection (Kaspi & Yuval, submitted)

In field cage choice experiments we examined how the presence of various animals, most of them common predators in fruit plantations in Israel, influences lek site selection. Male flies were allowed to choose between artificial trees containing predators and trees without predators, and their predator avoidance behavior was determined. Males did not avoid trees baited with the mantid *Sphodromantis viridis*, the ant *Cataglyphis niger*, the salticid spider *Plexippus paykulli*, the reptile *Agama stellio*, or the hornet *Vespa orientalis*. In contrast, trees containing the wasp *Vespula germanica*, or the honeybee *Apis melifera* (although *Apis* is not a predatory insect) were avoided in the choice of a lek site by medfly males. However, when the *V. germanica* were wingless or

desiccated their presence did not affect lek site selection. We discuss these results in relation to predator avoidance behavior and the predator-prey arms race model.

b. Objective 2: To determine quantitatively the energetic costs of lekking, and the effects of post-teneral nutrition on copulatory success of lekking males

b.1. Protein feeding increases the probability that males will join leks and subsequently copulate (Yuval et al 1998; Kaspi et al. 2000).

We tested the hypothesis that in male medflies, who invest considerable energy in sexual displays and courtship, foraging successfully for food affects their subsequent performance and copulatory success in leks. We investigated the interactions between individual size and diet on initiation of lekking behavior and copulatory success of male Mediterranean fruit flies. We found that protein-fed males were heavier, and contained more protein and less lipid reserves than sugar-fed males. Protein-fed males were more likely to emit pheromone in leks, and consequently were more likely to copulate than sugar-fed males. Furthermore, protein-fed males tended to start calling earlier than their nutritionally deprived competitors. Though size was not related to initiation of lek behavior, large males were more likely to copulate than small males. Among protein-fed males, large individuals tended to mate earlier than smaller ones. We suggest that generally, in lek mating systems, where a considerable investment of time and energy is required by males, foraging successfully for nutritional resources prior to engaging in territorial or courtship behavior is essential for reproductive success.

c. Objective 3: To experimentally analyse how females choose lek sites

c.1. Pheromone calling by lekking Mediterranean fruit flies: do females distinguish advertisement signals on the basis of male mating success? (Shelly, in press)

The purpose of this study was to test whether female medflies distinguished between the advertisement (pheromonal) signals of males with known low vs. high mating success. In laboratory trials, the observed distribution of matings among males differed significantly from that expected by chance, owing primarily to the higher than expected numbers of individuals with low (mated 0-1 days over 6 consecutive observation days) or high (mated 4 or more days) mating scores. [In reference to these scores, these two groups are termed "low" and "high" maters, respectively.] In the field, greater numbers of female sightings were made at artificial leks of high maters than low maters. This result apparently reflected a greater calling propensity among high maters. Slopes of female sightings vs. calling level did not differ significantly between leks of low and high maters,

suggesting that the observed relationship between calling activity and female sightings was independent of male mating status. Following the same protocol, a second experiment examined whether males used the signals of conspecific males to locate lek sites and, if so, whether signal attractiveness varied with male mating ability. Attraction of males to calling conspecifics was far weaker than that observed for females, and over 5 different trials a total of only 7 male sightings were made at any of the established leks.

c.2. Effects of male diet on female attraction. (Shelly et al. 2000)

In this study, we compared the levels of pheromone-calling and female attraction between groups of sugar-fed males vs. groups of protein +sugar -fed males at two spatial scales. First, we compared groups on different plants in the field (between-plant comparisons). Subsequently, we compared groups on the same plant using a field-tent (within-plant comparisons). All insects used in this study were from a 5-year old, mass-reared laboratory strain.

For the between-plant comparisons, tests were conducted in a coffee field, and four artificial leks - two with sugar-fed males and two with protein + sugar-fed males - were established at equal distances from a central female release point. Each male aggregation consisted of four transparent cups (ends covered with screen mesh), with each cup containing six males (i.e., 24 males per lek). Cups were suspended in the canopy of individual coffee plants, and 400 virgin females were released. Male groups were monitored every five minutes over a 90-minute test period, and the numbers of calling males and perching females were noted. Tests were run on 16 different days (yielding N = 32 for each lek type). We found no difference in the calling level between the two lek types, and an average of 7 males were calling per observation for both groups of sugar-fed and protein + sugar-fed males, respectively. However, the number of female sightings was significantly greater at leks of protein + sugar-fed males than at leks of sugar-fed males. An average of 6.8 female sightings were made per trial (i.e., over 10 observations) for leks of protein + sugar-fed males compared to only 3.6 sightings at leks of sugar-fed males.

