EVALUATION OF RICE MUTANT LINES FOR RESISTANCE TO BROWN PLANTHOPPER, NILAPARVATA LUGENS STALL

Mugiono

EVALUATION OF RICE MUTANT LINES FOR RESISTANCE TO BROWN PLANTHOPPER, NILAPARVATA LUGENS STALL

Mugiono *

ABSTRACT

EVALUATION OF RICE MUTANT LINES FOR RESISTANCE TO BROWN PLANT-HOPPER, NILAPARVATA LUGENS STALL. The most important and common insect in rice cultivation in South East Asia is brown planthopper, N.lugens Stall. Seven rice mutant lines produced by the National Atomic Energy Agency, Indonesia, were tested at IRRI, the Philippines for resistance to brown planthopper. Those mutant lines were Atomita 1,627/10-3/PsJ, Atomita 2, and 627/4-E/PsJ originated from Pelita I/1 which was irradiated with 0.2 kGy of gamma rays and A227/2/PsJ, A227/3/PsJ and A227 /5/PsJ, originated from early maturing mutant A23/PsJ/72K from irradiated Pelita I/1 which was irradiated with 0.1 kGy of gamma rays. Evaluation of resistance was carried out by seedling bulk screening, honeydew excretion, survival, and population build up tests by using brown planthopper biotype 1, 2, and 3. Results of these tests showed that the seven tested mutant lines were resistant to biotype 1 but susceptible to biotype 2. Reaction to biotype 3 showed that six mutant lines tested were moderately resistant and only one mutant of 627/4-E/PsJ was susceptible. Reactions the mutant lines to biotype 1, 2, and 3 were different from the resistant varieties, Mudgo or ASD-7. This indicated that mutant lines might be have gene(s) for resistance which differed from those of resistant varieties. The results showed that resistance to brown planthopper is possible to be introduced Indonesia rice varieties by means of mutations.

ABSTRAK

PENILAIAN KETAHANAN GALUR MUTAN PADI TERHADAP HAMA WERENG COKLAT, NILAPARVATA LUGENS STALL. Hama yang penting dan umum menyerang pertanaman padi di Asia Tenggara adalah hama wereng coklat, N. lugens Stall. Tujuh galur mutan padi yang dihasilkan oleh Badan Tenaga Atom Nasional, Indonesia dinilai ketahanannya terhadap hama wereng coklat di IRRI, Philipina. Galur-galur tersebut yakni Atomita 1, 627/10-3/PsJ, Atomita 2, dan 627/4-E/PsJ yang berasal dari seleksi varietas Pelita I/1 yang diradiasi dengan sinar gamma dosis 0,2 kGy dan galur A227/2/PsJ, A227/3/PsJ, dan A227/5/PsJ yang berasal dari seleksi mutan genjah

^{*} Centre for the Application of Isotopes and Radiastion, BATAN

Pelita I/1 yaitu A23/PsJ/72K yang diradiasi dengan sinar gamma dosis 0.1 kGy. Penilaian ketahanan dilakukan dengan cara uji kecambah bulk, uji embun madu, uji daya hidup, dan uji pembentukan populasi. Hasil pengujian menunjukkan bahwa ketujuh galur mutan memberikan reaksi tahan terhadap serangan hama wereng biotipe 1 dan rentan terhadap biotipe 2. Reaksi terhadap biotipe 3 menunjukkan, bahwa 6 mutan memberikan reaksi agak tahan dan hanya satu mutan 627/4-E/PsJ memberikan reaksi agak rentan. Reaksi galur mutan terhadap biotipe 1, 2, dan 3 berbeda dengan varietas tahan wereng yang lain seperti Mudgo dan ASD - 7. Ini menunjukkan bahwa mutan tersebut mempunyai gen ketahanan yang berbeda dengan varietas yang tahan. Hasil ini menunjukkan bahwa ketahanan varietas padi di Indonesia yang rentan terhadap hama wereng coklat dapat diperbaiki dengan mutasi.

