ERADICATION OF THE MELON FLY, *DACUS CUCURBITAE*, FROM KUME IS., OKINAWA WITH THE STERILE INSECT RELEASE METHOD

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The first successful demonstration of the sterile insect release method (SIRM) was against the screw-worm, *Cochliomyia hominivorax*, which was eradicated from Curacao Is. in 1954 (BAUMHOVER *et al.*, 1955). This fly was also eradicated from the Florida Peninsula with the same method in 1959 (LACHANCE *et al.*, 1967). The melon fly, *Dacus cucurbitae*, on Rota Is. and the oriental fruit fly, *D. dorsalis*, on Guam Is. were eradicated with SIRM till 1963 (STEINER, 1965). Since then many attempts have been made in a number of countries to control or eradicate insect pests with this method. However, it is our opinion that there has been no successful eradication of an insect pest with the SIRM among these trials, except for temporal annihilations or cases where large numbers of sterile insects were released to eradicate a pest just after its accidental introduction in small numbers. Thus there are still many problems which need to be solved to develop this unique method for practical eradications of noxious insects. Of particular importance in the SIRM are reliable procedures for obtaining precise information on the population parameters of wild and released insects.

In 1972, the Japanese Ministry of Agriculture and the Okinawa Prefectural Government started a large scale experimental project to eradicate the melon fly, *Dacus cucurbitae* COQUILLET, with SIRM from the 58.5 km² island of Kume, Okinawa. This was the first attempt to implement SIRM in Japan. For this purpose, a mass-rearing facility and an irradiation facility were established on Ishigaki Is., which is about 300 km southwest of Naha, the island of Okinawa, and in Naha, respectively (Fig.1).

Before sterile-fly releases were started, the population density of the wild melon flies on Kume Is. was estimated with the mark-release-recapture method (ITÔ *et al.*, 1974). The number of mature wild males in the peak season (November) of 1972 was estimated to be ca. 2.5 million. Meanwhile the production capacity of the mass-rearing facility just before the beginning of sterile insect releases was only 1-1.5 million.

In order to reduce the size of the wild fly population prior to the mass releases of sterile insects, control of the melon fly population was attempted by aerial distribution of cotton strings saturated with a solution of cue-lure (97%) and naled (3%), and with ground sprays of a mixture of protein hydrolyzate and technical malathion (IWAHASHI *et al.*, 1975). Possibly due to these treatments, which were

Fig. 1. Map of the Ryukyus. The three islands of Ishigaki, Okinawa and Kume are shown; other islands are omitted.

applied from November, 1972 to January, 1975, the density of the melon fly population in January, 1975, just before the beginning of mass-releases was estimated to be about 5% of the peak density. The fly density on Kume Is. at the bottom of yearly fluctuations was lower in early spring of 1974 (when the intensity of control was not so strong) than in 1975 (see Fig. 4) but as the population trends in Naha, the control area, and Kume Is. were remarkably different at the end of 1974, we can consider that the decrease in fly density in autumn of 1974 was mainly due to the effect of control practices.

METHODS

Fly pupae to be sterilized and released were reared in the mass-rearing facility on Ishigaki Is. (NAKAMORI *et al.*, 1975, KAKIHANA *et al.*, 1975, NAKAMORI *et al.*, 1976). They were transported weekly by air to Naha using a specially designed container and irradiated at a dose of 6-7 kR with a cobalt-60 source, 2 or 3 days before adult eclosion. The pupae were then air-shipped to Kume Is. and marked with a fluorescent dye (Whitex SKC[®], Satan Yellow[®], or Blaze Orange[®]) at a rate of 4 to 8 g per 100,000 pupae. Pupae in specially designed release buckets were placed in 170 release points distributed over the island. The mean distance between points was about 600 m. Pupae were usually left in release buckets until the next release in order to determine the rate of emergence.

To evaluate the ratio of sterile to wild males in the field, 50 monitor traps (plastic traps baited with cue-lure and naled) were distributed over the island. These traps were examined twice a month, and the melon flies caught were brought to the laboratory. Heads of these flies were carefully crushed on filter paper with a glass rod dipped in acetone, and the filter paper was examined under an ultraviolet lamp (2537 Å or 3650 Å/100 V) for fluorescent marks.

