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FORBEARANCE OF GENETICALLY DISTANT COTTON HYBRIDS TO BOLLWORM (*HELICOVERPA ARMIGERA*) IN DEPENDING OF THE LEVEL OF GOSSYPOL IN SEEDS

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Мақолада ғўза чигитидаги госсипол миқдорлари ва шаклларининг ғўза тунлами (Helicoverpa armigera) билан зарарланиш даражасига таъсири генетик-селекцион жиҳатдан асослаб берилган, турли миқдордаги ва шаклдаги госсиполга эга нав-намуналар иштирокида олинган дурагайларда ғўза тунламига бардошлиликнинг шаклланиши, чигитида (+)-госсипол миқдори ҳамда нисбатан ғўза тунламига бардошлилиги турлича бўлган селекцион ашёларни яратиш мумкинлиги келтирилган. Паст миқдордаги (+)-госсиполли генотиплар маълум даражада юқори миқдордаги (+)-госсиполли дурагайларга нисбатан камроқ зарарланганлиги аниқланди.

Калит сўзлар: *ғўза, госсипол, зарарланиш, авлод, Helicoverpa armigera, дурагайлаш, кўсак, бардошлилик, чигит.*

В статье представлены полученные в естественных условиях данные о толерантности старших поколений гибридов хлопчатника к хлопковому совку (Helicoverpa armigera) с различным уровнем (+)-госсипола в семенах. На основании полученных результатов выявлено, что уровень (+)-госсипола в семенах не обладает значительной устойчивостью к совку, т.е. подтверждена возможность отбора новых семей и линий хлопчатника с различными уровнями (+)-госсипола и толерантностью к совку. Генотипы с низким уровнем (+)-госсипола в определенной степени меньше поражались, чем гибриды с высоким (+)-госсиполом.

Ключевые слова*: хлопчатник, госсипол, повреждение, поколение, Helicoverpa armigera, гибридизация, коробочка, устойчивость, семена.*

This paper presents obtained data on the natural background about tolerance of senior generations of cotton hybrids to bollworm (Helicoverpa armigera) with different levels of (+)-gossypol in seeds. It is discovered that the level of (+)-gossypol in seeds does not significantly show resistance to cotton ballwor. It confirmed possibility of selecting new sorts and lines of cotton with different levels (+)-gossypol and tolerance to bollworm. Genotypes with a low level of (+)-gossypol affected by bollworm is less to a certain extent than hybrids with high (+)-gossypol level.

Keywords: *cotton, gossypol, damage, generation, Helicoverpa armigera, hybridization, bolls, resistance, seeds.*

Cotton varieties cultivated in our republic in terms of precocity, yield, fiber quality and other certain agronomy-valuable traits have an advantage over foreign variety samples. But recent environmental changes and the emergence of new populations of various pests set new challenges for scientists. Cottonseed provides a high quality protein that is currently under utilized because of the presence of a toxic compound called gossypol. Gossypol is biosynthesized by the free radical coupling of two molecules of hemigossypol. During this coupling reaction, two optically active enantiomers are formed. One of these is referred to as (+)-gossypol and the other as (-)-gossypol.

Cotton bollworm (*Helicoverpa armigera*) is one of the most dangerous cotton pests and is considered to be an urgent problem in the global sphere. Cotton bollworm (*Helicoverpa armigera*) is not only one of the most dangerous cotton pests, but also one of the most multifaceted and common pests of many crops.

In cotton (*Gossypium*) the ratio of (+)- to (-)-gossypol can vary from 98:2 to 31:69 in seed (Cass et al., 1991; Percy et al., 1996). Within the genus *Gossypium*, were found accessions from several species that have >92% (+)-gossypol in the seed. These include *G.mustelinum*, *G.anomalum*, and G.*gossypioides* (Stipanovic et al., 2005). Cass et al. (1991) first reported that *G.barbadense* had an excess of the (-)-enantiomer in the seed. It was found that some accessions of *G. darwinii*, *G.sturtianum*, *G.harknessii*, *G.longicalyx* and *G.costulatum* also produce an excess of (-)-gossypol in the seed [1].