INTRODUCTION

Rice is the most important staple food for more than three-fourths of the world's population and occupies at almost one-fifth of total world areas under cereals. Nine-tenth of the world's rice production is produced and consumed in Asia.

The humid tropical climate under which rice is mostly cultivated is also conductive to proliferation of insects, many of which are serious pests and cause severe damage to rice production. Together they insect all parts of the plant at all growth stages, and a few transmit virus diseases (1). Among the rice insect pests, brown plant-hopper, N.lugens Stall, has in recent years caused extensive damage to the rice crops in South East Asia. Large scale damage by brown planthopper has been reported in India, Indonesia, the Philippines, Sri Lanka, and Bangladesh (2).

The brown planthopper may also transmit virus diseases such as grassy and rugged stunt which can further reduce the crop yield (3,4).

The use of insecticide is considered as a practical method to prevent rice plants against this insect damage. However, the high cost

of insecticides and also the fact that insects feed at the base of teh rice plants, the use of insecticide can not effectively control this pest (5).

The uses of resistant varieties cause a reduction of insect population in the field. However, one major disadvantage associated with their use is the development of new biotypes which are able to overcome the resistance (6).

A large number of varieties were screened for its resistance to brown planthopper and several resistant varieties have been indenti-fied (7, 8, 9).

Many resistant varieties have been used in the IRRI's breeding program and many resistant breeding lines and released varieties have already developed (10).

No resistant varieties of Indonesian origin have been found and therefore Indonesia will have to depend on the introduced varieties either for direct production or for parental materilas in cross breeding. Resistant varieties from IRRI have been used in Indonesia.

The objective of the experiment was to investigate whether brown planthopper resistance can be introduced in Indonesian rice varieties by mutations.

Screening for resistance of mutant lines of the National Atomic Energy Agency, Indonesia, to brown planthopper has been conducted since 1976 and the reactions of some mutant lines will be reported.

MATERIALS AND METHOD

Seven mutant lines, Atomita 1 *, 627/10-3/PsJ, Atomita 2 *, and 627/E-4/PsJ, originated from Pelita I/1 which was irradiated with 0.2 kGy of gamma rays and A227/2/PsJ, A227/3/PsJ, and A227/5/PsJ, originated from early maturing mutant A23/PsJ/72K from irradiated Pelita I/1 which was irradiated with 0.1 kGy of gamma rays were used in these studies. Screening for resistance were conducted at IRRI's, the Philippines by the following methods.

apart in rows in 60x45x10 cm seed boxes. Each mutant line was placed randomly in a 10 cm long row in 3 replications. Each replication was placed in nylon cage with a nylon mesh top. The experiment of bulk screening test was conducted in a Complete Randomized Block Design (Figure 1). A susceptible and resistant check variety was planted randomly in each replication. The resistant checks were Mudgo for brown planthopper biotype 1 and 3, and ASD-7 for biotype 2. TN-1 variety was used as a susceptible check for all biotypes. Seven days after sowing, seedlings were infested with 2nd or 3rd instar nymphs obtained from the mass culture. Seedlings were uniformly infested as possible so that each seedling received 5-6 insects. Recording of the reaction was done when 90% of the susceptible check was killed. This rating was based on the following 0-9 visual grading scale which was the Standard Evaluation System for the brown planthopper (11).

^{*} New improved variety released by the Ministry of Agriculture in 1982 and 1983.

Rep. II Rep. III Rep. I G B H B E I F E F I C H C D A G D C G B H В E A C G D A E H D H J I F C I C G A C H F E I B D IEA J D H _A C В D E B

Fig. 1. Lay out of the experiment in the seedling bulk screening test.