Initially we had planned to release 1 million pupae per week, which was near the maximum number of pupae we could produce in our mass-rearing facility at that time. Although the ratio of marked (sterile) to unmarked (normal) males in the monitor traps, M/U , had decreased from March to July, 1975, there was no sign of decrease in the percent egg-hatch in Kume Is. as compared with the control area at Naha, thus we made some modifications in our mass-rearing system to increase the number of pupae up to 2 million per week. After March 1976, we attempted to further increase the number of pupae released up to 4 million per week to accelerate the eradication process. The actual number of pupae released exceeded 3.5 million per week in May, 1976.

Other modifications made in our SIRM project after September, 1975 were as follows:

1. To decrease the mortality of teneral adults due to predation, the design of release buckets was modified to discourage predators.
2. The number of release points was increased from 170 to 200, in order to concentrate sterile flies in localities where host plants were abundant.
3. In June, 1976 the so-called Taiwan-type monitor trap was replaced with the Steiner-type (STEINER, 1957), the bottom of which was covered with a saran-net to prevent the occurrence of a natural fluorescence in trapped flies due to decomposition of the insects (see below).

To evaluate the effect of sterile-fly releases on the hatchability of eggs, female flies were netted in fields on Kume Is. These females were reared individually in small plastic containers and eggs were collected with an artificial oviposition device (see IWAHASHI *et al.*, 1976). Eggs deposited in the device were placed on moistened filter paper in petri dishes and kept for 3 days at a constant temperature of 27°C after which the rate of egg-hatch was determined. Bi-monthly examinations of 5 monitor traps and collections of females were also made in Naha, which was the control area where no releases were made.

Infestation levels were checked monthly by collection of fruits of a wild cucurbit plant, *Bryonopsis laciniosa*, from many localities on Kume Is. More than 2,000 fruits were examined every month during the experimental period, except April and July, 1975. Fruits with apparent infestations or punctures were dissected within 2 or 3 days after they were collected and eggs or larvae found were recorded. Fruits containing unhatched eggs were recorded as uninfested. The remaining fruits were kept on moistened sand in plastic containers for 2 or 3 weeks at ambient temperatures to allow pupation and emergence of insects which had previously been overlooked.

RESULTS

Fig. 2 presents the results of sterile-fly releases on Kume Is. from February, 1975 to December, 1976. Open circles in Fig. 2-a indicate the number of sterilized

Fig. 2. Results of sterile insect releases on Kume Is. from February, 1975 to February 1977. Fig. 2-a shows the average number of sterilized pupae released per week (open circles) and estimated number of flies emerged (solid circles). Fig. 2-b shows the monthly ratio of marked (M) to unmarked males (U) caught in 50 monitor traps on Kume Is. Fig. 2-c shows the hatchability of eggs laid by female melon flies collected on Kume Is. (solid circles) and Naha, the control area (open circles).

pupae released per week, and solid circles the number of adults emerged, which was estimated from the number of pupae released and the rate of successful emergence of sampled pupae, Fig. 2-b shows the change in the ratio of marked to unmarked flies, M/U , caught in 50 monitor traps. No marked fly was captured in February and the M/U ratio was 1.83 in March, 1975. But the ratio decreased thereafter in spite of the increased numbers of sterile flies released. This was probably due to a seasonal rise in the population density of wild flies which exceeded the increase in numbers of released flies. The melon fly population usually decreases on Kume Is. during June and July (IWAHASHI *et al.*, 1975), but further decreases in the M/U ratio were observed even in these months. One reason for this may have been due to an underestimation of the ratio, because since June, determination of marked flies with an ultraviolet lamp became difficult due to the frequent occurrence of a green natural fluorescence in captured flies. This natural fluorescence was attributed to the decomposition of dead flies in Taiwan-type traps due to warm and humid conditions. Since all flies with only very faint fluorescence were recorded as 'unmarked' the

M/U ratio might have been underestimated during the warm seasons. The high mortality rate of released pupae was also considered to be another cause of the decrease in the M/U ratio (Fig. 2-a). However, we feel that the primary reason for the low M/U ratio was the insufficient number of flies released during this period.

The M/U ratio started to increase from autumn of 1975, when the number of pupae released approached 2 million per week, exceeded 2 in February, 1976 and kept increasing in successive months until it reached 10 in June, 1976. The ratio rose to 45 in September, 1976 and exceeded 90 in October of that year. After that the values stabilized around 100.

