

## New Mutant Cotton Lines of Good Yields and Resistance to Some Insects by $\gamma$ - Irradiation

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### ABSTRACT

$M_2$  through  $M_6$  of the  $\gamma$ -irradiated mutant lines,  $AP_1$  and  $AP_2$  were tested for insect resistance and good yields comparing to the control varieties,  $SR_2$  and  $R_1$ , in field and laboratory conditions. Field tests at Suwan Farm revealed seed weight, lint weight and % lint of  $AP_1$  to be the greatest while the highest weight of 10 fresh bolls was found in  $AP_2$ . Fiber quality of all varieties/lines were found to be similar and were within the rather high standard levels. Visual rating of damage caused by *H. armigera* (square damage) and *A. biguttata* (hopper burn) to the controls and mutants were similarly rated as moderately resistant and susceptible respectively. According to antibiotic test and chemical analysis of *H. armigera*, the small increased larval weight and the morphologically abnormal adult fed from leaves of  $AP_1$  and  $AP_2$  were believed to be partially caused by other chemical substances beside gossypol and flavonoids whose contents did not exceed the given toxic level inhibiting growth.

**Key words :** cotton, irradiation, mutant, insect resistance, *Heliothis armigera*, *Amrasca biguttata*

### INTRODUCTION

Aston and Winfield (1972) listed 46 groups of insects known to occur in cotton throughout the world; 42 are classified as economical importance in one or more of the cotton producing nations. Extensive studies have been made on antibiosis as a source of resistance to *Heliothis spp.* and Lukefahr *et al.* (1966) discussed utilization of high bud gossypol as a source of resistance. Plant breeding by radiation has long been undertaken and Saric (1961) reported that the irradiated plant seeds yielded differently depending on biological characters of the seeds. The appropriate dose for each plant must, therefore, be determined. Mutation breeding of cotton by radiation has been satisfactorily conducted in many countries, such as, Russia, Egypt, India, Pakistan, China, most of which were aimed at agronomic improvement. Iqbal *et al.* (1991) found cotton variety NIAB-78 irradiated with  $\gamma$ -rays became resistant to spotted bollworm and pink bollworm. Premskar (1986)

irradiated cotton pollen with 0.5 krad  $\gamma$ -rays and crossed them with cotton line 1412 which produced sticky hair on the leaves giving resistance to the jassid, *Amrasca biguttata*, with 7-30% more yield in  $M_6$ . The objective of the study is to use  $\gamma$ -radiation in inducing mutation of cotton variety  $R_1$  with hope that the mutant lines will possess more antibiotic resistance against the cotton insects and at the same time produce yield of good quantity and quality.

### MATERIALS AND METHODS

#### Field test

The experiment was undertaken during 1994-96 at Suwan Farm. The  $\gamma$ -irradiated mutant lines from  $M_2$  through  $M_6$  of  $AP_1$  and  $AP_2$ , were tested on insect resistance and yields against the control varieties,  $SR_2$  and  $R_1$ . RCB with 4 replicates was employed. Each replicate consisted of 2 control varieties and 2 mutant lines, each of 20 m long with spacing of row x plant = 0.5 x 1.0 m. Tagging was

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randomly made from 4 middle rows, 2 plants/row. Kind and number of insects were checked from the tagged plants for 4 consecutive weeks. Visual rating on damaging levels by *Heliothis armigera* and *Amrasca biguttata* from damaged squares and hopper burn, respectively, were also made out of tagged cotton. The average of 3-year data on quantity of yield as weight of 10 fresh bolls, weight of seeds and lint and % lint were checked while quality in micronaire, length, strength and uniformity of fiber were evaluated.

#### Antibiotic test

Bollworm larvae (2 days old) of uniform size were selected from the laboratory colony and individually weighed, then 1 larva was placed in each of 160 cups arranged in RCB. The test consisted of 4 replicates with 10 cups/control varieties (SR<sub>2</sub> & R<sub>1</sub>) and tested lines (AP<sub>1</sub> and AP<sub>2</sub>), 4 varieties and tested lines/replicate. Young terminal leaves removed from the plants and washed in a 0.15% sodium hypochlorite solution for a few minutes. Then the leaves were rinsed with water and dried. A few leaves of each variety/line were placed in each cup with moistened filter paper lining at the bottom. After 48 hours of feeding with young leaves, the larvae were reweighed and continued to be fed with artificial diet until all adults emerged. The following data were recorded for each insect : the increased larval weight after feeding on leaves for 48 hours, larval length, pupal weight, % pupation, % adult emergence and % morphologically abnormal adults. The average data of 3 years (1994-96) were reported.