A. Atomita 1
B. 627/10-3/PsJ
C. Atomita 2
D. 627/4-E/PsJ
E. A227/2/PsJ
J. ASD-7

2. Honeydew excretion test. Seeds of the mutant lines were sown in seed boxes of 60x45x10 cm. Seven days old seedlings were transplanted in clay pots, size 0 ($\emptyset = 6$ cm), 5 pots each variety. At thirty days after transplanting, 5 pots of each variety were prepared for the collection honeydew as shown in Fig. 2. First, the outer leaf sheath that was lost from the stem was cut at its base to prevent it from coming in contact with filter paper. The petri-dish was fixed in place by guiding the plant through the center hole. Treatment and replication markings were written on the filter paper with a pencil. The filter paper that will be used in this experiment has been treated

with bromocresol green solution (0.2 g/l ethanol). Forceps were used to avoid contact of the paper by moist hands. The plastic cup was then placed in an inverted position and the leaves were pulled through the center hole. The cup was held in place with tape. Five days old adult females were starved for 5 hours in container containing moist filter paper. Five adults were then introduced into each feeding chamber through the hole at the top of the cup and the hole was plugged with cotton. After 24 hours, the filter papers were removed and the area (mm²) of the honeydew spots provided an indirect measure of the feeding activity. Tracing paper was placed over graphpaper (mm²) and the number of square occupied by the spots was counted.

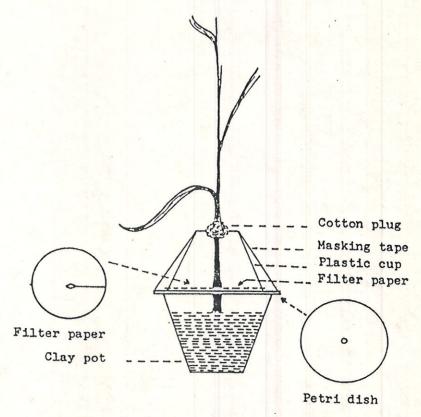


Fig. 2. Apparatus used for collecting honeydew in feeding studies.

- 3. <u>Survival of nymphs test.</u> Three, seven day seedlings were transplanted in clay pots size 3 (\emptyset = 10 cm), 5 pots each variety. At 30 days after transplanting, the plants were covered with a 8x70 cm nylon cage with a fine mesh screened window and 10 newly emerged nymphs of brown planthopper were placed in each cage. Insects were counted at 5, 10, 15, and 20 days after infestation.
- 4. Population buildup. Three, seven day old seedlings were transplanted in clay pots size 6 (\emptyset = 15 cm), 5 pots each variety. At 40 days after transplanting, the plants were covered with a 13x90 cm mylar cage with fine mesh screened windows and 10 newly emerged nymphs were placed in each cage. Number of insects were counted at 40 days after infestation and weight of insects were determined after the insects have been dried in the oven for 48 hours. The design for this experiment was a Complete Randomized Design with 5 replications.

RESULTS AND DISCUSSION

The reactions of mutant lines in the seedling bulk screening test to brown planthopper biotypes 1, 2, and 3 are given in Tabel 1. There were distinct differences in the reaction of mutant lines from the different biotypes of brown planthopper. All of the mutant lines were resistant to biotype 1 with a damage rating of 3 to 3.6, while the susceptible check TN-I and resistant check Mudgo were 9 and 2.3, respectively.

Reaction of the mutant lines to biotype 2 showed that all mutant lines were susceptible with a damage rating of 7. However, 2 mutant lines, Atomita 1 and A227/2/PsJ received a damage rating of 6.3.

Reaction of mutant lines of biotype 3 showed that all mutant lines were moderate resistant with a damage rating of 5 to 5.6, and only one mutant line, 627/4-E/PsJ, was susceptible with a damage rating of 7.

Based on the damage rating in Table 1, seemed that levels of resistance of the mutant lines were not similar with those of the resistant varieties, Mudgo or ASD-7. According to ATWAL et al. (12), Mudgo is resistant to biotypes 1 and 3, while ASD-7 is resistant to

biotypes 1 and 2. However, the mutant lines were only resistant to biotype 1 and moderate resistant to biotype 3.

These results indicated that the gene for resistance in the mutants is obviously different from the resistance gene of Mudgo.

