Assessment of Resistance in Potato Cultivars to Verticillium Wilt Caused by *Verticillium dahliae* and *Verticillium nonalfalfae*

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Abstract

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Verticillium wilt caused by *Verticillium* spp., also called potato early dying disease, is one of the most serious soilborne diseases affecting potato production in China. The disease has been expanding into most potato production areas over the past few years. Information on host resistance against Verticillium wilt among the potato cultivars in China is scarce, but it is critical for sustainable management of the disease. This study, therefore, evaluated 30 commercially popular potato cultivars against *Verticillium dahliae* strain Vdp83 and *Verticillium nonalfalfae* strain Vnp24, two well-characterized strains causing Verticillium wilt of potato in China. Both strains were isolated from diseased potato plants, and they were previously proven to be highly virulent. Ten plants of each cultivar were inoculated with the *V. dahliae* strain and incubated on greenhouse benches. Symptoms were rated at weekly intervals, and the relative area under the disease progress curve was calculated. The experiment was repeated once, and nonparametric analysis was used to calculate the relative marginal effects and the corresponding confidence intervals. Five resistant cultivars and four susceptible cultivars identified from the analyses were then challenged with the *V. nonalfalfae* strain. Cultivar responses to *V. nonalfalfae* were like those exhibited against *V. dahliae*, except for one cultivar. This study showed that resistance among potato cultivars exists in China against *Verticillium* spp. and that the resistance to *V. dahliae* identified in potato is also effective against the other *Verticillium* species that cause Verticillium wilt with a few exceptions. Cultivars identified as resistant to Verticillium wilt can be deployed to manage the disease until the breeding programs develop new cultivars with resistance from the sources identified in this study.

Potato (*Solanum tuberosum* L.) is an important crop globally, and China is the largest producer of potato, accounting for one-quarter of the world's production (Li et al. 2016). Since 2015, potato has become the fourth largest staple crop in China after rice, wheat, and maize. Verticillium wilt of potato, caused by *Verticillium* spp., is progressively becoming a serious problem in Shaanxi, Gansu, Xinjiang, and Guizhou Provinces within China (Chen et al. 2013; Liu et al. 1992; Wang et al. 2014; Zhang 2004), and it may result in up to 50% yield losses reported elsewhere (Dung et al. 2012).

The species within the genus *Verticillium* that causes extensive losses in potato is *Verticillium dahliae* (Johnson and Cummings 2015; Johnson and Dung 2010), and this species also threatens many dicotyledons, such as cotton, lettuce, tomato, strawberry, pepper, and sunflower (Inderbitzin and Subbarao 2014; Klosterman et al. 2009). The importance of *Verticillium nonalfalfae*, a relatively new species within the genus *Verticillium*, is becoming apparent, with reports of Verticillium wilt caused by this species on other crops, such as hops, spinach, solanaceous crops (including potato), and trees (Inderbitzin et al. 2011; Jing et al. 2018; Kasson et al. 2014; Schall and Davis 2009).

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Both *V. dahliae* and *V. nonalfalfae* form resting structures, like microsclerotia and resting mycelia, respectively (Inderbitzin et al. 2011), enabling them to survive unfavorable conditions (Bhat and Subbarao 1999; Inderbitzin et al. 2011; Klosterman et al. 2009). Microsclerotia survive for at least 10 years (Wilhelm 1955), and the survival duration of other resting structures formed by the species of the genus *Verticillium* are unknown. Fumigation with methyl bromide (MeBr) and chloropicrin was developed as an effective management technique against *V. dahliae*. However, agricultural fumigation with MeBr is no longer available as of 2015. Furthermore, the use of these broad-spectrum fumigants causes disproportionate negative impacts on the beneficial soil macro- and microbiota (Gurung et al. 2015; Klosterman et al. 2009). Many microorganisms have been reported to be able to suppress *Verticillium* spp., but additional commercial development and evaluation are needed before they can be deployed in commercial agriculture (Derksen et al. 2013).

