Study on the Mating Ability and Competitiveness of the Radiation Irradiated Males of Rice Stem Borer

(*Chilo suppressalis* Walker).

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방사선 조사(照射) 이화명나방의 교미능력 및 경쟁력에 관한 연구

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적 요

방사선을 투사한 이화명나방의 교미능력 및 경쟁력 조사를 5~7령 번데기에 gamma 선을 투사하여 조사하였다. 얻어진 결과는 다음과 같다.

1. 방사선 처리웅충(雄蟲)과, 정상적인 수컷에 매일 새로운 미교미자충(未交尾雌蟲)을 넣어 주었을 때 그들간에 교미 최고회수에는 유의차가 없었다.
2. 교미초일은 정상웅충의 경우 평소 1.5일이며 25.30 Krad 처리웅충은 2.4일이었다. 교미 일 간격은 정상웅충이 1.3 일인데 대하여 처리웅충은 3.0 일이었으며 방사선 투사는 교미력에 영향이 있었다.
3. 정상웅충(雄蟲) 1 두(頭)에 정상자충(雌蟲) 5 두와 10 두씩을 넣어서 정상웅충의 평균교미회수를 조사한 결과 1.2 회와 2.5 회이었다.
4. 교미경쟁력 실험에서 미교미자충(未交尾雌蟲) 5 두에 대하여 방사선 투사 웅충과 정상웅충의 비율을 1 : 0, 1 : 1, 2 : 1, 1 : 2, 5 : 1, 1 : 5, 0 : 1 로 하였을 때 난부화 감소율은 5 : 1(30Krad)에서 최대의 감소를 보였으며 이때의 난부화율(卵孵化率)은 26.6%이었다.
5. 방사선 투사는 교미능력에 다소의 영향을 미치나 처리웅충에 의한 산란수(産卵數) 감소의 영향을 고려한다면 이화명충에서 웅성불임 기술의 이용은 가능할 것으로 생각된다.

Abstract

Five to seven day old pupae were irradiated with gamma rays, the mating ability, and competability of the irradiated males were examined. The results obtained are summarized as follows:

1. There is no difference in percentage of males that mated, nor in the maximum number of matings

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of the males, when individual matings were made, and fresh females were provided every day for his life time.

2. The average days for the first mating of the emerged were 1.5 days for the control, and 2.4 days for the males irradiated with 25 and 30 Krad. The average days between first and second mating were 3.0 days for the irradiated males, and 1.3 days for the control.
3. The average matings of a male were 1.2 and 2.5 times when 5 and 10 females were provided to a normal male respectively. No significant different in average numbers of mated female per male were observed between the normal and irradiated males.
4. To test the mating competitiveness of the irradiated and normal males (1 : 0, 1 : 1, 2 : 1, 1 : 2, 5 : 1, 1 : 5, and 0 : 1) were confined together with five virgin females. The greatest reduction in hatching percentage was found with 5 : 1 : 1 (30 Krad) where 26.6% was hatched.
5. The results showed that the irradiated males were not fully competitive with normal males, but if the reduced number of eggs produced by the female mated with irradiated male is considered, it might be used sterile male techniques in rice stem borer control.

Introduction

One of the important requirements for the successful application of the sterile male technique is to sterilize the insects without serious damage to their mating ability or other biological behaviour. Several species of moths have been studied to test the suitability of the sterile males for release program. There has been a considerable amount of accumulated data on the mating competitiveness of the sterile males indicating that the insects can be sterilized without any apreciable alteration of the mating behaviour if treated with adequate dosages at a suitable of the life stage.

Mating ability of a moth has been routinely tested by holding normal females with various ratios of sterile to normal males and testing the fertility of the eggs produced. The mating of the tested insects is usually assessed by the presence of spermatophores in the bursa copulatrix of the female (Ouye et al, 1964).

The work reported here is a continuation of the feasibility tests on the use of the sterile male technique in the control of rice stem borer, *Chilo suppressalis* (Walker). The purpose of this work is to investigate the effects of radiation on the mating ability and competitiveness of the irradiated males of the insect.

Materials and Methods: General

A large number of overwintered larvae were collected in the field, and stored in a refrigerator (5°C) with pieces of corrugated carton board into which the larvae had crawled. They were transferred into incubators which maintained at 25±2°C. temperature and 90% relative humidity. The pupae were removed every day and sexed.

Five to seven day old pupae were irradiated, placing them in a small glass vial. Less than twenty pupae were irradiated at once. The Co-60 sources were pole types, and the dose rates were 4.56×10^4 rad/hr. and 3.71×10^4 rad/hr. at a distance of 10cm from the two sources. The temperature was 18±2°C. at the time of irradiation.

The calibration of the dose rates was made by Fricks chemical dosimetric methos, the exposure time was less than 30 minutes.