The results of honeydew in the feeding studies are shown in Table 2 and Fig. 3. Significant differences were recorded in the amount of honeydew excreted from the mutant lines compared with the Mudgo or ASD-7, and the susceptible check TN-1.

The amount of honeydew excreted in the mutant lines by biotype 2 was much larger than that of biotypes 1 and 3 and the excretion produced by biotype 3 was larger than that of biotype 1. These findings indicated that biotype 1 was not suitable to feed on the mutant lines. While biotypes 2 and 3 were able to feed the mutant lines during the testing periode, but not as much as that of the susceptible check.

The amount of honeydew excreted in all mutant lines by biotype 1 was significantly different from that of the susceptible check TN-I but not significantly different from the resistant check, Mudgo and ASD-7. From the results of biotype 2 feeding test, all mutant lines were significantly different from the susceptible check TN-I, Mudgo,

and resistant check ASD-7. However, in the biotype 3 test, some mutant lines were not significantly different compared with the resistant check Mudgo except for the mutant line A227/2/PsJ, which with all were significantly different from TN-I or ASD-7.

According to LEE and PARK (13), the amount of honeydew excreted by brown planthopper is less when feeded on a resistant variety than on a susceptible one. Based on this assumption on the mutant lines were resistant to biotype 1, moderately resistant to biotype 3, and moderately susceptible to biotype 2.

The results of survival test were presented in Table 3 and Fig.4. Survival of nymphs at 20 days (Table 3) infestation was less on all mutant lines compared with the susceptible check TN-1. Survival of nymphs of biotype 1 on all mutant lines were significantly different from the susceptible check TN-1 and the resistant check Mudgo, except for the mutants of Atomita 1, 627/10-3/PsJ, and A227/5/PsJ no signiticant differences were found compared with the resistant check Mudgo.

After fifteen days of infestation the survival of nymphs of biotype 1 on all mutant lines was significantly reduced, indicating that all of the mutant lines possessed a strong antibiosis or resistance to biotype 1 (Result after 20 days are listed in Table 3).

Survival of nymphs of biotype 2 on mutant lines was significant-ly different from the resistant check, ASD-7, but not from the sus ceptible check TN-1, indicated that all mutant lines were susceptible to biotype 2. Survival of nymphs of biotype 3 on some mutant lines were not significantly different from the resistant check, Mudgo. The mutant lines 627/4-E/PsJ, A227/2/PsJ, and A227/5/PsJ possessed a lower antibiosis effect compared with Mudgo variety.

Number and weight of insects in the population buildup were presented in Tables 4 and 5, Fig. 5 and 6. On all mutant lines signifi cantly smaller populations of biotypes 1 were observed compared with that on the susceptible check TN-1. However, in the biotype 2 screening, higher populations were developed on all mutant lines but no significant differences compared with susceptible check, TN-1 or Mudgo were observed. Also in the biotype 3 test the number of insect on all mutant lines were higher than that on Mudgo , but significantly lower compared with the susceptible checks TN-I and ASD-7. Susceptible check varieties offered more favorable condition for insect multiplications than the resistant ones. The insects on the susceptible varieties were able to complete their generation in a short time and started with the second and succeeding generations earlier and larger compared with a those of the resistant varieties and most mutant lines. It was particularly interesting to note that the reactions of the mutant lines towards the population buildup of biotype 3 was significantly lower than those of the susceptible checks (TN-1 and ASD-7). The insects showed a slower growth rate and lower survival on these mutant lines. The production and hatching of eggs was reduced, which would explain why the population showed a slower crease compared with the susceptible checks. However, in regard with the biotype 1. most mutants had a greater capacity to interrupt the insect's population buildup and some proved to be as resistant as Mudgo.

