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CREATION OF PROMISING BREEDING MATERIAL OF COTTON SPECIES G.BARBADENSE L.

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Annotation.

Diseases cause significant damage to agriculture in Uzbekistan, especially cotton production. The causal agent of the disease is *Fusarium oxysporum f.sp.vasinfestum* (FOV), one of the economically serious soil pathogens causing significant damage to cotton crops worldwide. During the last decades, this pathogen has been detrimentally affecting cotton production in many countries including Uzbekistan. Varieties of diseases of the genus *Fusarium* are known to play a significant role in the damage of cotton plants, from the moment of seedling emergence to the formation and accumulation of raw cotton yield. The paper presents the results of phytopathological studies in the development of a new variety of cotton fine-fiber cotton resistant to *Fusarium* wilt.

As a result of phytopathological studies, the average value of the trait in the standard varieties Surkhan-14 and Surkhan-16 was 2.72 and 2.61 points, respectively. In indicator variety Bukhara-6 the average value of the trait was 4.35 points. From the point of view of breeding it was found that the most stable should be attributed to hybrids F₆[F₁₂ (C-6037 x Termez-42) x F₁₂ (C-6013 x 5904-I)], F₆[F₁₃ (C-6037 x C-6013) x F₁₂ (5904-I x C-6800)], F₁₀ L-54 x L-102, F₉ 9453-I x L-130 as the average value of the trait was from 1.1 to 1.94 points. As for the other hybrids, they showed themselves as average resistant to this pathogen, with the trait value ranging from 2.11 to 3.41 points. Obtained resistant hybrids, due to the fact that the experiments were conducted on natural wilt background, as well as conducted inoculation in laboratory conditions in each generation.

Keywords: *cotton, breeding, hybrid, line, pathogen, trait, selection, resistance genus Fusarium, morphology, fiber, quality*

INTRODUCTION

At present, breeding and introduction of fine-fiber cotton varieties are being carried out in Egypt, USA, Sudan, Tajikistan, Turkmenistan and other countries. Pathogens of the genus *Fusarium* are widespread in nature, living on plant residues, in soil and on dead plant parts [16] and are capable of destroying organic compounds [2, 12]. Most pathogens of the above genus are phytotrophs (plant pathogens). Pathogens of the same species can affect plants from a wide variety of families [15]. In Egypt, where *Fusarium* root rot affecting cotton seedlings (*Gossypium* spp.) is the most damaging disease [14]. Variegated root rot is most severe during the sprouting phase, but can also develop throughout the vegetation of plants [5]. Previous research has found that most Pima-type varieties grown in California are more susceptible to FOV race 4 than varieties - Upland (*G.hirsutum* L.). Evaluation for resistance to FOV race 4 should be carried out throughout breeding [16].

Fusarium oxysporum f.spp. vasinfectum (FOV) is causing significant damage to cotton production in the United States, as evidenced by the isolation of FOV race 4 in California. In 2003, 17 isolates of *F.oxysporum* were isolated from seed imported from Australia to California for forage production [13]. Significant differences in varietal susceptibility to the FOV race 4 pathogen were observed in both Pima and Acala varieties. [14]. Diseases caused by pathogens of the genus *Fusarium* are considered the most damaging in flax and cotton [3, 4, 6, 7, 8].

To counteract the emerging threat of *Fusarium* wilt of cotton in Uzbekistan, the distribution of race/genotype of the pathogen *F.oxysporum f.sp. vasinfectum* (FOV) [11], as well as the pathogenicity of the most frequently occurring FOV races. Several QTL loci contributing to cotton resistance to FOV have been mapped using SSR markers [9, 10].

MATERIALS AND METHODS

The executors of the applied project A-2021-317 in 2021-2023 conducted research both in laboratory and field conditions of the Research Institute of Cotton Breeding, Seed Production and Agrotechnology (RICBSPA).

A significant part of the experiments was conducted in the fields of the production department of RICBSPA and in the breeding greenhouse of the greenhouse complex "Fitotron", and station variety testing was laid in parallel in the conditions of Tashkent and Surkhandarya regions.

Temperature conditions in Tashkent and Surkhandarya regions in the period of 2021-2023 years in the field experiments were favorable, sowing was carried out at the optimum time. Plants developed at constantly rising temperatures, and hot summer and warm autumn allowed to complete harvesting of experimental seed cotton annually by September 25. Agrotechnical measures carried out in the field during the period of field trial establishment related to the experiment are typical for the cultivation zones.