Crop rotation is an efficient way to reduce pathogen propagules and manage many plant diseases. However, owing to the wide host range and the long-term persistence of *Verticillium* spp. in soils, rotation may not be economically feasible in many regions (Dung et al. 2013). The only exception is that the rotation of susceptible crops with broccoli has been proven to be effective in reducing soilborne propagules and disease incidence in several cropping systems (Hao et al. 2003; Njoroge et al. 2009; Subbarao and Hubbard 1996; Subbarao et al. 2007; Xiao et al. 1998). More research (Inderbitzin et al. 2018) has shown that the incorporation of broccoli alters the soil microbiomes from disease conducive to disease suppressive, and this is deemed to be the mechanism by which broccoli reduces soilborne propagules.

Host resistance remains the most effective long-term management method, because other control methods are cost prohibitive, potentially damaging to the environment, or of limited feasibility (Hayes et al. 2007). *V. dahliae* exists as two pathogenic races (race 1 and race 2) in lettuce. Race 1 and race 2 were also described in tomato (Alexander 1962; Vallad et al. 2006), and the pathogenicity of isolates (race 1 or race 2) from lettuce and tomato is strongly correlated (Maruthachalam et al. 2010). In tomato, widespread use of cultivars carrying the race 1 resistance gene *Ve+* led to the outbreak of race 2 isolates that predominate many tomato production regions (Alexander 1962; Pegg and Brady 2002). The existence of a similar race structure of *V. dahliae* populations in potato is also known, and thus, resistant cultivars offer the most effective method of managing this disease (Jansky 2009; Jansky and Miller 2010; Jansky and Rouse 2003; Simko and Haynes 2016). Many popular potato cultivars are still susceptible to Verticillium wilt (Arbogast et al. 1999; Frost et al. 2006; Jansky 2009; Jansky and Miller 2010), despite significant efforts to breed for resistance. Resistance against Verticillium wilt from diploid *Solanum* species could be transferred to tetraploid potato, and diploid *Solanum* species could be a good source of resistance. Thus, a number of *Solanum* clones resistant to Verticillium wilt, such as LRC18-21, LRC373-5, C287, and C545, have been released (Concibido et al. 1994; Frost et al. 2006; Jansky and Rouse 2000, 2003; Lynch et al. 2004).

There is limited information on the resistance response of major commercial potato cultivars to Verticillium wilt in China. Given that the disease has become a major production constraint on potato, information on the status of Verticillium wilt resistance in contemporary commercial cultivars is critical to managing the disease in production fields. The objectives of this study were to evaluate the major commercial potato cultivars from China for resistance against *V. dahliae* and test if responses identified to *V. dahliae* also confer resistance against *V. nonalfalfae*. This information is critical not only to inform cultivar deployment but also, for the breeding programs.

Materials and Methods

Potato cultivars and plant growth conditions. A total of 30 potato cultivars were collected from seed companies and research institutions in China (Table 1). These cultivars are uniquely suited to the different production regions in China, and they are regularly planted by growers. Potato tubers from each cultivar were cut into several suitable-sized pieces (3 × 3 × 3 cm), each with at least one bud eye. A single piece was sown into each plastic pot (upper diameter of 13.5 cm, bottom diameter of 8.9 cm and height of 10.5 cm) filled with a mixture of sterile nursery substrate, sterile sand, and sterile soil (1:2:4 by volume).

Pathogen inoculation. Two fungal strains *V. dahliae* strain Vdp83 and *V. nonalfalfae* strain Vnp24, both isolated from Verticillium wilt-infected potatoes, were cultured on potato dextrose agar (PDA) for 14 days. Then, distilled water was added to the PDA plates to wash off the spores with the help of a glass spreader. The concentration of the spore suspension was adjusted to 2×10^6 conidia ml^{-1} before inoculation (Tai et al. 2013). Potato seedlings were inoculated at the approximately four- to six-leaves stage (~4 to 5 weeks after sowing) following the method published by Tai et al. (2013). A

Table 1. The cultivars, their source, median disease rating (MDR), and relative area under the disease progress curve (RAUDPC) obtained from two independent inoculation experiments, each with the *Verticillium dahliae* isolate Vdp83 and the *Verticillium nonalfalfae* isolate Vnp24