For the within-plant comparisons, data were collected using a field-caged guava tree. One cup of sugar-fed males and one cup of protein + sugar-fed males were suspended in the canopy of the tree; each cup held five males of a given type. Thirty females were released from the base of the tree, and the numbers of calling males and perching females were noted for both cups at 2.5 minute intervals over a 90-minute period. We found no difference in either male calling or female sightings. For both male groups, an average of 2.5 males were calling per observation, and an average of

approximately one female sighting was recorded per cup per observation. These results suggest that adult diet has little effect on male signal attractiveness over short distances (within a canopy). However, in the between-plant tests, females were sighted more frequently at groups of protein + sugar-fed males, suggesting males fed a more complete diet have a greater "long-range" attrcativeness than do males fed only sugar.

d. Objective 4: to establish whether copulatory success of lekking sterile male flies may be enhanced by specific pre-release diets and judicious dispersal strategies

d.1. Trimedlure and the mating competitiveness of irradiated male Mediterranean fruit flies (Shelly 1999).

Medfly males are attracted to trimedlure, a synthetic lure widely used in monitoring and eradication programmes. Previous work with wild males revealed that males exposed to trimedlure had a mating advantage over unexposed males under laboratory conditions. In this study we examined whether exposure to trimedlure increased the mating competitiveness of irradated males from a long established medfly colony (Maui 53), relative to non-irradiated males recently colonized (Maui 95). The latter achieved a disproportionate number of matings compared to unexposed males. However, in tests conducted 1 day after exposure, Maui 53 males mated significantly more often than Maui 95 males. In tests conducted 3 and 7 days after exposure irradiated males enjoyed the same level of mating success as unirradiated males. Exposure to trimedlure resulted in a slight, but insignificant increase in calling activity of Maui 53 males. These results suggest that manipulating the olfactory environment of sterile males prior to release could improve their sexual performance in the field.

d.2.. Post-teneral protein feeding improves sexual competitiveness but reduces longevity of mass reared sterile male Mediterranean fruit flies (Kaspi & Yuval 2000)

We examined how post-teneral nutrition during the first 4-8 days after emergence affects performance and copulatory success in leks of mass-reared sterile (TSL strain) males. We found that protein and sugar fed males were significantly more likely to emit pheromone (call) in leks, and more likely to copulate than males fed only sugar. Sterile males, that had access to water and apples following four days feeding on protein and/orsugar were significantly more likely to copulate than their starved competitors who had access to water alone. However, after 24 hours of starvation, four day protein-fed males suffered a higher mortality than sugar-fed males. More work is necessary to determine the optimal protein formulation that will maintain a balance between hastened mortality and increased sexual competitiveness of sterile males.

Discussion

a. Scientific implications

a.1. Medfly leks and lekking behavior (reviewed in Field et al. 2000)

Despite the passage of two decades since the medfly was identified as a lek-mating organism, our understanding of the evolutionary forces driving lek formation in this species remains rudimentary. This is not due to a lack of research effort into medfly mating behavior, but rather because such research has rarely been framed with evolutionary issues specifically in mind or been used to explicitly test evolutionary hypotheses. Understandably, the emphasis has been on experiments designed to bring immediate improvements in the quality of mass-reared males, to lead toward development of superior attractants for baiting and monitoring programmes, or to understand the proximate mechanisms determining successful courtships. However, basic and applied research questions are never mutually exclusive, and we believe that attempting to place medfly mating behavior in an explicit evolutionary setting can yield practical benefits, just as practically-oriented research has already begun to benefit our evolutionary understanding, by providing critical empirical data for testing theoretical models. Most importantly, an evolutionary framework can facilitate ongoing critical evaluation of empirical studies, aiding the resolution of experimental ambiguities and contradictions, and speeding the conversion of an otherwise haphazard accumulation of results into an orderly, coherent body of knowledge.

Many questions and uncertainties remain. Below we identify lines of research that appear to hold promise for teasing apart the influences of various ecological factors, and suggest some experiments critical to resolving outstanding issues.