Gossypol, which is found in pigment glands, was identified in 1915 (Withers and Carruth) as the tox-

ic component in cottonseed. Bottger et al. (1964) showed that gossypol was toxic to cotton aphids, lygus bugs, salt-marsh caterpillars, thurberia weevils, and bollworms. Jenkins et al. (1966) showed that the grape colaspis and leaf beetle preferred feeding on glandless compared to glanded cotton cultivars. Gossypol inhibits the growth and development of many insect pests including the beet armyworm, bollworm, cabbage looper and the salt-marsh caterpillar [2].

Terpenoids that are biosynthetically related to gossypol also occur in the foliage. Besides gossypol, these terpenes include hemigossypolone and heliocides H_1, H_2, H_3 and H_4 (Gray et al., 1976; Stipanovic et al., 1977a; 1977b; 1978a; 1978b). These compounds are also involved in insect resistance. In an artificial diet study, Stipanovic and collaborators established the effective dosage that is required to reduce growth of the tobacco budworm larvae [*Heliothis virescens* (F.)] by 50% (ED_{50}) (Stipanovic et al., 1977a; Williams et al., 1987). Although gossypol is toxic, field studies show that the levels of heliocides and hemigossypolone correlate better with resistance than gossypol [3].

Cotton bollworm (Helicoverpa armigera), the main rodent pests. In particular, the elements of the cotton bollworm moth is a great loss, he is the glory and the loss of knots, dry or completely fall [4].

Zhang Jhin study carried out at the United States that the resistance of cotton bollworm and sucking pests in addition to the gossypol, along with other terpenoid compounds, flavonoids, fatty acids and tannin material is provided [5]. Thus, gossypol does not appear to be the primary source of chemical resistance to *Helicoverpa* in cotton foliage.

Extensive tests on the toxicity of (+)- and (-)-gossypol to insect pests are not available. Chinese scientists report a study with *Helicoverpa armigera* in which larvae were raised on artificial diets containing either (+)- or (-)-gossypol from the 3rd instar through pupation to the moth stage [6].

The larvae raised on the (+)-gossypol diet matured more slowly, and percent survival to the adult was lower. Stipanovic et al. showed that racemic, (+)- and (-)-gossypol were equally effective at reducing days-to-pupation, pupal weights and survival of *Heliothis zea*.

Stipanovic et al. showed that the leaves and stems of *G.hirsutum marie galante* that exhibit high levels of (+)-gossypol in the seed falls within the normal 3:2 range. Thus, regulation of the (+)- to (-)-gossypol ratio in foliage appears to be under separate regulation from that in seed. Therefore, we expect the plants exhibiting a high level of (+)-gossypol in the seed to behave normally with respect to susceptibility to phytophagous insects [7].

More than a decade ago, scientists at the USDA, Cotton Pathology Research Unit, Southern Plains Agricultural Research Center in College Station, Texas began a backcross breeding program to incorporate the high (+)-gossypol trait from moco cotton into agronomically acceptable cotton cultivars. This backcross breeding program shows two dominant genes are responsible for the high percentages of (+)-gossypol in moco cottonseed. However, there is very limited information on how level (+)- and (-)-gossypol correlated with their insect and disease resistance under field conditions. We have now initiated an investigation on how the percentage of (+)-gossypol in seed could influence resistance to insects and diseases. We investigated resistance of some local cotton varieties, USA accessions and hybrids that exhibit different percentages of (+)-gossypol to *Tetranychus urtical* and *Rizactonia solani*. We found that the percent of (+)-gossypol in seeds does not affect to their resistance. [8].