		Vdp83		Vnp24	
Cultivars	**Breeding institution**	**MDR[a]**	**RAUDPC**	**MDR[a]**	**RAUDPC**
Helan 15	—	3	2,589.20		
Longshu 6	Gansu Academy of Agricultural Sciences	3	2,327.35	2	567.06
Zaodabai	Liaoning Benxi Potato Research Institution	3	2,314.46		
Longshu 3	Gansu Academy of Agricultural Sciences	3	1,528.98		
L0527-4	Gansu Academy of Agricultural Sciences	3	1,961.09		
Longshu 9	Gansu Academy of Agricultural Sciences	2.5	1,845.38		
Caishu	—	3	1,743.28		
L0529-2	Gansu Academy of Agricultural Sciences	2	1,766.08		
Zhuangshu 3	Zhuanglang County Agricultural Technology Extension Center	2	1,901.30		
Tianshu 12	Tianshui Institute of Agricultural Sciences	1	1,580.57		
Xindaping	Gansu Anding District of Dingxi City Agricultural Technology Center	1.5	1,858.07	2	522.54
Longshu 10	Gansu Academy of Agricultural Sciences	2	1,669.30		
Atlantic	United States Department of Agriculture	2	1,310.55	2	413.46
Longshu 12	Gansu Academy of Agricultural Sciences	1	1,792.23		
Longshu 7	Gansu Academy of Agricultural Sciences	1.5	1,515.85	2	483.85
Jinshu 16	High Latitude Crops Institute, Shanxi Academy of Agriculture Sciences	1	1,263.51		
L0109-4	—	1	1,539.56		
Huangyangyu	—	1	1,286.76		
Jizhangshu 14	Zhangjiakou Academy of Agricultural Sciences	1	1,565.84		
Zhongshu 19	Institute of Vegetables and Flowers, Chinese Academy of Agricultural Sciences	0	956.23	0	288.04
Longshu 11	Gansu Academy of Agricultural Sciences	0	1,104.15	1	407.74
Longshu 13	Gansu Academy of Agricultural Sciences	0	1,051.44		
Zhongshu 20	Institute of Vegetables and Flowers, Chinese Academy of Agricultural Sciences	0	973.48		
L0227-18	—	0	1,073.84		
Hongyun	Hunan Agricultural University and Hunan Province Potato Engineering Technology Research Center	0	897.22		
Z2011-1	—	0	950.87		
Qingshu 9	Biotechnology Research Center of Qinghai Academy of Agriculture and Forestry	0	817.50	0	172.50
Longshu 8	Gansu Academy of Agricultural Sciences	0	824.00	0	179.06
Zhongshu 18	Institute of Vegetables and Flowers, Chinese Academy of Agricultural Sciences	0	817.50	0	174.69
Zhongshu 21	Institute of Vegetables and Flowers, Chinese Academy of Agricultural Sciences	0	817.50		

[a] MDR was calculated from the final disease evaluation.

sterile scalpel was used to make three incisions down the root, and 40 ml spore suspension was then poured into the wounded root zone. Seedlings were inoculated again 1 week after the first inoculation. Seedlings in the control treatments were similarly treated with distilled water. Ten plants serving as one replication of each cultivar were inoculated with *V. dahliae* and arranged in a randomized block design. The experiment was conducted in a greenhouse with the photoperiod set to ~14 h, and the temperature was set to 22 to 25°C (day) and 11 to 14°C (night). The seedlings were manually watered every 2 days, and no other management measures (e.g., pest or disease control or fertilization) were used. The experiment was repeated once.

Resistance screening was carried out in two stages. In the first stage, seedlings of all 30 cultivars were inoculated with Vdp83 to evaluate their responses to *V. dahliae*. Cultivars Longshu 3 and Atlantic, which were previously confirmed as susceptible (Chen et al. 2013; Nair et al. 2016; Whitworth and Davidson 2008), and cultivar Qingshu 9, which was previously confirmed as resistant (Cai and Lu 2017), were included in this experiment as checks.