Firstly, most of the tentative conclusions concerning lek evolution rely on evidence from only one or a few studies. Inevitable variations among studies in the origin, rearing and handling of insects, experimental methodology and analysis make it likely that even the most carefully designed and executed of studies can produce ambiguous or inconclusive results. In medflies, the potential for discrepancies between studies is perhaps compounded by the fact that this insect has relatively recently colonized a variety of new habitats worldwide, and different populations have possibly undergone (or are undergoing) adaptation to local conditions. It therefore may be necessary to accumulate numerous tests of the same hypothesis under differing ecological conditions before robust conclusions emerge. Ideally, consensus on the evolutionary influence of an ecological factor should be quantitatively assessed after taking multiple similar studies into account. Far from being a

redundant exercise, repeating experiments performed by other researchers on different medfly populations may highlight critical ecological factors that influence mating behavior and thus prove essential to the task of understanding its evolution.

At the large-scale spatial level, it would be useful to repeat studies which apply a multivariate analysis to confirm which factors determine the favored sites for male display within the habitat. Ideally, such studies would be longitudinal in nature and would track the location of calling males in relation to seasonal patterns of host availability within seasons, and fluctuations in these patterns among seasons. Combined data from different medfly populations, climates and habitats would provide a rich database with which to identify universal factors determining lek locations. Should the pattern of clumped lek distributions be borne out by such studies, the next task would require tracking of female distributions (feeding sites, oviposition sites and movements among them) and the demonstration of a correlation between female distribution and the male calling sites.

Concerning specific hypotheses for lek evolution, the effect of predation is one area that has recieved intense empirical attention recently (Kaspi & Yuval, submitted and references therein) and should be pursued further. To clarify whether the formation of leks in trees with large volumes and dense canopies is in part a response to predation, it would be desirable to measure predation rates in trees of different size and canopy structure. The information already obtained on attack rates at different lek sizes could be supplemented by data on the rate of *successful* attacks at different lek sizes, which would indicate whether individuals displaying in larger groups benefit from increased vigilance. As this would entail measurement of predation rates on naturally displaying males, the data would be difficult to obtain, but would be well worthwhile as they would clinch the argument for the role of predation in driving male aggregation.

Further investigations should focus on testing whether the proximate mechanism by which males aggregate is indeed by cueing on the pheromone emissions of other males, and if so, which are the active components in the blend. A positive result would add credibility to the hypothesis that low-quality males are attracted to leks occupied by superior males that can be distinguished by the quality of their pheromone. This hypothesis could then be investigated in an experiment similar to that performed by Shelly (in press), who tested attraction of males and females to calling males of low and high mating ability, with the difference that the calling males should be classified with respect to their ability to attract conspecifics on the basis of their pheromone alone. Classifying

them by their mating success leaves open the possibility that the high-mating males were successful not due to quality of their pheromone, but due to the efficacy of their courtship, which is of no evolutionary consequence for lek formation. If both females and males concurred in their choice of males, the pheromone blends of attractive and unattractive males could then be compared and the physiological basis for attraction identified.

One of the most interesting research directions to pursue is the putative interaction between predation and female preferences for large leks in determining optimal lek size. Whilst large leks appear to increase male survival by decreasing per-capita attack rate, this may not be true for females, at least judging by the behavior observed by J. and M. Hendrichs (unpublished). This may be explained by the fact that female vigilance toward predators is at its lowest when receiving courtship, so females may be particularly sensitive to the risk of predation when visiting a lek. This not only brings female interests into conflict with that of males with respect to predation, but also sets up a counterbalance to any preference females might have for mating in larger leks due to the opportunity to compare males, as larger leks attract more predators. Thus it could be an important selective force acting to set an upper boundary to lek sizes. Further experiments measuring female arrival rates whilst manipulating lek sizes and predator attack rates would be extremely valuable.

a.2. Protein nutrition and reproductive success

We have been able to establish that post teneral protein diet significantly increases male sexual success. This is true for wild, colonized and sterile males, in Israel and Hawaii. Thus there is a strong link between male foraging ability and sexual success, as well as an opportunity to improve the performance of sterile males by feeding them on a protein rich diet before release. This treatment however, may backfire, if sterile male are unable to forage in the field. Further experiments are needed to clarify this issue.