The irradiated pupae were held in large glass cylinders which were kept in an incubator. Each cylinder was provided with a water soaked roll of dental cotton that was remoistened daily. The matings were made on the morning following the adult emergence. Further details of the experimental methods will be explained for each experiment.

Results and Discussion

1. The mating ability of the irradiated males.

a. The number of matings during life of the

male.

Each of emerged male moths from irradiated pupae was confined individually with a female in a small glass bottle (8.5cm in diameter and 8cm in height), which was provided with a water-soaked roll of dental cotton, remoistened daily.

Ten replicates were used for each treatment. A parchment paper was provided for oviposition on the inside wall of the bottle. A fresh virgin female was provided every day until the irradiated male died; consequently the females were of constant age even though the male became progressively older.

The removed females were preserved separately in 75% alcohol until the examination for spermatophore in the bursa copulatrix. The presence of spermatophores was regarded as evidence of mating. The results obtained are shown in table 1.

Table 1. Mating performance of males when a fresh female was provided daily throughout the male's life time.

Doses (Kr)	Mated male(%)	Max. no. of mating for a male	Mated females (%)	Total no. of females provided	Longevity of male* (days)
0	100.0	2	37.5	32	3.3(1-5)
25	100.0	2	30.0	40	4.0(1-7)
30	100.0	2	30.0	40	4.0(1-9)

*Average longevity of the treated males (range in parenthesis).

There is no difference in percentage of males that mated, nor in the maximum number of matings of the males. The percentages of mated females were 37.5% for the control, and 30% for the treated males.

The day of first mating and the interval between first and second mating are shown in table 2.

Table 2. Average time of first mating and interval between matings when a fresh female was provided daily to a male throughout his life time.

Doses (Krad)	First mating (days)	Interval (days)
0	1.5	1.3
25	2.4	3.0
30	2.2	3.0

The time of first mating averaged 1.5 days for the control and 2.4 and 2.2 days for the irradiated males depending on the dosage. The average interval between first and second mating was 3.0 days for the treated males, while it was 1.3 days for the control.

This may mean that the irradiation affects the vigour of mating and/or causes some delay in matings. It was observed that 28% out of the 37.5% of the total matings were made in the first two days in the control group, 15% out of the 30% in the treated groups.

b. Mating ability of the sterile males when 5 or 10 virgin females were provided together in a cage.

Five or ten normal females were confined together with a single freshly emerged male moth in a screen cage (15×15×30 cm) in which rice seedlings were provided.

The male moths were removed after five days confinement. Ten replicates were used. The female moths were preserved in 75% alcohol after their death.

Matings were determined by dissection of the bursa copulatrix. The results obtained are shown in Table 3.

The total number of mated females was greater when 10 females were provided per male than when 5 females were provided, but the percentage of mated females seemed to be smaller with the greater number of females per male in the case of the irradiated males. The average number of matings per male was greater when the greater number of females was provided, especially for the control group.

The average numbers of matings per male were 1.2 and 2.5 respectively for the normal males when 5 and 10 females were provided. No significant increase in total number of matings occured for the treated males, even though a considerable increase in the maximum

Table 3. Number of mated females when 5 or 10 virgin females were provided to one male in a cage for a period of five days.

Doses (Krad)	Mated females	Matings per male			Hatched eggs(%)	Females surviving after 5 days
		Average	Max.	Min.		
			1 : 5			
0	24.0	1.2	2	1	38.14	9
25	34.0	1.7	3	1	8.69	8
30	24.0	1.2	2	0	0	12
			1 : 10			
0	25.0	2.5	4	1	32.28	13
25	18.0	1.8	3	1	7.63	13
30	18.0	1.8	4	1	0	12

number of matings was observed.

2. Mating competitiveness of rice stem borer with different ratios of irradiated and normal males.

Five virgin females were confined with various ratios of irradiated to normal males per female(1 : 0 : 1, 1 : 1 : 1, 2 : 1 : 1, 1 : 2 : 1, 5 : 1 : 1, 1 : 5 : 1, 0 : 1 : .1)

The size and the conditions of the mating cages were the same as in experiment 1-b. Ten replicates were used. The eggs were removed with a piece of the rice leaf on which they had been deposited and placed in a petri dish in which filter paper and moistened roll of dental cotton were provided to maintain moisture.

The results are shown in Table 4.

Table 4. Mating competitiveness of treated males of rice stem borer.