#### Chemical analysis

Young leaves were removed from each variety/line, put in plastic bags and kept in the freezer for 2 days before being lyophilized at low temperature. The dried leaves of each one were finely ground with blender, put back into sealed plastic bags, kept in the refrigerator before sending out for chemical analysis at Mississippi Chemical Lab.

## RESULTS AND DISCUSSION

#### Field test

The field test of 3-year average from sampled plants found seed and lint weights of AP<sub>1</sub> to be the highest (Table 1) and significantly differed from the others while the highest weight of 10 fresh boll was noticed in AP<sub>2</sub> and significantly differed from AP<sub>1</sub> and SR<sub>2</sub>. However, % lint of the controls and mutant lines were similarly fallen within satisfactory level (above 35%) with AP<sub>1</sub> the highest.

According to HIV analysis of fiber quality, Table 2 shows the fiber lengths of the control varieties and the mutant lines to be of medium with AP<sub>2</sub> the highest. Micronaire, fiber strength and UI of all varieties/lines were determined to be similarly under desirable values, high designated group and very high designated group, respectively. Results of the analysis were found to be similar to those of some cotton cultivars evaluated at Weslaco, Texas by Smith *et al.* (1994), which apparently implied Thai cotton fiber to be of equal quality to US cotton fiber.

**Table 1** Average agronomic performance of cotton varieties/lines from sampled plants at Suwan Farm during 3 consecutive years of 1994-96.

Var/Line	Seed Wt. (g)	Lint Wt. (g)	% Lint	Wt. of 10 fresh bolls (g)
SR <sub>2</sub>	164.0 b	97.0 b	39.9	181.32 a
R <sub>1</sub>	135.6 a	80.1 a	38.6	196.50 b
AP <sub>1</sub>	170.1 c	116.5 c	41.3	182.16 a
AP <sub>2</sub>	143.0 a	94.5 b	40.4	199.10 b

Means not followed by the same letters differ significantly as determined by DMRT  
(p = 0.05)

Table 3 and 4 show visual rating of damages caused by *Heliothis armigera* (square damage) and *Amrasca biguttata* (hopper burn) of the controls and mutants to be moderately resistant and susceptible, respectively. It is not that unusual for the plant to be resistant to some pests while susceptible to the others. The glabrous types of cotton, resistant to *Heliothis spp.* and the other pests, are susceptible to the cotton leafworm, *Spodoptera littoralis* (Kamel, 1965) and jassid, *Empoasca spp.* (Reed, 1974). In the study, the emphasis was on resistance to *H. armigera*, one major cotton insect pest, since the insect is quite

difficult to be controlled by other methods, whereas *A. biguttata* could still be controlled by chemicals. However, the development of multiple resistance to more kinds of insects in one plant should be pursued in the future.

It was also noticed that the plant bug, *Megacoelum biseratense* (new record of this genus and species in Thailand) started to become serious pest lately since the insect caused a lot of flower buds to drop as much as did by *H. armigera*. The tarnished plant bug, *Lygus lineolaris* and *Lygus hesperus* were found to be the economic cotton pests in the eastern half of US cotton belt and the

**Table 2** Comparison of average fiber quality of 4 cotton varieties/lines during 3 consecutive year (1994-1996).

Var/Line	Fiber length <sup>1/</sup> (inch)	Micronaire <sup>2/</sup> (unit)	Fiber strength <sup>3/</sup> (g/tex)	UI <sup>4/</sup> (ratio)
SR <sub>2</sub>	1.03	4.3	27.0	85.3
R <sub>1</sub>	1.05	4.1	26.2	85.6
AP <sub>1</sub>	1.06	3.8	27.3	86.2
AP <sub>2</sub>	1.10	3.9	26.9	85.0

  

<sup>1/</sup> Fiber length (in.) and descriptive designation	<sup>2/</sup> desirable values = 3.5-4.9, best value = 3.9-4.2		
Below 0.97	short		
0.97 - 1.10	medium		
1.11 - 1.28	long		
<sup>3/</sup> $\frac{1}{8}$ -inch gauge strength (g/tex)	<sup>4/</sup> Uniformity ratios and descriptive designation		
	Fiber length group and descriptive designation		
	Medium (0.97 - 1.10 inch)		
20 - 22	low	77 - 79	low
23 - 25	average	80 - 82	average
26 - 28	high	83 - 85	high
		> 85	very high

**Table 3** Visual rating and level of resistance of square damage caused by *H. armigera* from sampled plants of each var/line, the 3-year average at Suwan Farm (1994-1996).