The dry weight of insects in the population buildup (Table 5) was also a good measure for effective growth. The dry weight of biotype 1 insects on Atomita 1 , 627/10-3/PsJ , A227/4-E/PsJ , A227/3/PsJ, and

A227/5/PsJ were not significantly different from the resistant check Mudgo, but Atomita 2 and A227/2/PsJ showed a significant difference from the resistant check. The dry weight of biotype 2 insects on all mutant lines were significantly different from the resistant check ASD-7. In the biotype 3 test, the dry weight of insects of all mutant lines were significantly different from the susceptible check TN-1. Only Atomita 1 and 2 showed a significant difference from Mudgo. These results indicated that biotype 3 insects were not able to grow normally on the mutant lines although these mutant lines were not resistant to biotype 3.

CONCLUSIONS

Seven mutant lines were tested for evaluation or resistance to brown planthopper biotypes 1, 2, and 3. All of the mutant lines indicated to be resistant to biotype 1 but susceptible to biotype 2. It was proved that six mutant lines were moderatelly resistant to biotype 3 and only mutant line, 627/4-E/PsJ was susceptible.

Reactions of the mutant lines to biotype 1, 2, and 3 differed from those of Mudgo and ASD-7, and probably the mutants had different genes for resistance.

Results of the other test, showed that honeydew excretion excretion, survival, population buildup, and dry weight of insects seemed to have different characteristic between the mutant lines and the check varieties. There were also interesting differences among the mutants in its reaction to biotypes 1, 2, and 3 in various tests. The data in Table 2, 3, 4, and 5 indicated that the mutant lines seemed

to have moderate reactions to these tests, which might indicate symptoms of tolerance.

It is encouraging that the released varieties, Atomita 1 and 2 seem to be very promising in these respects. Extensive trials in field experiments will prove the practical significance of the observations.

ACKNOWLEDGEMENTS

The author wishes to acknowledge the UNDP and IAEA for fellowship grant to IRRI, which made this research possible.

The author wishes to express special thanks to Dr.E.A. Heinrichs and Mr. Fill Medrano for their ancouragements and valuable advise.

Thanks are also due to Mr. Ising Domingo for his valuable technical assistance.

Also most grateful acknowledgement to Dr. K. Mikhaelsen, for his advise in preparing the manuscript and to Dr. Moch. Ismachin and Mr. Hendratno, M.Sc. for their ancouragement.

REFERENCES

- 1. PATHAK, M.D., Ecology of common insect pest of rice, Ann. Rev. Entomol. 13 (1968) 257.
- 2. DYCK, V.A., "The brown planthopper problem", Brown Planthopper Symposium, Int. Rice Res. Conf., IRRI, Los Banos, Philippines, April 18-22 (1977).
- 3. RIVERA, C.T., S.H. OU, and IIDA, Grassy stunt disease of rice, transmission by the planthopper, *Nilaparvata lugens*, Plant Disease Reptr. 7 50 (1966) 453.
- 4. LING, K.C., Ragged stunt disease, IRRN 5-6-7 (1977).
- 5. HEINRICHS, E.A., "Chemical control of brown planthopper", Brown Planthopper Symp., Intl. Rice Res. Conf., IRRI, Los Banos, Philippines, April 18-22 (1977).

- 6. GLASS, E.H., Integrated pest management. Radtionale, potential weeds and improvement, ESA Special Publ. 75 2 (1975) 141 p.
- 7. PATHAK, M.D., CHENG, C.H., FORTUNE, M.E., Resistance to Nepothetic impiticeps and Nilaparvata lugens in varieties of rice, Nature 223 (1969) 502.
- 8. PATHAK, M.D., "Resistance to insect pest in rice varieties Rice Breeding, Intl. Rice Res. Inst., Los Banos, Philippines (1978) 738 p.
- 9. PATHAK, M.D., and KUSH, G.S., "Studies on varietal resistance to brown planthopper at IRRI", Brown Planthopper Symp., Intl. Rice Res. Inst. Conf., Los Banos, Philippines, April 18-22 (1977).
- 10. KUSH, G.S., and BICHEL, H.H., "Breeding for disease and pest resistance", Rice Breeding, Intl. Rice Res. Inst., Los Banos, Philippines (1972) 738 p.
- 11. IRRI, Standard Evaluation System for Rice, Intl. Rice Testing Program, IRRI, Los Banos, Philippines (1980).
- 12. ATWAL, D.S., PATHAK, M.D., BACALANGO, E.H., and PURA, C.D., Genetics of resistance to brown planthopper abd green leafhopper in Oryza sativa L., Crop Science 11 (1976) 747.
- 13. LEE, J.O., and PARK, J.S., Studies on varietal resistance of rice to the plant and leafhopper (in Korean), Res. Rep. (Soil Sci., Fert., Plant, Prot., Micol.) 18 (1976) 67.

