

**1419 Wilt Resistance of New Varieties and Lines of Cotton (*Gossypium hirsutum* L.) to
Different Geographic Populations of the Pathogen *Verticillium dahliae* Kleb. in
Uzbekistan**

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Abstract

Herein we consider the resistance of the new Uzbek wilt resistant varieties and lines of cotton (*Gossypium hirsutum* L.) towards different geographic populations of the fungal pathogen *Verticillium dahliae*, that were isolated from severely diseased plants found in widely cultivated varieties of cotton in various soils and climatic regions of the Republic of Uzbekistan. During first fruit set, plants were artificially inoculated in the field at the beginning of July. Plants were inoculated near the root collar using a medical syringe with a pathogen conidial suspension - 1×10^6 propagules/ml. Ten plants, of each line or variety were inoculated. The virulence of the isolates was assessed according to the number of plants showing wilt, and according to the intensity of disease as demonstrated by discoloration of the interior of the stalk. Many of the varieties and lines studied did not show a high degree of resistance to the new virulent isolates of the pathogen. Exceptions were the variety Bukhara-8 and lines LS-6593 and LS-6595, which showed hyper-sensitivity to the virulent isolates 58, 53 and 52, and thus appear to be a valuable source of germplasm for breeding. Lines L-155, L-408, L-842, L-866, L-1708, L-387, and also the varieties Omad, C-8284 and C-8288 showed some resistance to virulent *V. dahliae* isolates 28, 30, 32, 36, 40 and 44. These lines and varieties also appear to have potential as resistant germplasm. Therefore, it will be beneficial to introduce these resistant germplasm stocks into the breeding process to create new more wilt resistant varieties of cotton. These data will also aid in providing preliminary recommendations to districts that cultivate cotton taking into account the specific virulent isolates of *V. dahliae* that were collected from that district.

Wilt Resistance of New Varieties and Lines of Cotton of the Species *G. hirsutum* L. after Inoculation with Different Geographic Populations of the Fungus *Verticillium dahliae* Kleb in Uzbekistan. Marupov A., Kim R.G., 2007

Introduction

In the present integrated system to combat *Verticillium* wilt of cotton, breeding for wilt resistant varieties of cotton is of primary importance. Academician S.M. Mirakhmedov has elaborated a fundamentally new approach to accomplish this goal. He crossed the wilt resistant wild subspecies *Gossypium hirsutum* L *ssp. mexicanum* (Tod). Mauer, V. Nervosum (W) with the early maturing variety C-4727, and with the following backcrosses created the wilt resistant varieties Tashkent-1, Tashkent-2, Tashkent-3 and Tashkent-6. These possess high productivity and fiber quality. However, because of evolving pathogen-host plant interactions, resistance to the new more virulent isolates of *Verticillium dahliae* is no longer observed.

The decline in resistance has prompted this study of *V. dahliae* and the virulence of its various races and populations to different varieties and lines of *G. hirsutum* and how other environmental factors affect performance.

Materials and Methods

Isolation of monospore isolates of *V. dahliae* Kleb. – Pathogen isolates were originally collected from diseased plants in autumn by placing infected plant stalks in a moist chamber. The stalks (5 mm in length) were placed in Petri dishes and then into a thermostat with humidity of 60-70% and a temperature of 24-26C° within 3 -5 days. The fungus was then transferred to glass test-tubes containing Czapek's agar medium. Alternatively, the cut stalks from the diseased plants were placed directly on Czapek's medium in test-tubes, omitting the moist chamber (Khokhryakov, 1964). In the present investigation, we studied new isolates of Wilt Resistance of New Varieties and Lines of Cotton of the Species *G. hirsutum* L. after Inoculation with Different Geographic Populations of the Fungus *Verticillium dahliae* Kleb in Uzbekistan. Marupov A., Kim R.G., 2007

V. dahliae from different geographical origins in the Republic of Uzbekistan, that were isolated from different varieties of field cotton in - seven Provinces (see Table 1).

Plant inoculation - Ten plants of each line or variety were inoculated in the field with each pathogen isolate by use of a medical syringe. The suspension of *V. dahliae* was diluted to a concentration of 1×10^6 conidia ml^{-1} according the methodology of Yakutkin (1973), and Marupov (2003). Suspensions of 7-9 days-old monospore isolates were prepared according to the method of Tarunina (1973). A Goryayev cell was used to calculate the spore concentrations. Inoculations were begun in early July during flower bud development by injecting 0.1 ml of pathogen suspension into the plant near the root collar. Plants were labeled, and subsequent data was recorded at the first appearance of disease symptoms, the number of diseased plants showing disease symptoms, and the intensity of disease symptomology. The virulence of isolates was assessed according to the number of plants showing wilt symptoms, the intensity of leaf wilting and necrosis, and also the duration of the incubation period. The intensity of leaf wilting was defined according the scale of Thamos and Korharos (1978). The degree of wilting during the period of vegetation was measured by visually inspecting the plant and recording the results on a 1 to 4 scale (resistant, faint, medium and severe wilt), and in at the end of vegetative growth after harvest the stalks were cut and the degree of necrosis was observed and rated on a 1 to 4 scale [healthy plant (no necrotic spots); faint (insignificant distribution of necrotic spots on the whole of cut of stalk); medium (distribution of necrotic spots over the whole of the cut of stalk and severe (complete necrosis or dead plant)].