The second-stage screening was to test if cultivar responses against *V. dahliae* were similar in *V. nonalfalfae*. To test this, *V. nonalfalfae* strain Vnp24 was inoculated on 9 of the 30 cultivars screened against *V. dahliae*. The nine cultivars were selected based on varying levels of resistance exhibited against *V. dahliae*. Five of the nine cultivars had few or no symptoms, and the other four cultivars exhibited high susceptibility to *V. dahliae*. Among the five resistant cultivars, three had 0% incidence against *V. dahlia*, and the remaining two had ≤25% incidence. Among the four susceptible cultivars, one had 100% Verticillium wilt, and the other three had >50% incidence. This experiment was also conducted two times with 10 plants per cultivar inoculated during each experiment.

Disease assessment and data analysis. Disease severity caused by *V. dahliae* and *V. nonalfalfae* was assessed weekly for 4 weeks after inoculation for a total of five assessments with *V. dahliae* and four assessments with *V. nonalfalfae*. Diseased plants were scored on a rating scale designed for Verticillium wilt in potatoes (Rowe 1985): 0 = no visible symptoms; 1 = some chlorosis, especially in older leaves; 2 = general chlorosis and some necrosis and wilting; and 3 = severe wilting or death. For each week of assessment, all disease-rating data from each experiment of each cultivar over time were collected and ranked, and mean ranks of each experiment of each cultivar at different rating times were calculated. The area under the disease progress curve (AUDPC) was then calculated as $\Sigma_1^{n-1}\left[\left(\frac{yi+yi+1}{2}\right)\times(ti+1-ti)\right]$, where n is the total number of assessments, yi is mean rank at ith assessment, and $ti+1-ti$ is the interval (days) between two consecutive assessments (Derksen et al. 2013). AUDPC values were then normalized as relative area under the disease progress curve (RAUDPC) = AUDPC/($t_n - t_1$), where t_1 and t_n are the dates of the first and last assessments, respectively (Jansky 2009).

Nonparametric data analyses (Shah and Madden 2004) were performed to compare cultivar resistance or susceptibility (i.e., RAUDPC) using SAS (version 8.1). Relative marginal effects (RMEs) and 95% confidence intervals of RMEs were generated with the PROC MIXED procedure and the LD_CI (confidence intervals for the relative treatment effects of longitudinal data) macro (Hu et al. 2015; Shah and Madden 2004). RME values with nonoverlapping confidence intervals were considered significantly different. A cultivar with a higher RME value tends to be more susceptible to the pathogen. The data from cultivar responses to *V. nonalfalfae* were also analyzed using the same procedure.

Results

Resistance assessment of potato cultivars to *V. dahliae*. Based on the five disease assessments in each experiment, cultivars Zhongshu 18, Zhongshu 21, Qingshu 9, Longshu 8, Hongyun, Zhongshu 19, Z2011-1, Zhongshu 20, Longshu 13, L0227-18, and Longshu 11 developed little or no symptoms of Verticillium wilt, and the median disease rating on these cultivars was 0 (median disease rating values were calculated on the data from the last date of disease assessment). Within this group, Zhongshu 18, Zhongshu 21, Qingshu 9, Longshu 8, and Hongyun did not show the foliar symptoms of Verticillium wilt or the vascular discoloration (Fig. 1). The corresponding RAUDPC values on these cultivars were 817.50, 817.50, 817.50, 824.00, and 897.22, respectively (Table 1). The remaining cultivars developed severe symptoms, especially cultivars Helan 15, Longshu