Table 1. Reaction of mutant lines to brown planthopper in seedling bulk screening test.

No.	No. of line	Damage rating *)		
		Biotype 1	Biotype 2	Biotype 3
A. B. C. D.	Atomita 1 627/10-3/PsJ Atomita 2 627/4-E/PsJ A227/2/PsJ	3 3.6 3.6 3	6.3 7 7 7 6.3	5 5 7 5,6
F. G. H. J.	A227/3/PsJ A227/5/PsJ TN-I Mudgo ASD-7	3 3 9 2.3	7 7 9 - 3	5.6 5.6 9 1.6

^{*)} Average of 3 replications
Damage rating by Standard Evaluation System

Table 2. Amount (mm²) of honeydew excreted in feeding studies *).

No.	No. of line	Biotype 1	Biotype 2	Biotype 3
Α.	Atomita 1	12.0 b	237.0 d	128.8 cd
В.	627/10-3/PsJ	16.6 b	262.0 d	127.8 cd
C.	Atomita 2	17.0 b	254.0 d	143.0 cd
D.	627/4-E/PsJ	22.0 b	382.0 c	156.2 cd
E.	A227/2/PsJ	28.6 b	186.2 d	190.8 c
F.	A227/3/PsJ	50.4 b	237.0 d	114.2 cd
G.	A227/5/PsJ	37.4 b	188.4 d	120.4 cd
Н.	TN-I	801.8 a	570.4 a	890.0 a
1.	Mudgo	11.8 b	463.4 b	7.0 d
J.	ASD-7	15.8 b	36.0 e	454.6 b

^{*)} Average of 5 replications.

Means in column followed by common letter are not significantly different at 5% level by DMRT.

Table 3. Percentage survival of brown planthopper in survival and development of nymphs test *).

No. No. of line		Survival (arcin scale)**)		
		Biotype 1	Biotype 2	Biotype 3
A. B. C. D.	Atomita 1 627/10-3/PsJ Atomita 2 627/4-E/PsJ A227/2/PsJ	50.99 bc 48.68 bc 55.71 b 55.88 b 57.04 b	62.40 a 73.15 a 65.18 a 74.35 a 68.13 a	54.55 bc 51.09 bc 52.37 bc 64.02 ab 64.02 ab
F. G. H. J.	A227/3/PsJ A227/5/PsJ TN-I Mudgo ASD-7	55.88 b 52.20 bc 68.31 a 41.48 c	63.73 a 65.18 a 78.93 a - 35.44 b	61.20 bc 62.82 ab 73.62 a 42.69 c

^{*)} Average of 5 replications

Table 4. Number of insects in population buildup *).

No.	No. of line	Biotype 1	Biotype 2	Biotype 3
A. B.	Atomita 1 627/10-3/PsJ	74.8 c 120.0 c	504.0 a 531.4 a	263.2 cd 227.6 cd
C. D. E.	Atomita 2 627/4-E/PsJ A227/2/PsJ	228.6 b 63.6 c 102.6 c	451.2 ab 597.8 a 577.8 a	300.0 bc 253.6 bc 135.2 cd
F. G. H. J.	A227/3/PsJ A227/5/PsJ TN-1 Mudgo ASD-7	81.0 c 130.0 bc 464.8 a 75.4 c 59.6 c	449.2 ab 363.8 ab 604.6 a 544.4 a 145.2 b	146.8 cd 146.2 cd 594.2 a 76.6 d 404.4 b

^{*)} Average of 5 replications

Means in a column followed by a common letter are not significantly different at 5% level by DMRT

^{**)} Data transformed based on arcin transformation

Means in a column followed by a common letter are not significantly different at 5% level by DMRT

Table 5. Dry weight of insects (mg) in population buildup *).