The diseases cause significant damage to agriculture in Uzbekistan, especially cotton production. The pathogen is *Fusarium oxysporum f.sp.vasinfectum* (FOV), one of the economically serious soil pathogens causing significant damage to cotton crops worldwide. During the last decades, this pathogen has been detrimentally affecting cotton production in many countries including Uzbekistan. Disease varieties of the genus *Fusarium* are known to play a significant role in the damage of cotton plants, from the moment of seedling emergence to the formation and accumulation of raw cotton yield.

For breeding, it is important to determine the tolerance to the pathogen FOV in a timely, short time and reliably in laboratory conditions. Because in field conditions, under unfavorable soil and climatic conditions for the pathogen, it may not harm the plant.

During the study, hybrids F₆-F₁₀ of *G.barbadense* L. species, were artificially infected with the pathogen *F.oxysporum f.sp. vasinfectum* among which resistant samples were selected. Samples of the pathogen were collected from cotton fields where Fusarium infestation reached in Bukhara region. Then a monospore isolate was isolated in laboratory conditions.

Samples of infected leaves, stems and roots were used for isolation of pure culture of the pathogen causing Fusarium wilt of cotton. For this purpose, the surface of infected samples of the plant under study was disinfected. Infected plant parts were cut out 0.5-1 cm in size under sterile conditions using a sterile scalpel. After treatment with spirit flame, 10 Petri dishes were placed in a humid chamber and then the Petri dishes were placed in thermostats at 24-25⁰C for growth of the pathogen. Petri dishes were observed from the third day and sowing of fungal mycelium was carried out from the surface of samples with pathogens in a spirit flame, bacterial hooks into tubes with potato-glucose agar, potato-sugar agar and Chapek's agar.

Leaves, stems and roots of cotton samples infected with Fusarium wilt isolated in Bukhara region were placed in a humid chamber where its clear separation from the stem compared to the root part of the plant was observed.

Pure culture of FOV pathogen was diluted in sterile water at the rate of 5*10⁶ CFU/mL and inoculated plants at the rate of one drop (0.05 ml) in the phase of appearance of 2-4 true leaves. At the same time, the root system of young plants of cotton samples was immersed in the FOV suspension for 10 minutes.

The main criterion for determining the level of pathogenicity of isolates was the analysis of the conducted studies of infected and non-infected (control) variants of phytopathogen studies. Artificially evaluated on 1).

infected plants were a 5-point scale (Fig.

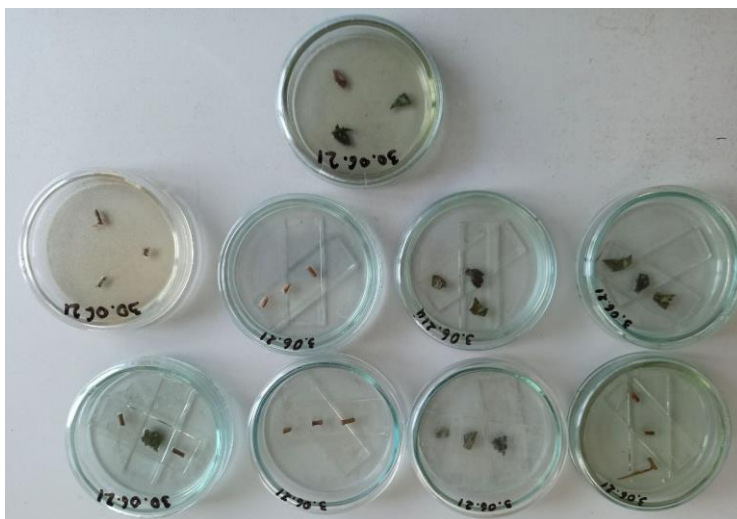


Figure 1. Identification of the pathogen causing Fusarium wilt of cotton from root, leaf and stem parts