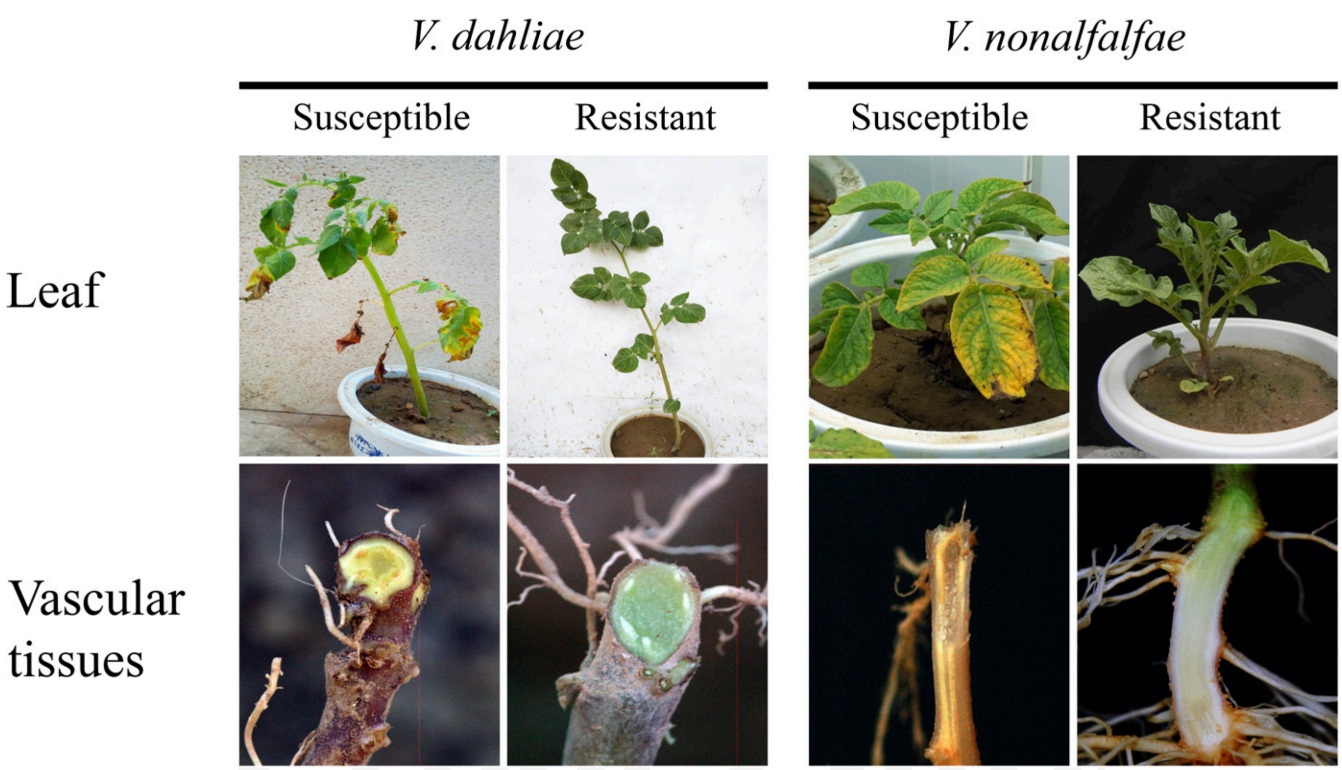

Fig. 1. Symptoms caused by *Verticillium dahliae* or *Verticillium nonalfalfae* in susceptible and resistant potato cultivars.

6, and Zaodabai, with their median disease rating being 3 (Fig. 1). Nearly all plants of these cultivars were dead by the end of the experiment in which they were inoculated with the *V. dahliae* strain. The corresponding RAUDPC values on these cultivars were 2589.20, 2327.35, and 2314.46, respectively.

Statistical analysis indicated significant differences in the response of cultivars ($P < 0.01$) (Table 2), but experiments ($P > 0.87$) or cultivar-experiment interactions ($P = 1.0$) did not influence cultivar responses to Verticillium wilt. The RME values of cultivar responses and the corresponding 95% confidence intervals are given in Table 3. Zhongshu 18, Zhongshu 21, Qingshu 9, Longshu 8, and Hongyun did not exhibit wilt symptoms, and their RME values were 0.06, 0.06, 0.06, 0.09, and 0.15, respectively (Table 3). The RME values of these five cultivars were significantly lower than those of the remaining 25 cultivars. Other than these five cultivars, another six cultivars (Longshu 11, L0227-18, Longshu 13, Zhongshu 20, Z2011-1, and Zhongshu 19) showed only slight symptoms. The RME values of these cultivars were significantly lower than those the remaining cultivars, except cultivars Huangyangyu and Jinshu 16 ($P < 0.05$). Cultivars from Helan 15 to Atlantic in Table 3 showed typical symptoms of Verticillium wilt; some cultivars, such as Helan 15, Longshu 6, and Zaodabai, were highly susceptible to Vdp83, with RME values of 0.98, 0.94, and 0.93, respectively (Table 3). The RME values of these three cultivars were significantly higher than on other cultivars ($P < 0.05$).