No.	No. of line	Biotype 1	Biotype 2	Biotype 3
A. B. C. D.	Atomita 627/10-3/PsJ Atomita 2 627/4-E/PsJ A227/2/PsJ	29.48 cd 38.79 cd 101.20 b 26.78 cd 55.68 c	119.36 bcd 220.90 a 189.83 ab 165.38 abc 157.30 abc	79.00 cd 54.20 cde 88.21 c 64.46 cde 21.48 e
F. G. H. J.	A227/3/PsJ A227/5/PsJ TN-I Mudgo ASD-7	25.92 cd 27.70 cd 136.08 a 17.61 d 16.90 d	100.34 de 65.37 de 168.41 abc 152.20 abc 34.30 e	15.99 e 30.75 de 162.62 a 17.42 e 154.92 ab

^{*)} Average of 5 replications

Means in a column followed by a common letter are not significantly different at 5% level by DMRT

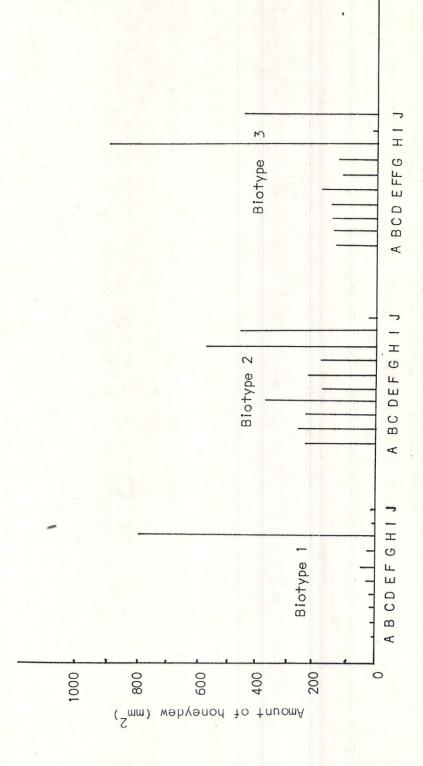


Fig. 3. Amount (mm²) of honeydew excreted in feeding studies.

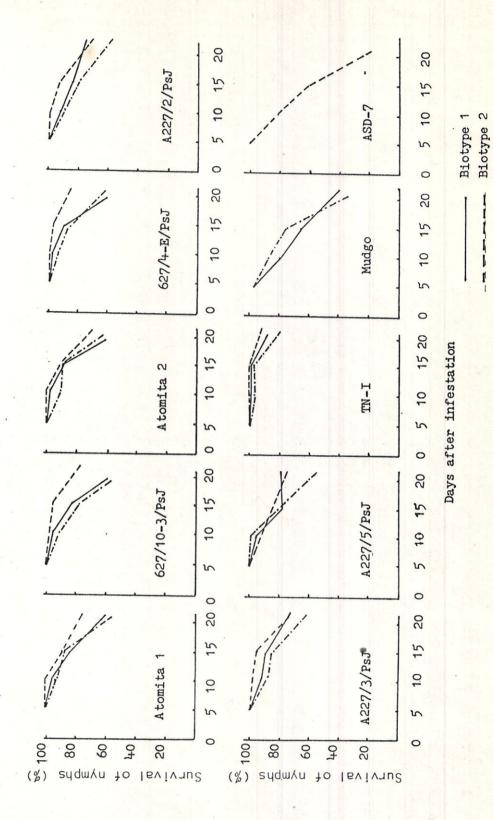


Fig. 4. Survival of nymphs on different rice mutant lines and check varieties.

Biotype 3

11.