Lusas et al. detected the larvae of bollworms high reproductive potential and eat mainly the nutrition of cotton, shrub, soybeans, sunflower seeds, macaroni, corn and pomegranate crops. In particular, it has been determined that the larvae of the larvae are mainly concentrated on the pelvic egg [9].

In crop breeding, incl. and cotton are widely used various methods of hybridization. Researches of many scientists have revealed that the effectiveness of each method of hybridization to a small extent depends on both the purpose of crossing and the biology of culture.

By joint researches of USDA and Uzbek scientists under PI-480 (UB-ARS-43) and Uzb2-31001-TA-08 projects toward developing of cultivars that exhibit a high ratio of (+)-gossypol in the seed by using of Uzbek cultivars and American lines has shown that:

-it is possible to transfer the high (+)-gossypol seed trait from U.S. cotton accessions into Uzbek cultivars;

-these Uzbek cotton hybrids developed to date have >93% (+)-gossypol. Thus, it is possible to introduce the high (+)-gossypol seed trait into Uzbek cotton lines to provide plants with agronomic traits suitable for growing in Uzbekistan;

-these plants are no more susceptible to insect pests and pathogens than normal cotton cultivars.

Studies of previous years investigated the character of inheritance and variability of diseases resistance to such pathogens as *Verticillium dahliae*

Kleb, *Thelaviopsis bazicola*, *Rhizoctonia solani* and evaluation of developed progenies for resistance to *Helicoverpa armigera (Hubner)* in artificial conditions, the total of gossypol and (+)-gossypol in seeds and other important parameters of various hybrid generations [10].

Based on the above researches, we analyse the some research results regarding to *Helicoverpa armigera* resistance of progenies developed with the participation of ecologically-geographically and genetically distant hybridization in the paper.

Results of the first observations of the first set of hybrids under isolated conditions showed that the $F_4BC_3S_1$-47-8-1-17 x S-6532 with the level of (+) - gossypol 91.4% was not affected by *Helicoverpa armigera* (Table-1).

Among the studied only in the combination $F_4BC_3S_1$-47-8-1-17 x S-6524 with a high level of (+) - gossypol (91.2%), a relatively high affection with *Helicoverpa armigera* was observed. In the remaining cases, there was no significant difference of affection in depending of the level of (+)-gossypol.

The second observation of the damage of plants with a different level of (+)-gossypol in seeds showed resistance of high (+) - gossypol (91.4%) progenies to *Helicoverpa armigera*. It should be noted that all combinations with the level of (+)-gossypol over 91%, in contrast to the hybrids of the progenies with a low level of (+) - gossypol, were less affected (5 and lower percentages).

Results of the 3-observation of the damage by *Helicoverpa armigera* showed that the pest is being moved depending the level of (+)-gossypol. In other words, among the hybrids of this generation with a high level of trait, only in two cases, i.e. in the combinations $F_4BC_3S_1$-47-8-1-17 x S-6524 and $F_4BC_3S_1$-1-6-3-15 x S-6524, the damage were observed at the level 15% and 10%, respectively. The remaining progenies were affected up to 5 %. However, the percentage of affection with *Helicoverpa armigera* increased (except for the combination $F_4BC_3S_1$-47-8-1-17 x S-6530) at hybrids with a relatively low level of (+) - gossypol.

Results of first observation of hybrids F_6 under the conditions of a greenhouse, showed that all combinations with both a high (above 90%) and low (below 70%) level of (+) - gossypol were affected by *Helicoverpa armigera* (Table-2). Among the studied, the hybrid $F_6BC_3S_1$-1-6-3-15 x S-6530 with a relatively high content of (+) - gossypol (90.8%), was less affected (6%).