Resistance assessment of potato cultivars to *V. nonalfalfae*. Disease development caused by *V. nonalfalfae* was assessed four times. In our investigation, Qingshu 9, Zhongshu 18, Longshu 8, and Zhongshu 19 developed incipient Verticillium wilt with median disease ratings of 0 (Fig. 1), and the corresponding RAUDPC values were 172.50, 174.69, 179.06, and 288.04, respectively (Table 1). In contrast, Longshu 6 as well as cultivars Xindaping and Longshu 7 showed typical symptoms of Verticillium wilt with median disease ratings of 2 (Fig. 1), and the corresponding RAUDPC values were 567.06, 522.54, and 483.85, respectively. Atlantic and Longshu 11 showed general symptoms with median disease ratings of 2 and 1, respectively, and the corresponding RAUDPC values were 413.46 and 407.74, respectively.

Statistical analysis indicated significant differences in the response of cultivars ($P < 0.01$) (Table 4) but no significant effects of experiments or cultivar × experiment interaction. The RME values of cultivar responses and the corresponding 95% confidence intervals are given in Table 5. Among the nine cultivars, Qingshu 9, Zhongshu 18, Longshu 8, and Zhongshu 19 had RME values of 0.11, 0.18, 0.21, and 0.39, respectively, and the RME values of these cultivars were significantly lower than those of the other cultivars. Longshu 11 and Atlantic showed general symptoms with RME values of 0.53 and 0.58, respectively; Xindaping, Longshu 6, and Longshu 7 were highly susceptible to Vnp24 with high RME values of 0.89, 0.86, and 0.75, respectively.

Discussion

Soil fumigation and use of resistant cultivars are two widely used methods to control Verticillium wilt in the different cropping systems in which the disease is prevalent. The combination of MeBr and chloropicrin, two effective soil fumigants against Verticillium wilt (Wilhelm et al. 1961), has been phased out, or their use is restricted to quarantine purposes in China based on the Montreal Protocol. The least expensive way of managing Verticillium wilt is to plant resistant cultivars (Jansky and Rouse 2003). Of the 30 potato cultivars tested, 11 cultivars were identified as resistant to *V. dahliae*; five of these cultivars (Zhongshu 18, Zhongshu 21, Qingshu 9, Longshu 8, and Hongyun) showed virtually no visual symptoms of Verticillium wilt either on the leaves or in vascular tissues. These five cultivars are good candidates for use in those regions where Verticillium wilt is an issue subject to their agronomic performance. Huangyangyu and Jinshu 16 showed slight Verticillium wilt symptoms with median disease ratings of 1; they showed no significant differences from cultivars, such as Atlantic, with typical Verticillium wilt symptoms or several cultivars showing only slight symptoms, such as Longshu 11. These two cultivars could not be defined for their response levels to Vdp83 with the disease rating scale used in this research (0 to 3); if a wider and more precise range scale was used, such as a scale of 1 to 10 (Goth and Haynes 2000) or a scale of 0 to 5 (Derksen et al. 2013), the resistance levels of such cultivars could be more precisely defined. Such knowledge will determine the importance of these cultivars to resistance breeding programs in potato.