In view of the above, we studied the resistance of the presented cotton samples to FOV in laboratory conditions. The studies were conducted in the laboratory "Cotton biotechnology" at RICBSPA. The strains of the fungus causing fusariosis were first grown on potato-agar nutrient medium to create an infectious environment in laboratory conditions. First, 400 g of potatoes were diced. Then boiled for 40 minutes, after a certain time was placed in a beaker and sterilized water

was added up to a volume of 1000 ml. The cooled solution was poured into 500 ml flasks, 10 g agar was added to each flask and placed in an autoclave for sterilization with a stopper. The flasks with potato-agar nutrient medium were then sterilized for one hour at 120°C and 1 atm pressure. The potato agar nutrient medium flasks removed from the autoclave were cooled to 25°C and a certain amount of antibiotic was added to them. After autoclaving, the nutrient media containing antibiotic was poured into 30 Petri dishes in laminar boxes and kept for some time for cooling. The strains of fungi causing Fusarium wilt of cotton were sown in a laminar box. The roots, rhizomes and stems of the plant were cut into pieces, then the cut pieces were placed in small pieces of gauze and sterilized in sodium hypochlorite (17%) in 200 ml beakers and sterilized on sterile water. It was incubated in sodium hypochlorite solution for 1.5 minutes and rinsed in sterile water for 1 minute. The cuttings and sterilized pieces of seedlings were planted on previously prepared nutrient media in Petri dishes. The above biomaterial was grown in Petri dishes in the thermostat for 10 days at a temperature of 24-26°C and humidity of 70-75%, the most favourable for pathogen development.

The isolated fungus *Fusarium oxysporum*, in the form of pure culture, was examined under a microscope (Fig. 2) and, having made sure of it, was used in the preparation of suspension to form an infectious background.

After the formation of 3-4 true leaves on the cotton plant in the root neck area, inoculation with a syringe of the fungus suspension was carried out and a favourable environment for the development of fungal spores was created in the laboratory (Fig. 3). Phenological observations were carried out on 3, 5 and 8 days after inoculation.

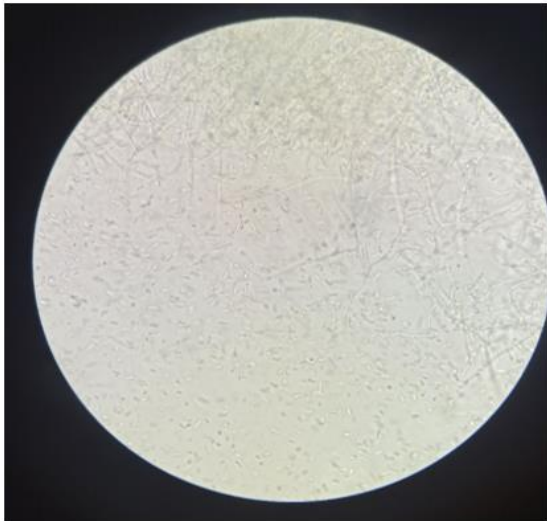


Figure 2: The isolated pathogen *Fusarium oxysporum*, as pure culture, examined under microscope



Figure 3: The inoculation process

The degree of *Fusarium* wilt damage was assessed according to a 5-point system. 0 - no change; 1 - slight colour change (in the form of areas with spots in the longitudinal section of the stem); 2 - extended stem colour change (light colour); 3 - moderate colour change covering more than half of the stem; 4 - manifested in the form of severe darkening of the stem vascular system (Fig. 4).