Results of monitoring of susceptibility to *Helicoverpa armigera* at the first observation showed that the pest is populated depending on the level of (+)-gossypol. In other words, among F_6 hybrids with a high level of (+) -gossypol, only in two cases, i.e. at F_6L-10/04 x BC_3S_1-47-8-1-17 and $F_6BC_3S_1$-1-6-3-15 x S-6524, a high degree of sussceptibility were observed (23% and 21%, respectively). It can be noted that hybrids of this generation with a high level of (+) - gossypol were affected by *Helicoverpa armigera* from 6% ($F_6BC_3S_1$-1-6-3-15 x S-6530) up to 17% ($F_6BC_3S_1$-47-8-1-17 x S-6532).

Thus, it was found that the studied progenies F_6 with high (+)-gossypol were relatively resistant to the cotton bollworm in comparing to progenies with low level (+)-gossypol. These results indicate that the level of (+) - gossypol does not significantly affect on resistance to *Helicoverpa armigera*.

Results of study the resistance to *Helicoverpa armigera* in ecologically geographically distant hybrids showed that both families with a high level (+) - gossypol and low - are affected by the cotton shovel to the same extent. It was found that the genotype of US samples has a significant effect on the manifestation of tolerance in the offspring and does not exclude the possibility of creating genotypes resistant to a cotton bollworm with a high (+) - gossypol level.

Results of studies of another group of hybrids developed between US accessions and Uzbek cultivars with a high level of (+)-gossypol in seeds showed that their affection related to the initial form genotype involved in hybridization. For example, such hybrids with a low level of (+)-gossypol in seeds as F_5Bukhoro-8 x BC_3S_1-1-6-3-15 (62%), F_5Turon x BC_3S_1-1-6-3-15 (67%), F_5Surkhon -14 x BC_3S_1-1-6-3-15 (79%) and $F_5$9871-I x BC_3S_1-1-6-3-15 (75%), developed from a relatively resistant parent form (BC_3S_1-1-6-3-15) with high (+)- gossypol in seeds (93.8%), were affected with *Helicoverpa armigera* in less degree (with respective affection 25%, 50%, 40% and 40%).

Among the hybrids F_5 with a high level of (+) - gossypol in the seeds, only in one case the damage was 50% ($F_5BC_3S_1$-1-6-3-15 x S-6530), and in the remaining cases susceptibility were 55-65%. In contrast to the above, hybrids of this generation with a low level of (+) - gossypol in seeds, differed in relative tolerance to the *Helicoverpas armigera*. For example, susceptibility of F_5S-6532 x BC_3S_1-47-8-1-17 and F_5S-6532 x BC_3S_1-1-6-3-15 - with low (+)-gossypol were 15% and 20%, respectively. The remaining hybrids of this generation with a low level of (+)-gossypol affected by a *Helicoverpa armigera* from 30% up to 45% (Table 3).

Table 1.