In our previous study, other than *V. dahliae*, many *V. nonalfalfae* isolates were obtained from Verticillium wilt-infected potato plants, and they were subsequently confirmed to be pathogenic on potato (Jing et al. 2018). Several cultivars resistant to *V. dahliae* strain Vdp83 (Zhongshu 18, Qingshu 9, Longshu 8, and Zhongshu 19) were also resistant to *V. nonalfalfae* strain Vnp24. None of these cultivars showed any symptoms of Verticillium wilt when challenged with either pathogen. In contrast, Longshu 11 was more susceptible to *V. nonalfalfae* than to *V. dahliae*, with a median disease rating of 0 against the *V. dahliae* isolate Vdp83 and a median disease rating of 1 against the *V. nonalfalfae* isolate Vnp24. Xindaping, Longshu 6, Longshu 7, and Atlantic were susceptible to both pathogens and showed typical symptoms. Jing et al. (2018) had compared the pathogenicity between *V. dahliae* and *V. nonalfalfae* on potato and indicated that *V. nonalfalfae* exhibited lower virulence. This phenomenon was not obvious in this study, because the isolates of *V. nonalfalfae* used in the two studies were different.

The results presented in this study suggest that the responses of nearly all cultivars against *V. dahliae* were concordant with their responses to *V. nonalfalfae* with the lone exception being Longshu 11, which was resistant to *V. dahliae* but was not resistant to *V. nonalfalfae*. Gurung et al. (2015) showed that lettuce cultivars resistant to *V. dahliae* were also resistant to isolates of *Verticillium isaacii* and *Verticillium klebahnii*. Thus, cultivars identified as resistant to the more dominant pathogen are likely to carry resistance to other species of the pathogen that generally are less widespread and cause disease in niche environments. However, inconsistency in the response of potato cultivars to different *Verticillium* spp. has been reported previously (Concibido et al. 1994). Three clones differed in their responses to *V. dahliae* and *Verticillium albo-atrum*. In this study, the isolate Vnp24 was the most virulent strain among 20 *V. nonalfalfae* isolates tested for pathogenicity before this experiment (data not shown). The high level of virulence in this strain may partially explain why Longshu 11, which was resistant to Vdp83, was less resistant to Vnp83. Atlantic, an international cultivar that is widely grown worldwide, showed typical leaf necrosis and vascular discoloration when inoculated with *V. dahliae* with a median disease rating of 2; the cultivar was also susceptible and showed typical symptoms to

Table 2. Results from the nonparametric analysis of Verticillium wilt responses of 30 potato cultivars against *Verticillium dahliae* strain Vdp83 using the PROC MIXED procedure in SAS

		Wald-type statistic				ANOVA-type statistic			
Effect	***n***	**df*N***	**df*D***	**χ^2**	***P* value**	**df*N***	**df*D***	***F***	***P* value**
Cultivar	30	29	236	1,330.79	<0.0001	29	236	45.89	<0.0001
Experiment	2	1	236	0.03	0.8690	1	236	0.03	0.8692
Time	5	4	236	298.54	<0.0001	4	236	74.64	<0.0001
Cultivar × experiment	60	29	236	6.43	1.0000	29	236	0.22	1.0000

ANOVA, analysis of variance; df*D*, denominator degree of freedom; df*N*, numerator degree of freedom.

V. nonalfalfae with a median disease rating of 2. In contrast to these results, Saremi and Amiri (2010) reported that Atlantic was a moderately resistant cultivar against wilt, Webb et al. (1978) characterized it as a tolerant cultivar against Verticillium wilt, and yet, others (Nair et al. 2016; Whitworth and Davidson 2008) considered it as a moderately susceptible cultivar. Thus, responses of the same cultivars vary based on the pathogen isolates and the disease rating scales used, making comparisons between studies extremely difficult.

This study showed that Qingshu 9, Zhongshu 18, Longshu 8, and Zhongshu 19 were resistant to both *V. dahliae* and *V. nonalfalfae*. Of these cultivars, Qingshu 9 was also resistant to late blight (Hu et al. 2014); Zhongshu 18 was resistant to potato mosaic virus X and Y but slightly susceptible to late blight (Bian 2014); and Longshu 8 was resistant to late blight, potato mosaic virus, and potato leaf-roll virus (Li et al. 2010). These cultivars could, therefore, be good germplasm resources for potato resistant breeding programs, and until new

Table 3. Mean ranks, relative marginal effect value, variance, and confidence intervals of Verticillium wilt severity ratings on 30 potato cultivars inoculated with *Verticillium dahliae* strain Vdp83