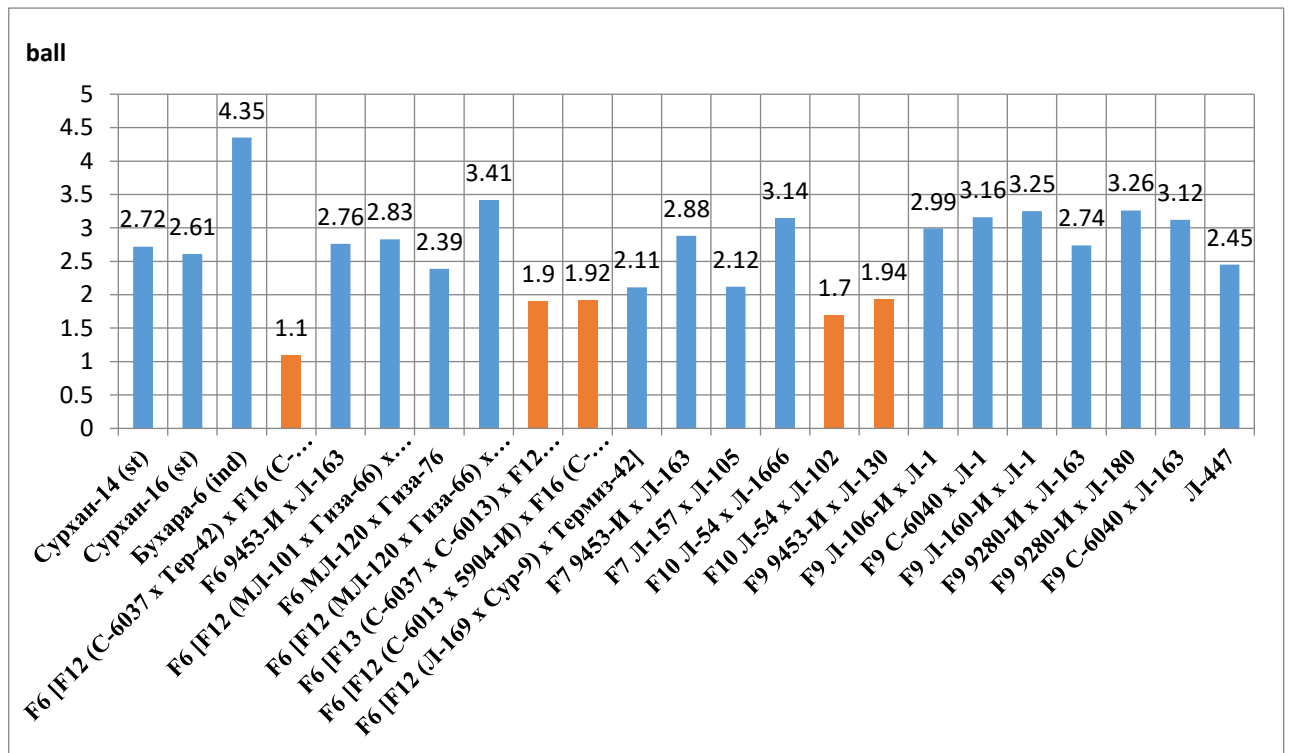
Figure 4. Test for resistance to FOV pathogen isolates

The degree of *Fusarium* wilt lesion was assessed using a 5-point system: 0 - no change; 1 - slight discoloration (in the form of areas with spots in the longitudinal section of the stem); 2 - extended discoloration of the stem (light colour); 3 - moderate discoloration covering more than half of the stem; 4 - manifested as a darker stem colour than in 1 and 2; 5 - the plant died.

According to the results of previously conducted research 2019- 2023 for evaluation in the field in the station variety testing and parallel reproduction in 2023, the following breeding materials are recommended and studied in the conditions of Tashkent and Surkhandarya regions: L-16, L-402 and L-457, along with the standard variety Surkhan-103 and indicator variety 8763-I.

As can be seen from the results of research conducted in 2023 in the conditions of Tashkent region, where the field experiment was laid down according to the methodology of varietal testing in the station nursery with the participation of the above-mentioned selection material, as well as variety-standard Surkhan-103 and indicator variety 8763-I. All lines and standard variety Surkhan-103 of zero branching type and indicator variety 8763-I of sympodial branching type were involved in the experiment.

Analysing the trait "early maturity" we can see that it is equal to 116 days in the variety-standard Surkhan-103, as it can be seen from Table 1 in the variety-indicator 8763-I - 134 days, while in L-16, L-402 and L-457 it is respectively equal to 107, 103 and 106 days.



In terms of raw cotton weight, as it can be seen from Table 1, which is characterised by the following values: in the variety-standard Surkhan-103 is equal to 3.3 g, in the variety-indicator 8763-I - 2.9 g, while in L-16, L-402 and L-457 it is respectively equal to 3.7, 3.6 and 3.6 g.

One of the main attributes determining the quality of fibre is "staple fibre length" in inches, which, as it can be seen from Table 1, is characterised by the following values: in the standard variety Surkhan-103 is 1.39 inches, in the indicator variety 8763-I - 1.41 inches, whereas in L-16, L-402 and L-457 it is 1.43, 1.43 and 1.44 inches, respectively.

Micronaire size is characterised by the following values in Surkhan-103 standard variety equals 4.4 and in 8763-I indicator variety equals 4.2 inches, whereas in L-16, L-402 and L-457 it equals 3.9, 3.7 and 3.6, respectively.