Dynamics of affection of cotton hybrids F_4 by *Helicoverpa armigera*

№	Combinations	(+)-gossypol, %	Total gossypol, %	1-Observation				2-Observation				3-Observation			
				Density of population, 10 plants,n			Affected, %	Density of population, 10 plants,n			Affect-ed, %	Density of population, 10 plants,n			Affected, %
				Eggs,n	Cat-erpil-lars,n	Affected buds, n		Eggs,n	Cat-erpil-lars,n	Af-fected buds, n		Eggs,n	Cat-erpil-lars,n	Af-fected buds, n	
1.	$F_4BC_3S_1$-47-8-1-17 x S-6524	75.6	0.87	0.05	0	0	5.0	0.05	0	0.05	5.0	0	0.1	0.05	10.0
2.	$F_4BC_3S_1$-47-8-1-17 x S-6530	71.9	0.46	0.05	0	0	5.0	0.05	0	0.05	5.0	0	0.05	0.1	5.0
3.	$F_4BC_3S_1$-47-8-1-17 x S-6532	75.6	2.03	0.15	0	0.05	15.0	0.05	0	0.1	5.0	0.1	0	0.05	10.0
4.	$F_4BC_3S_1$-1-6-3-15 x S-6524	76.7	1.36	0.05	0	0	5.0	0.05	0.05	0.05	10.0	0.1	0	0.1	10.0
5.	$F_4BC_3S_1$-1-6-3-15 x S-6530	75.2	1.20	0.15	0	0	15.0	0.05	0.05	0.15	10.0	0	0.1	0.2	10.0
6.	$F_4BC_3S_1$-1-6-3-15 x S-6532	78.5	0.56	0.25	0	0.1	25.0	0.05	0.05	0.15	10.0	0	0.1	0.2	10.0
7.	$F_4BC_3S_1$-47-8-1-17 x S-6524	91.2	1.20	0.8	0	0.1	80.0	0.3	0	0.25	30.0	0.05	0.1	0.3	15.0
8.	$F_4BC_3S_1$-47-8-1-17 x S-6530	92.1	1.35	0.05	0	0.1	5.0	0.05	0	0.1	5.0	0.05	0	0.05	5.0
9.	$F_4BC_3S_1$-47-8-1-17 x S-6532	91.4	0.79	0	0	0	0	0	0	0	0	0.05	0	0	5.0
10.	$F_4BC_3S_1$-1-6-3-15 x S-6524	92.4	1.45	0.15	0	0.05	15.0	0.05	0	0.1	5.0	0.05	0.05	0.1	10.0
11.	$F_4BC_3S_1$-1-6-3-15 x S-6530	93.1	1.23	0.15	0	0.15	15.0	0	0.05	0.1	5.0	0.05	0	0.1	5.0
12.	$F_4BC_3S_1$-1-6-3-15 x S-6532	92.3	1.64	0.05	0	0.05	5.0	0.05	0	0.05	5.0	0.05	0	0	5.0

Table 2.

Dynamics of affection of cotton hybrids F_6 with *Helicoverpa armigera*

№	Combinations	(+)-gossypol, %	Total gossypol, %	1-Observation				2- Observation				Average degree of affecting for 2 observation,%
				Density of population, 10 plants,n			Affected, %	Density of population, 10 plants,n			Affected, %	
				Eggs,n	Caterpillars,n	Affected buds, n		Eggs,n	Caterpillars,n	Affected buds, n		
	$F_6BC_3S_1$-47-8-1-17 x S-6524	91,1	0,65	12	1	3	13.0	1	3	9	4.0	8.5
	$F_6BC_3S_1$-47-8-1-17 x S-6530	91,8	1,03	14	2	8	16.0	2	4	15	6.0	11.0
	$F_6BC_3S_1$-47-8-1-17 xS-6532	91,8	1,32	16	1	5	17.0	3	5	11	8.0	12.5
	$F_6BC_3S_1$-1-6-3-15 x S-6524	90,0	1,12	18	3	6	21.0	2	6	13	8.0	14.5
	$F_6BC_3S_1$-1-6-3-15 x S-6530	90,8	1,45	6	0	0	6.0	0	2	6	2.0	4.0
	$F_6BC_3S_1$-1-6-3-15 x S-6532	91,0	0,87	10	0	2	10.0	1	0	4	1.0	5.5
	$F_6BC_3S_1$-47-8-1-17 x S-6524	77,6	0,89	7	0	4	7.0	0	2	0	2.0	4.5
	$F_6BC_3S_1$-47-8-1-17 x S-6530	74,4	1,23	6	2	8	8.0	0	5	13	5.0	6.5
	$F_6BC_3S_1$-47-8-1-17 x S-6532	78,4	0,92	8	0	3	8.0	1	2	9	3.0	5.5
	$F_6BC_3S_1$-1-6-3-15 x S-6524	75,4	0,91	17	4	8	21.0	2	8	15	10.0	15.5
	$F_6BC_3S_1$-1-6-3-15 x S-6530	77,3	1,16	10	1	3	11.0	1	3	9	4.0	7.5
	$F_6BC_3S_1$-1-6-3-15 x S-6532	78,0	0,68	12	2	4	14.0	1	5	8	6.0	10.0
	F_6L-10/04 x BC_3S_1-1-6-3-15	81,0	1,19	20	3	7	23.0	3	6	15	9.0	16.0
	F_6L-10/04 x BC_3S_1-1-6-3-15	70,0	0,98	5	1	2	6.0	0	3	5	3.0	4.5

Table 3.