Results and Discussions

The results of these studies on resistance show that the varieties and lines of cotton differ in the genetic nature of their resistance to wilt incited by new virulent populations of

the fungal pathogen *V. dahliae*, that is they possess a race specific resistance characterized by hypersensitivity to specific pathogen isolates, and non-race specific or horizontal (field) resistance that can be characterized as tolerance. Specific examples include the early maturing dwarf line L-155 which exhibits resistance to (isolates: 3, 10, 15, 17 and 20), and shows a high degree of wilt resistance and relatively high degree of tolerance to the pathogen (i.e. it possesses horizontal or non-race specific resistance). Relatively high wilt resistance was observed with the varieties Omad, C-8284, C-8288, and also with the lines L-408, L-842, L-866, L-1708 and 387 towards several isolates of *V. dahliae*. The variety C-9082 exhibited high resistance towards isolates 3 and 10. The varieties C-6524, C-5621, Tashkent-6, Namangan-77 and the lines L-275, L-70, L-052 were highly susceptible to the majority of the studied isolates of *V. dahliae*.

Wilt resistance of the varieties and lines of cotton towards new more virulent isolates 52, 53, 54, 55 and 58, which were isolated in widely cultivated varieties of cotton from different geographic regions of the Republic of Uzbekistan, was characterized by as shown by both the phenotypic disease response, and the wilting and cut stalk assay. After inoculation, 30 to 60 percent of the plants showed disease symptoms. The difference between phenotype assessment and assessment by observing the cut stalk, in general, was insignificant and varied from 0 to 10%, and only in five cases up to 20%. The highest wilt resistance was observed in the variety Bukhara-8 towards virulent isolate 58. This isolate was collected from the variety Omad in Tashkent province. In the case of Bukhara-8, wilted plants were not observed during the whole vegetative period as judged by both phenotypic symptoms, as well as by the cut stalk. Thus, Bukhara-8 appears to be super sensitive to isolate 58.

A similar lack of symptoms was observed with the line LS-6593 to isolate 53, this isolate was collected from the variety An-Bayaut-2 (Syrdarya Province), and also with the

line LS-6595 to isolate-55, that was collected from the variety Akdarya-6 (Samarkand province).

Relatively high resistance to new virulent isolates of the fungus *V. dahliae* were observed for line LS-6593 to isolate 52 that was collected from the variety C-4727 (Karakalpak SRIoCP, Tashkent Providence), and also LS- 6595 to isolate 58 collected from the variety Omad (Tashkent province). About 10% of these plants showed phenotypic wilt after inoculation. A similar number of diseased plants were observed when assayed by the cut stalk assay. Thus, these lines do not exhibit hypersensitivity to virulent isolates 52 and 58.

Most of the studied varieties and lines possess relatively little resistance to new virulent populations of the fungus *V. dahliae*. Therefore, Verticillium wilt remains a serious problem, especially in view of the appearance of new more aggressive strains of *V. dahliae*; with up to 100% of existing varieties being negatively affected.

These results show that the studied varieties and lines have a relatively low degree of resistance towards new virulent isolates of *V. dahliae*. Exceptions are the variety Bukhara-8, which exhibits hypersensitivity towards virulent isolate 58, and lines LS-6593 and LS-6595 which also exhibits hypersensitivity towards virulent isolates 53 and 52. These varieties and lines represent superior as breeding stock. Lines L-155, L-408, L-842, L-866, L-1708, L-387, and also the varieties Omad, C-8284 and C-8288 exhibit complex wilt resistance to the studied isolates 28, 30, 32, 36, 40 and 44 of *V. dahliae*; these also may provide superior breeding stock but only to these less aggressive *V. dahliae* isolates. Therefore, it may be advantageous to involve them in a breeding program to create new more wilt resistant varieties of cotton perhaps by combining them with the new early maturing, high-yielding varieties C-8286 and C-8290 which have fiber quality corresponding to international standards. This may provide complex resistance to more virulent populations of the fungus *V.*

dahliae such as those that have developed in the more than 50 thousand ha currently planted in the Republic of Uzbekistan.

Based on these results it is possible to make preliminary recommendations for planting specific cotton varieties for cultivation depending upon the range of distribution of these isolates of *V. dahliae*.

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Table 1. Collection site for isolates of *Verticillium dahliae*.

Isolate #	Virulence	Variety ¹	District	Province
28	Mild	C-6524	Shakhrikhan	Andijan
30	Mild	C-6524	Asaka	Andijan
32	Mild	C-2609	Andijan	Andijan
36	Mild	Akkurgan-2	Uyichirchik	Namangan
40	Mild	C-6524	Uyichirchik	Namangan
44	Mild	Akkurgan-2	Bagdad	Fergana
52	Severe	C-4727	Karakalpak	Tashkent
53	Severe	An-Bayau-2t	Syrdarya	Syrdarya
54	Severe	An-Bayaut-2	Dzizak	Dzizak
55	Severe	Ak-Darya-6	Samarkand	Samarkand
58	Severe	Omad	Tashkent	Tashkent

¹Cotton variety from which the pathogen was collected.