Table 1

Results of testing of selection-significant linear material in the station variety trial in 2023, in conditions of Tashkent region

№	Variety and line name	Early maturity day	Fibre length inch	Micro-neir	Specific breaking strength g.f/tex	Fiber Output		Raw cotton yield as of October 10		Fiber yield	
						%	% to st	c/ha	% to st	c/ha	% to st
1	Surkhan-103st	116	1.39	4.4	37.6	34.1		38.1		13.0	
2	8763-I ind	134	1.41	4.2	38.3	28.4		25.6		7.3	
3	L-16	107	1.43	3.9	40.3	38.4	112.6	42.4	111.3	16.3	125.4
4	L-402	103	1.43	3.7	41.5	38.2	112.0	43.5	111.4 2	16.6	127.7
5	L-457	106	1.44	3.6	42.2	37.9	111.	42.1	110.5	16.0	123.4

No less important feature determining the quality of fibre and as a consequence its price is the feature "specific breaking strength", which is characterised by the following values in the variety-standard Surkhan-103 is equal to 37.6 g.f/tex, in the variety-indicator 8763-I - 38.3 g.f/tex, whereas in L-16, L-402 and L-457 it is respectively equal to 40.3 g.f/tex, 41.5 g.f/tex and 42.2 g.f/tex.

Profitability of fine-fibre cotton varieties along with the quality determines the value of the trait "fibre yield", which is characterized by the following values in the variety-standard Surkhan-103 is equal to 34.1%, in the variety-indicator 8763-I - 28.4%, while in L-16, L-402 and L-457 it is respectively equal to 38.4%, 38.2% and 37.9%.

And the main attributes on which production is oriented is raw cotton yield, and textile industry "fibre yield" on a certain date, which characterises the rate of yield of raw cotton and ultimately the quality of fibre.

Characterizing the total yield of raw cotton on 10 October 2022, that it in the variety-standard Surkhan-103 was - 38.1 c/ha, in the variety-indicator 8763-I - 25.6 c/ha, and in L-16, L-

402 and L-457 it was respectively 42.4 c/ha, 43.5 c/ha, and 42.1 c/ha, or yield increase was in relation to the variety-standard respectively 111.3, 114.2 and 110.5%.

The main feature determining the profitability in Uzbekistan of this or that variety is the yield of fibre on October 10, as it allows to receive fibre of high quality and to free the land for sowing wheat in the optimal time, i.e. contributes to the solution of the problem of food independence of the republic.

At the same time, the fibre yield is composed of the total yield of raw cotton and fibre output.

According to the total fibre yield, it was found that it was 13.0 c/ha in the variety-standard Surhan-103, 7.3 c/ha in the variety-indicator 8763-I, and in L-16, L-402 and L-457 it was 16.3 c/ha, 16.6 c/ha and 16.0 c/ha respectively, or the increase in fibre yield on October 10 was 125.4, 127.7 and 123.4% in relation to the variety-standard respectively.

Based on the analysis of the results of research in the conditions of Tashkent region it should be concluded that L-16, L-402 and L-457 have ultra-fast maturity, high yield and quality of fibre, as well as yield of raw cotton and fibre, and recommended for re-evaluation in the control nursery and breeding in 2022.

For the purpose of reliable evaluation of L-16, L-402 and L-457 in parallel the above-mentioned lines were evaluated in the station test laid in 2022 in the conditions of Surkhandarya region, where the field experiment was laid in accordance with the methodology of small competitive (station) variety trial with the participation of the above-mentioned breeding material, as well as standard variety Surkhan-103 and indicator variety 8763-I.

Analysing the trait "early maturity" it can be seen that it in the variety-standard Surkhan-103, as it can be seen from Table 2 it is equal to 117 days, in the variety-indicator 8763-I - 134 days, while in L-16, L-402 and L-457 it is respectively equal to 106, 105 and 107 days.

In terms of raw cotton weight, as can be seen from Table 2, which is characterised by the following values: in the standard variety Surkhan-103 it is 3.2 g, in the indicator variety 8763-I - 2.8 g, while in L-16, L-402 and L-457 it is respectively 3.7, 3.7 and 3.9 g.

One of the main features determining the quality of fibre is the "staple fibre length" in inches, which, as it can be seen from Table 2, is characterised by the following values: in Surkhan-103 standard variety is 1.39 inches, in 8763-I indicator variety is 1.40 inches, whereas in L16, L-402 and L-457 it is 1.42, 1.43 and 1.44 inches, respectively.

Micronaire size is characterised by the following values in the standard variety Surkhan-103 is 4.2 and in the indicator variety 8763-I is 4.3 inches, whereas in L-16, L-402 and L-457 it is 3.9, 3.7 and 3.6, respectively.