Fig. 2-c and columns 4 and 8 of Table 1 show the results of the experiment on hatchability of eggs laid by females collected on Kume Is. (solid circles) and at Naha, the control area (open circles). Fifteen females caught on Kume Is. in July, 1975 were examined for a total of 66 days, and 10 of these laid 417 eggs, of which 91% hatched. This value is similar to that obtained in the control area and higher than the rate obtained in the treated area in June. In August, 13 of 56 females laid 272 eggs and the percent egg-hatch decreased to 64. From September onwards, the percent egg-hatch on Kume Is. decreased consistently, that is, 58 in September, 45 in November, 29 in March, 1976, and 20 in May. In July, 1976, 10 of 300 females examined for a total of 42 days laid 670 eggs, of which 1.5% (10 eggs) hatched. In September, only 3 of 524 females, examined for a total of 51 days, laid 200 eggs, and 9 of these hatched (4.5%). Although percent egg-hatch was higher in September than in July, this may have been due to sampling errors related to the very small number of females which laid eggs. In December we were unable to obtain a value for percent egg-hatch on Kume Is., since 383 females examined for a total of 45 days laid no eggs.

As mentioned above, detection of marked flies under an ultraviolet lamp became difficult during the warm seasons in 1975 due to the occurrence of natural fluorescence. To overcome this problem, ITÔ and IWAHASHI (1976) presented a method to estimate the ratio of sterile to normal males based on percent egg-hatch. Their method uses FRIED's equation (FRIED, 1971), which was designed to estimate the 'sexual competitiveness' of sterile males, C , at a given ratio of sterile to normal males, that is,

$$C = \frac{H_n - H_c}{H_c - H_s} \cdot \frac{N}{S} \quad (1)$$

where

H_n = % egg-hatch in matings of $N\sigma^{\uparrow}\sigma^{\uparrow} \times N\varphi\varphi$

H_s = % egg-hatch in matings of $S\sigma^{\uparrow}\sigma^{\uparrow} \times N\varphi\varphi$

H_c = % egg-hatch at a given ratio of $S\sigma^{\uparrow}\sigma^{\uparrow}$ to $N\sigma^{\uparrow}\sigma^{\uparrow}$ in the presence of $N\varphi\varphi$.

N = Number of normal males.

S = Number of sterile males.

H_s is zero in our experiment when an irradiation dose of 6-7 kR was applied to pupae 2 or 3 days before adult eclosion. If we know H_n , H_c and C , we can estimate the ratio of sterile to normal males (S/N) from Eq. 1, that is,

$$\frac{S}{N} = \frac{H_n - H_c}{H_c} \cdot \frac{1}{C} \quad (2)$$

For the value of C , 0.7 was tentatively used, based on the results of laboratory experiments on sexual competitiveness of irradiated melon flies (TERUYA and ZUKEYAMA, 1975). Substituting values of percent egg-hatch on Kume Is. for H_c and those in the control area for H_n , estimated values of S/N were calculated (10th column of Table 1). The values thus obtained were higher than M/U during the warm season in 1975.

TERUYA and ZUKEYAMA (in preparation) showed that females irradiated at 6-7 kR are infecund. Thus, we can also estimate the ratio of sterile to normal *females* based on the ratio of females which oviposited (including both fertile and sterile eggs) to the total number of females collected on Kume Is. and Naha.

That is,

$$R = C_f / C_o \quad (3)$$

and

$$\frac{S_f}{N_f} = \frac{T_f - RT_o}{RT_o} \quad (4)$$

where

S_f = Number of sterile females in treated area.

N_f = Number of normal females in treated area.

C_f = Number of females collected in control area.

C_o = Number of females collected in control area and which oviposited eggs.

T_f = Number of females collected in treated area.

T_o = Number of females collected in treated area and which oviposited eggs.

Substituting values shown in Table 1 for T_f , T_o , C_f and C_o in Eq. 4, expected values of S_f/N_f can be obtained (11th column of Table 1). If the actual ratio of sterile to normal flies is not different between sexes, S_f/N_f can be used also as an index indicating the ratio of sterile to normal males, and can be compared directly with estimated values of S/N and M/U . As shown in Table 1, the values of S_f/N_f were nearly equal to those of S/N , but higher than those of M/U during the warm season in 1975. These three values agreed well in March and May, 1976, when the natural fluorescence disappeared. The large difference among these values in July and September was probably due to sampling error in T_o and H_c and also to under-estimation of the M/U ratio (see below).