Ratio*	Mated females (%)	No. of eggs/F.	No. of eggs/ egg mass	Hatched (%)	Expected (%)	Adjusted**(%)
			25 Krad			
0 : 1 : 1	55.70	129.3	35.92	78.62	—	—
1 : 0 : 1	50.60	93.6	25.30	24.92	—	18.07
1 : 1 : 1	57.30	88.7	21.63	59.83	50.7	41.04
2 : 1 : 1	57.70	105.6	27.08	57.68	45.0	47.11
1 : 2 : 1	59.60	108.3	25.79	68.42	60.9	57.31
5 : 1 : 1	59.20	119.2	31.37	40.72	34.4	37.54
1 : 5 : 1	65.90	110.3	26.90	65.76	61.6	56.10
			30 Krad			
1 : 0 : 1	45.50	37.6	18.25	0.3	—	—
1 : 1 : 1	53.40	92.3	17.42	42.82	39.0	30.57
2 : 1 : 1	53.70	95.7	19.53	43.62	25.0	32.28
1 : 2 : 1	55.40	93.9	19.78	66.32	50.9	48.16
5 : 1 : 1	54.80	87.8	18.68	26.62	13.7	18.07
1 : 5 : 1	57.90	124.6	23.07	63.28	65.8	60.98

*; Ratio of irradiated: Normal male: Female

**; Hatched eggs as per cents of total number of eggs produced by females mated with normal males(See Text)

The treated males were not fully competitive with untreated males, regardless of the irradiation doses. The greatest reduction of egg hatch obtained with 5 : 1 : 1 (30 Krad), in which 26.6% of the eggs hatched. Competition of the irradiated males has been studied for several insects. Walker and Brindly(1963) reported the results of a study on the effects of X-ray irradiation on *Ostrinia nubilalis* (H.), in which the hatchability of the eggs produced by the females confined with males in the ratio of 8 : 4(sterile: normal males), was 39.4%, while it was 1.1% when the females were mated with irradiated males only (35 Kr of X-ray in adult stage).

Flint and Kressin (1968) reported that in their experiments with *Heliothis verescens* (F.) using a 3 : 1 : 1 ratio of sterile males: normal males: normal females, the hatchability of the eggs produced by the females was 52.4%, while the expected egg hatch was 22.3%, or 30.1% lower.

The data of the present study suggest that the irradiated males were not fully competitive with untreated males, even though the irradiated males were not significantly different in apparent mating behaviour from the normal males. The reduction in the competitive ability of the treated males may be attributed to the reduction in mating vigour suggested by the delayed mating and longer interval between the first and the second matings (Table 3).

Secondly, the calculated reduction in mating competitiveness of the irradiated males may be an indirectly affected from the reduced number of eggs produced by the females mated with the irradiated males, since the percentages of hatched eggs were calculated from the number of hatched eggs and the total number of eggs produced. If the females mated with treated males produce smaller number of eggs than these mated with the normal males, it may result in overestimated hatchability, resulting in an under-estimation of the competitive ability of the irradiated males. Table 5 shows the number of eggs per female obtained from experiment 1-a.

The number of eggs produced by an unmated female was less than the number produced by a mated female regardless of whether she mated with a treated or an untreated male. The average number of eggs laid by mated females was 75.98, while it was 52.07 eggs for the unmated females.

Table 5. The number of eggs produced by a female.

Doses (Krad)	Number of eggs per female*	
	Mated	unmated
0	96.37 (10—235)	57.88(33—150)
25	62.75 (15—110)	49.33(8—215)
30	68.83 (38—178)	49.0 (10—90)
Ave	75.98	52.07

*; Average number of eggs and range in parenthesis.

Flint and Kressin (1969) reported that the virgin females of *H. verescens* (F.) produced an average of 49.6 eggs, while the mated females produced 85.2—438.8 eggs depending on the conditions of the sperm transfer: the mated females without sperm in the spermathecae or stomatophore mated females with sperm in the spermathecae or stomatophore and mated females with sperm in the stomatophore laid 98.6 eggs and 85.2 eggs respectively, while mated females with sperm in their spermathecae laid 438.8 eggs. They concluded that the presence of sperm in the spermathecae, rather than the act of mating, was required for the normal oviposition.

In Table 5, the females mated with treated males produced less eggs than those mated with the normal males. Flint and Kressin (1969) reported that, when the females had crossed with irradiated males, the percentages of mated females without sperm increased (26% to 48%), and the number of eggs per female was 178 for the mated females without sperm, while it was 501 eggs for the mated females with sperm. Thus the transfer of the sperm increased the oviposition of the females.

The data shown in Table 5 suggested a similar trend in oviposition, and such a reduction of the oviposition of females mated with irradiated males might have some relation with the reduction in the appearent competitiveness.

Thus it might be reasonable that in evaluating the mating competitiveness of an insect we should consider both the direct effects of the irradiation: mating vigour, sperm transfer, etc, which are the detrimental effects of the irradiation on mating, and the indirect effects:

sperm transfer, sperm competability, etc, which are likely to reduce oviposition by the females.

The figures shown in the last column of the Table 4 are calculated as the ratio of the hatched eggs to the total number of eggs produced by the females mated with normal males. The figures show that the reduction in competitiveness of the irradiated male seems not to be serious from a practical point of view. This kind of defect can be overcome by release of large numbers of treated males.

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