No less important feature determining the quality of fibre and as a consequence its price is the feature "specific breaking length", which is characterised by the following values in the grade-standard Surkhan-103 equals 37.8 g.f/tex, in the variety-indicator 8763-I - 39.9 g.f/tex, whereas in L-16, L-402 and L-457 it is respectively equal to 41.7 g.f/tex, 42.6 g.f/tex and 42.9 g.f/tex (Table 2).

The average value of the trait "fibre yield" is characterized by the following values in the variety-standard Surkhan-103 is equal to 34.1%, in the variety-indicator 8763-I - 30.7%, while in L-16, L-402 and L-457 it is respectively equal to 39.4%, 38.7% and 38.8% (Table 2).

Table 2

Results of testing the best linear material in the station variety trial in 2022, under conditions of Surkhandarya region

№	Variety and line name	Early maturity day	Raw weight of one box gr	Fibre length inch	Micro-neir	Specific breaking strength g.f/tex	Fiber Output		Raw cotton yield as of October 10		Fiber yield	
							%	% to st	c/ha	% to st	c/ha	% to st
1	Surhan-103 st	117	3.2	1.39	4.2	37.8	34.1		38.8		13.2	
2	8763-I ind	134	2.8	1.40	4.3	39.9	30.7		35.1		10.8	
3	L-16	106	3.7	1.42	3.9	41.7	39.4	115.5	49.1	126.5	19.3	146.2
4	L-402	105	3.7	1.43	3.7	42.6	38.7	113.5	46.0	118.6	17.8	134.8
5	L-457	107	3.9	1.44	3.6	42.9	38.8	113.8	48.4	124.7	18.8	142.4

As can be seen from the results of the analysis of the conducted research, which are presented in Table 2, the yield of raw cotton on 20.09.2022 in the variety-standard Surkhan-103 was - 36.9 centners/ha, in indicator variety 8763-I - 32.1 centners/ha, and in L-16, L-402 and L-457 it was 46.1 centners/ha, 44.3 centners/ha, 47.1 centners/ha respectively, or the yield increase was 124.9, 118.6 and 127.6% in relation to the standard variety respectively.

Characterizing the total yield of raw cotton on 10 October 2022, that it in the variety-standard Surkhan-103 was - 38.8 kg / ha, the variety-indicator 8763-I - 35.1 kg/ha, and in L-16, L-402 and L-457 it was respectively 49.1 kg/ha, 46.0 kg/ha, and 48.4 kg/ha or yield increase was in relation to the variety-standard respectively 126.5, 118.6 and 124.7%.

Fibre yield is known to be the product of raw cotton yield and fibre yield. According to the total fibre yield, it was found that it was 13.2 c/ha for the standard variety Surkhan-103, 10.8 c/ha for the indicator variety 8763-I, and 19.3 c/ha, 17.8 c/ha and 18.8 c/ha for L-16, L-402 and L-457, respectively, or the fibre yield increase on 10 October was 146.2, 134.8 and 142.4% in relation to the standard variety, respectively.

CONCLUSIONS.

Based on the analysis of the conducted studies on the value of the trait "resistance to FOV pathogens", the following has been established:

1. As a result of the conducted studies, the average value of the trait in standard varieties Surkhan-14 and Surkhan-16 was 2.72 and 2.61 points, respectively. In indicator variety Bukhara-6 the average value of the trait was 4.35 points. From the point of view of selection it was established that the most stable should be attributed to hybrids $F_6[F_{12} (C-6037 \times \text{Termez-42}) \times F_{12} (C-6013 \times 5904-I)]$, $F_6[F_{13} (C-6037 \times C-6013) \times F_{12} (5904-I \times C-6800)]$, $F_{10} L-54 \times L-102$, F_9

9453-I x L-130 as the average value of the trait was from 1.1 to 1.94 points. As for ostolnye hybrids, they showed themselves as average resistant to this pathogen, with the value of the trait was in the range from 2.11 to 3.41 points. Obtained resistant hybrids, due to the fact that the experiments were conducted on natural wilt background, as well as conducted inoculation in laboratory conditions in each generation.

2. In the conditions of Tashkent and Surkhandarya regions L-16, L-402 and L-457 have ultrashort maturity, high yield and quality of fibre, as well as yield of raw cotton and fibre, and recommended for re-evaluation in the competitive variety trial and further multiplication of the above lines in 2024 in the greenhouse complex "Fitotron".

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