Fig. 3 shows changes in percent infestation of fruits of *B. laciniosa* by the melon fly before, and during, the mass-releases on Kume Is. If it can be assumed that the percent infestation of fruits reflects the density of the melon fly population even when the density of the fly population is very low, percent infestation can be presented

Fig. 3. Percent infestation of wild cucurbit fruits, *Bryonopsis laciniosa*, on Kume Is. before and during the mass-releases of sterile insects.

with a logarithmic scale such as shown in Fig. 3. The mean percentage of infested fruits in the pre-release period (from June, 1972 to January, 1975) ranged from 2.56 to 23.62. During the first year or so of the SIRM program, the percent infestation was low, fluctuating between about 3.9 to 0.4%, but there was no sign of remarkable decrease. After May, 1976, however, it decreased rapidly and reached zero in August. In September, when 5081 fruits were examined, the percent infestation rose to 0.06. From October, 1976 to April, 1977, a total of 73,788 fruits of *B. laciniosa* was gathered on Kume Is. and examined in Naha, but there was no sign of melon fly infestation during these seven months.

Fig. 4 shows changes in the number of wild melon fly males caught in 50 monitor traps on Kume Is. (solid circles) and those in five traps in the control area at Naha (open circles; cited from YAMAUCHI *et al.*, 1976). Traps distributed on Kume Is. caught both marked and unmarked flies during the SIRM project, the latter were counted as wild males. It is noted that after the beginning of the SIRM the number of unmarked flies on Kume Is. had increased.

In April, 1975 the number of unmarked flies on Kume Is. was 405 per 1,000 trap-days. This density was ten times higher than that recorded in February, when mass-releases were initiated, and nearly the same as that obtained before suppression. Although the numbers of melon flies caught in traps at Naha were always higher than those on Kume Is., populations at both locations fluctuated in the same manner from August, 1975 to July, 1976. After July, 1976, the melon fly population on Kume Is. began to decrease, in spite of the fact that the population in the control area was

increasing. This suggests that the suppressive effect of mass-releases had reached a level which was sufficient to decrease the population of the next generation. As mentioned earlier, there was no sign of infestation of fruits by the melon fly on Kume Is. since October, 1976. However, unmarked flies were still found in monitor traps, probably due to the fact that marking of flies with fluorescent dye powder was not perfect. In February, 1977 the number of unmarked flies decreased to 4.2 per 1,000 trap-days. This value was about 0.4% of the peak density, however, for the above-mentioned reason, it is considered to be an overestimation of the wild fly density.

DISCUSSION

As mentioned earlier, Itô *et al.* (1974) estimated that the number of male flies inhabiting Kume Is. in the peak season of 1972 was about 2.5 million. It was also estimated that the fly population decreased to about 5% of the peak season density, following the application of control treatment (IWAHASHI *et al.*, 1975). We initially released 1 to 1.5 million pupae per week. The releases made from February to July, 1975 were not sufficient to control the wild melon fly population on Kume Is. After September, 1975, pupal releases were increased to 1.5-2 million per week. As a result, the ratio of M/U began to rise and percent egg-hatch began to decrease. In spite of this decrease in the percent egg-hatch, percent infestation of cucurbit fruits did not decrease markedly during the same period. These results suggest that a decrease in egg-hatch to about 30% was not enough to reduce the density of the next generation of *D. cucurbitae*. Actually, the numbers of unmarked flies on Kume Is.

Fig. 4. Monthly changes in abundance of male melon flies caught in 50 monitor traps on Kume Is. (solid circles) and in 5 ones in Naha, control area (open circles).

and those in the control area fluctuated in the same manner in the latter half of 1975 (Fig. 4).

Based on studies with a model of the SIRM process for the melon fly, Irô (in preparation) suggested that releases of 4 million sterilized pupae per week could remarkably accelerate the eradication process on Kume Is. From February, 1976 we increased the number of pupae released per week, and, in May and subsequent months it reached 3.5-4 million per week. The ratio of M/U increased remarkably after February. This ratio increase was less due to a decrease in the wild melon fly population as to the increase in the number of flies released (the first half of 1976 in Fig. 4). When the number of pupae released reached the maximum level of 3.5 to 4 million per week since May, a further increase in the M/U ratio was recorded.