Resistance hybrids with different levels of (+) - gossypol in seeds to *Helicoverpa armigera* (in Petri dishes)

№	Initial forms and hybrids	(+)- gossypol, %	Affected,%
1	S-6524 (standart)	77.0	55,0
2	S-6530	70.0	45,0
3	S-6532	75.0	55,0
4	L-10/04	61.0	40,0
5	L-16/04	63,0	50,0
6	BC_3S_1-47-8-1-17	93.3	45,0
7	BC_3S_1-1-6-3-15	93.8	35,0
8	F_5Turon x BC_3S_1-47-8-1-17	62,0	75,0
9	F_5Turon x BC_3S_1-1-6-3-15	67,0	50,0
10	F_5Bukhoro-8 x BC_3S_1-47-8-1-17	64,0	65,0
11	F_5Bukhoro -8 x BC_3S_1-1-6-3-15	62,0	25,0
12	F_5Surkhon-14 x BC_3S_1-47-8-1-17	75,0	50,0
13	F_5Surkhon-14 x BC_3S_1-1-6-3-15	79,0	40,0
14	$F_5$9871-I x BC_3S_1-47-8-1-17	60,0	65,0
15	$F_5$9871-I x BC_3S_1-1-6-3-15	75,0	40,0
16	F_5Surkhon-100 x BC_3S_1-1-6-3-15	74,0	35,0
17	F_5S-6532 x BC_3S_1-47-8-1-17	90,9	55,0
18	F_5S-6532 x BC_3S_1-1-6-3-15	92,8	65,0
19	F_5L-10/04x BC_3S_1-47-8-1-17	80,0	60,0
20	F_5L-16/04x BC_3S_1-47-8-1-17	65,0	40,0

Results indicate that another group of hybrids obtained with the participation of US samples with high (+) - gossypol differ in their stability from those with low levels (+) - of state gossypol. Their affection occurred depending on the paternal genotype involved in the hybridization.

Among the hybrids F_6, comparative resistance with a low level of (+) - gossypol and a relatively high affection of buds with a high level of (+)-gossypol (over 90%) are also observed. For example, the damage of buds with a low (+)-gossypol was from 30% ($F_6BC_3S_1$-1-6-3-15 x S-6524) up to 40% ($F_6BC_3S_1$-1-6-3-15 x S-6532 and F_6L-10/04x BC_3S_1-47-8-1-17), and with a high level of (+) - gossypol - from 55% ($F_6BC_3S_1$-47-8-1-17 x S-6530 and $F_6BC_3S_1$-1-6-3-15 x S-6524) up to 70% ($F_6BC_3S_1$-47-8-1-17 x S-6532). The similar dates of damaging of buds with different levels of (+) - gossypol were observed at hybrids up to F_7.

Thus, on the basis of the obtained results of the study of comparative resistance to *Helicoverpa armigera* among hybrids of different generations in petri dishes, it was established that progenies with a different levels of (+) - gossypol, a definite pattern is observed for affection. Genotypes with a low level of (+)- gossypol are affected by *Helicoverpa armigera* to a certain extent less than hybrids with high (+)-gossypol level. Although, the incidence of the initial accession BC_3S_1-1-6-3-15 with a high level of gossypol does not preclude the possibility of developing of resistant genotypes with a high level of (+)-gossypol, which requires additional studies in this direction. Proven high efficiency of the method of ecologo-geographically and genetically distant hybridization in the creation of naturally tolerant to cotton bollworm (*Helicoverpa armigera*) with a complex genetic basis of new recombinants, families and cotton lines with a high level of (+) - gossypol in seeds.

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