a.3. Exposure to Trimedlure and reproductive success

In addition to the diet ingested by males in the days following eclosion, their olfactory environment also impacts their subsequent performance in the field. This is best exemplified by the effect of exposure to Trimedlure, which significantly enhanced the copulatory success of both wild and sterile males. Shelly's work with other tephritids

suggests that the olfactory environment of emerging sterile males may be manipulated in a manner that will increase the succes of the sterile insect technique.

b. Potential impact on agriculture, industry, environment, policy, legislation

Although the research directions outlined above are primarily directed towards answering a theoretical question in behavioral ecology, there also exist potential avenues whereby such research could make a positive contribution to improving the efficacy of medfly control programmes. Tests of the hypotheses of lek formation will provide us with detailed knowledge about preferred lek locations in various habitats and climates, enabling more judicious selection of sites for monitoring traps and thus improving the ability to detect and respond to infestations. Such studies could provide the key to understanding variations in male attractiveness, and be a step forward in improving the quality and mating competitiveness of mass-produced males *vis à vis* their wild counterparts. If specific components of the male pheromone could be identified as responsible forincrease attractiveness, they could also be used to manufacture more effective chemical baits and lures. It is our hope that the future will see more interaction between applied empirical research on medfly mating behavior and theoretical modelling of lek evolution, to mutual profit.

Finally, our finding that manipulating the males during the post teneral period, when they are in the pre-release facility, whether via their diet or olfactory environment, has immediate practical implications.

List of all publications resulting from the project:

1. Field, S.A., R. Kaspi & B. Yuval. 2000. Why do calling medflies cluster? Assessing the empirical evidence for models of medfly lek evolution. Florida Entomologist (in press).
2. Kaspi, R. & B. Yuval. 1999a. Lek site selection by male Mediterranean fruit flies. Journal of Insect Behavior 12:267-276.
3. Kaspi, R. & B. Yuval. 1999b. Mediterranean fruit fly leks: factors affecting male location. Functional Ecology 13: 539-545.
4. Kaspi R., P.W. Taylor & B. Yuval. 2000. Diet and size influence sexual advertisement and copulatory success of males in Mediterranean fruit fly leks. Ecological Entomology (in press).
5. Kaspi, R. & B. Yuval. 2000. Post-teneral protein feeding improves sexual competitiveness but reduces longevity of mass reared sterile male Mediterranean fruit flies. Annals of the Entomological Society of America (in press).
6. Shelly, T.E. 1999. Trimedlure and the mating competitiveness of irradiated male Mediterranean fruit flies (Diptera: Tephritidae). Environmental Entomology. 28: 780-786.
7. Shelly, T.E. Male signalling and lek attractiveness in the Mediterranean fruit fly. Animal Behaviour. (in Press).
8. Shelly, T.E., S. Kennelly & D.O. McInnis. Effect of adult diet on signaling activity, mate attraction, and mating success in male Mediterranean fruit flies (Diptera: Tephritidae). Florida Entomologist (in Press).
9. Yuval, B., & J. Hendrichs. 1999. Behavior of flies in the genus *Ceratitis*. In: Fruit Flies (Tephritidae): Phylogeny and Evolution of Behavior. (Aluja, M. & A. Norrbom, eds), CRC Press, Boca Raton. pp. 429-456.
10. Yuval, B., R. Kaspi, S. Shloush & M.S. Warburg. 1998. Nutritional reserves regulate male participation in Mediterranean fruit fly leks. Ecological Entomology 23:211-215.
11. Yuval, B., R. Kaspi, S.A. Field, S. Blay, and P. Taylor. 2000. Effects of post-teneral nutrition on reproductive success of male Mediterranean fruit flies. Florida Entomologist (in Press).

Submitted:

12. R. Kaspi & B. Yuval. Effect of predators on lek site selection by male Mediterranean fruit flies. Oecologia. (submitted April 2000).
13. R. Kaspi, I. Feitelson, T. Drezner & B. Yuval. A novel method for rearing the progeny of wild Mediterranean fruit flies using artificial fruit. Phytoparasitica (submitted April 2000).
14. Shelly, T.E.. Lek size and female settlement patterns in the Mediterranean fruit fly. Animal Behaviour (submitted May 2000).