From Fig. 4 we can see that the population in the control area increased after July, 1976, while that on Kume Is. was on the decrease. At the same time, a remarkable decrease in the percent egg-hatch took place. These results suggest that our mass-releases started to suppress and reduce the wild fly population on Kume Is. since July, 1976. The sharp reduction of percent infestation of host fruits took place from May, 1976 onward (Fig. 3). This rapid decrease started from May, 1976 when the M/U ratio reached about 7 and the rate of egg-hatch decreased to 20%. Twenty to thirty percent in the rate of egg-hatch during peak population season may be a critical point for the success of an eradication project with SIRM.

During the SIRM program on Kume Is., all flies released were marked with a fluorescent dye powder. The monitor traps on Kume Is. thus caught both marked and unmarked flies. As mentioned previously, we recorded as marked only those flies which showed unmistakable fluorescence. It was considered that the M/U ratio was always lower than the actual ratio of sterile to normal flies. That is to say, the number of unmarked flies caught in monitor traps during the SIRM was always larger than the true number of wild males. The M/U value reached a constant level around 100 from October, 1976 onward. This could mean that marking with fluorescent dye was not complete or some flies lost their dye before they were trapped. If we assume that 99% of the released flies were properly marked on the ptilinum, the M/U would never exceed 100 even when the population had been eradicated.

Since we do not have data on the percent of marked released flies in those months when the number of unmarked flies caught in monitor traps on Kume Is. began to decrease, we do not know exactly when the wild population began to decrease rapidly, however it was probably 1 or 2 months earlier than July, 1976. We consider that eradication of the melon fly population on Kume Is. with the SIRM was achieved by April, 1977.

SUMMARY

The sterile insect release method was applied to eradicate the melon fly,

Dacus cucurbitae, from the 58.5 km² island of Kume, in the Okinawa Islands group. Weekly releases of 1 to 1.5 million flies irradiated as pupae with 6-7 kR from a cobalt-60 source did not decrease the wild melon fly population. Releases of 1.5-2 million pupae per week made from September, 1975 to January, 1976 decreased the percent egg-hatch of females caught on Kume Is., but did not decrease the percent infestation significantly. The number of pupae released was increased from February, 1976 to accelerate the eradication process. When the number of pupae released exceeded 3.5 million per week, a rapid increase in the ratio of marked (sterile) to unmarked (wild) flies, a remarkable decrease in percent egg-hatch, and a decrease in percent infestation of fruits were observed. There has been no sign of melon fly infestation in wild cucurbit fruits from October, 1976 to the present time (April, 1977), despite the fact that more than 70,000 fruits were carefully examined. The eradication of the melon fly from Kume Is. was thus achieved by April, 1977, after the release of 264 million sterile fly pupae.

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不妊虫放飼法による久米島のウリミバエ根絶

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沖縄本島の西約 80 km に位置する面積約 58.5 km² の久米島でウリミバエを根絶するために、大規模な不妊虫放飼実験を行なった。誘引剤による抑圧防除ののち、1975年2月から不妊虫の放飼を開始した。放飼効果の判定は、久米島に配置した50個のトラップに誘殺される放飼虫(放飼虫は全てマークを付してある:M)と野生虫(U)の比、同島で採集したメスの産む卵のふ化率、野生寄主(オキナワスズメウリ)の寄生果率によった。1975年2月から8月までは週100~150万匹の蛹を放飼したが、上記の示標によると放飼の効果はほとんど現われなかった。すなわち、無防除地区のメスの産んだ卵のふ化率が75~90%であったのに対し、久米島のものでほとんど同じレベルであった。そこで放飼数を増加し、1975年9月から1976年1月まで150~200万匹を放飼した結果、M/U比は上昇し、卵のふ化率は低下した。したがって不妊虫放飼の効果がある程度現われてきたことが伺われるが、早急に本種を根絶するにはなお不十分であると思われた。そこで、1976年2月以降再び放飼数を増加した。放飼数が週350万匹のレベルに達した1976年5月にM/U比は7となり、その後も増加し続け9月には90となり、それ以後は約100を維持した(M/U比がこれ以上増加しなかったのはマークが完全でなかったことによる)。卵のふ化率は1976年5月には20%に低下した。7月にはさらに1.4%に低下した(それ以後は採集したメスのほとんど全てが放飼した不妊虫となり、信頼性のあるデータは得られなかった)。

以上のことから、久米島のウリミバエは不妊虫放飼法によって根絶したものと考えられる。なお、1975年2月の放飼開始時点から、1977年3月末までの総放飼数(蛹)は約2億8千万匹であった。