Var/Line	Damaging level	Resistant level
SR <sub>2</sub>	1.7	moderately resistant
R <sub>1</sub>	1.5	moderately resistant
AP <sub>1</sub>	1.3	moderately resistant
AP <sub>2</sub>	1.2	moderately resistant

**Table 4** Visual rating and level of resistance of hopper burn caused by *A. biguttata* from sampled plants of each var/line, the 3-year average at Suwan Farm. (1994-1996)

Var/line	Damaging level	Resistant level
SR <sub>2</sub>	3.3	susceptible
R <sub>1</sub>	2.8	susceptible
AP <sub>1</sub>	2.8	susceptible
AP <sub>2</sub>	2.9	susceptible

Damaging and Resistant Levels (1 = resistance, 2 = moderately resistance, 3 = susceptible, 4 = highly susceptible)

irrigated area of western US, respectively (Niles, 1980). Due to the similarity of the environments of cotton growing regions of the 2 countries, there is a chance that the tarnished plant bug in Thailand will soon cause great damage to the cotton as occurring in USA. The other kinds of insects, such as aphids, whiteflies, pink bollworm, cotton stainer, etc. were also encountered but in small numbers only. Damages caused by these insects were not apparent, which was possibly contributed to antibiosis resistance of the cotton.

#### Antibiotic test and chemical analysis

Laboratory results (Table 5) showed the increased larval weight and larval length of AP<sub>1</sub> and AP<sub>2</sub> to significantly differ from both controls whereas only pupal weight of AP<sub>1</sub> was found to be significantly different from the other varieties/lines. As for pupation, the 2 mutant lines were

found to be a little higher than those of the controls while % adult emergence were the same to all. It was also revealed that 35% of the emerging adults from AP<sub>1</sub> and AP<sub>2</sub> were morphologically abnormal, such as twisted wings, deformed legs etc., while none was encountered in the controls. The adult abnormality and small increased larval weight of the mutant lines may be due to the toxicity of some chemical substances in the leaves which AP<sub>1</sub> and AP<sub>2</sub> had at higher content than SR<sub>2</sub> and R<sub>1</sub>.

But since chemical analysis of leaves of the controls and mutants showed % gossypol and flavonoids, the most toxic substances in cotton (Hedin *et al.*, 1983), to be not much different from one another (Table 6) and did not exceed 1.2%, (in case of gossypol) the level supposed to inhibit larval growth as stated by Shaver *et al.* (1970). The

**Table 5** Average antibiotic effect of cotton varieties/lines leaves on selected developmental parameters of *H. armigera*, 1994-96.

Var/Line	Mean increased larval wt (g)	Mean larval length (day)	Mean pupal wt (g)	% <sup>1/</sup> pupation	% adult emergence
SR <sub>2</sub>	0.016 b	19.4 b	0.28 b	40	40
R <sub>1</sub>	0.010 b	20.5 b	0.30 b	40	40
AP <sub>1</sub>	0.007 a	18.0 a	0.26 a	60	40 <sup>2/</sup>
AP <sub>2</sub>	0.008 a	18.5 a	0.29 b	50	40 <sup>2/</sup>

Means not followed by the same letters differ significantly as determined by DMRT

(p = 0.05)

<sup>1/</sup> expressed as percentage of 10 larvae fed on each var or line/rep.

<sup>2/</sup> 35% of emerged adults were morphologically abnormal.

**Table 6** Content of gossypol and flavonoid in cotton leaves of control varieties and mutant lines.

Var/Line	Gossypol (%)	Flavonoids (%)
SR <sub>2</sub>	0.27	0.99
R <sub>1</sub>	0.19	0.85
AP <sub>1</sub>	0.26	0.90
AP <sub>2</sub>	0.31	0.89

above results, therefore might be partially caused by the action of other chemicals. The extraction of other substances beside gossypol or separate bioassay study of each kind of flavonoids such as quercetin, rutin, etc., must be furtherly investigated. Yet, it was also evident that gossypol and flavonoids in every control variety/line caused more or less growth reduction to the tested insects.

Although the source of antibiotic resistance to *H. armigera* in the 2 mutant lines has not been known yet, the lines can successfully transfer the resistant trait from M<sub>2</sub> through M<sub>6</sub>. Evidently AP<sub>1</sub> and AP<sub>2</sub> express similar or even better results compared to those of SR<sub>2</sub> and R<sub>1</sub> in terms of resistance to some insects and quantity/quality of yields. However, to use the resistant lines as principal control method is not quite effective since, as already mentioned, the cotton has a great deal of pests at every stage of development, the integrated control may, thus, be more practical in order to ultimately obtain maximum good yield.

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