Cultivar	Mean ranks	Relative marginal effect	Variance	95% Confidence low limit	95% Confidence up limit
Helan 15	59.5	0.98	0.000	0.98	0.98
Longshu 6	57.0	0.94	0.008	0.91	0.96
Zaodabai	56.0	0.93	0.008	0.90	0.94
L0527-4	52.5	0.87	0.025	0.82	0.90
Zhuangshu 3	51.0	0.84	0.025	0.80	0.88
Xindaping	49.0	0.81	0.217	0.66	0.90
Longshu 9	48.5	0.80	0.017	0.77	0.83
Longshu 12	44.5	0.73	0.042	0.68	0.78
L0529-2	44.0	0.73	0.117	0.63	0.80
Caishu	42.0	0.69	0.050	0.63	0.74
Longshu 10	40.5	0.67	0.025	0.63	0.71
Tianshu 12	35.5	0.58	0.033	0.54	0.63
Jizhangshu 14	34.5	0.57	0.158	0.46	0.66
Longshu 7	33.0	0.54	0.208	0.43	0.65
L0109-4	32.5	0.53	0.050	0.48	0.59
Longshu 3	32.0	0.53	0.058	0.46	0.59
Atlantic	26.5	0.43	0.025	0.39	0.47
Huangyangyu	25.0	0.41	0.025	0.37	0.45
Jinshu 16	25.0	0.41	0.042	0.36	0.46
Longshu 11	21.5	0.33	0.030	0.29	0.37
L0227-18	19.5	0.32	0.025	0.28	0.36
Longshu 13	18.5	0.30	0.030	0.26	0.34
Zhongshu 20	15.0	0.24	0.008	0.22	0.27
Z2011-1	13.0	0.21	0.042	0.16	0.27
Zhongshu 19	12.5	0.20	0.017	0.17	0.23
Hongyun	9.5	0.15	0.000	0.15	0.15
Longshu 8	6.0	0.09	0.038	0.06	0.16
Qingshu 9	4.0	0.06	0.004	0.04	0.08
Zhongshu 21	4.0	0.06	0.004	0.04	0.08
Zhongshu 18	4.0	0.06	0.004	0.04	0.08

Table 4. Results from the nonparametric analysis of Verticillium wilt responses of nine potato cultivars to *Verticillium nonalfalfae* strain Vnp24 using PROC MIXED procedure in SAS

		Wald-type statistic				ANOVA-type statistic			
Effect	*n*	df*N*	df*D*	χ^2	*P* value	df*N*	df*D*	*F*	*P* value
Cultivar	9	8	51	1,586.24	<0.0001	8	51	198.28	<0.0001
Experiment	2	1	51	0.44	0.5059	1	51	0.44	0.5089
Time	4	3	51	33.61	<0.0001	3	51	11.2	<0.0001
Cultivar × experiment	18	8	51	8.96	0.3455	8	51	1.12	0.3658

ANOVA, analysis of variance; df*D*, denominator degree of freedom; df*N*, numerator degree of freedom.

Table 5. Mean ranks, relative marginal effect value, variance, and confidence intervals of Verticillium wilt severity ratings on nine potato cultivars inoculated with *Verticillium nonalfalfae* strain Vnp24

Cultivar	Mean ranks	Relative marginal effect	Variance	95% Confidence low limit	95% Confidence up limit
Xindaping	16.50	0.89	0.056	0.63	0.94
Longshu 6	16.00	0.86	0.139	0.50	0.94
Longshu 7	14.00	0.75	0.028	0.66	0.82
Atlantic	11.00	0.58	0.028	0.50	0.66
Longshu 11	10.00	0.53	0.028	0.45	0.60
Zhongshu 19	7.50	0.39	0.000	0.39	0.39
Longshu 8	4.25	0.21	0.090	0.11	0.40
Zhongshu 18	3.75	0.18	0.063	0.10	0.34
Qingshu 9	2.50	0.11	0.028	0.07	0.26

resistant cultivars emerge from these programs, they are likely to remain the sole sources of resistance for Verticillium wilt management.

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