

Black knot [*Apiosporina morbosa* (Schw.)] resistance in imported and domestic *Prunus domestica* L. germplasm and cultivars

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Abstract

Black knot *Apiosporina morbosa* (Schw.) (BK) is an important fungal disease of *Prunus domestica* and other *Prunus* species in North America. BK causes economic losses in the plum growing regions of northern and eastern U.S., and eastern Canada. Relatively few *P. domestica* commercial cultivars are resistant to BK. Evaluation of resistance to BK in *P. domestica* plums including an evaluation of infection of new shoot growth following pruning was carried out over four years. The 43 genotypes evaluated included 23 accessions from the U.S. National Plant Germplasm System (NPGS)-Davis, California and cultivars and selections from the U.S., Canada, and Europe. Based on the evaluations, genotypes were classified as highly resistant, moderately resistant, susceptible or highly susceptible. Highly resistant genotypes, without visible symptoms made up 44% of the germplasm evaluated and of these, 68% were accessions from the U.S. NPGS collection. While the inheritance of resistance remains to be determined, these evaluations indicate that sources of high level resistance are available in *P. domestica* that can be utilized in breeding highly resistant commercial cultivars.

Black knot (*Apiosporina morbosa* (Schw.) (BK), is a fungal disease of plum species that is found throughout North America and is particularly serious in the northern and mid-Atlantic U.S. and eastern Canada. Host species include *Prunus cerasifera* Ehrh., *P. domestica* L., *P. pennsylvanica* L., *P. salicina* Lindl., *P. serotina* Ehrh., and *P. virginiana* L. (McFaden-Smith et al., 2000; Sutton and Waterson, 1970). BK conidia appear in the spring followed by ascospores which are then responsible for the spread of the disease (Cornell Univ., 1970; Sutton and Waterson, 1970). Disease symptoms first develop on shoots at the base of petioles. Abnormal growths ("knots") follow usually one year after infection and these knots harden by the end of the growing season (Douglas 2008; McFadden-Smith et al., 2000; Ritchie et al., 1975). The portions of a branch distal to a knot become stunted, and knots may enlarge to girdle branches causing their death (Babadost, 2000). Trees with multiple infections

become dwarfed and misshapen, markedly reducing their productivity (Snover and Arneson, 2002). Infected trees lose value after a few years. BK is a disease of economic significance to U.S. and Canadian *P. domestica* plum growers. In some parts of the northeast and midwest, and in Canada, BK is a major limiting factor for commercial fresh plum and prune production (Cornell Univ., 2007; Gourley, 1962). Losses in plum due to BK have been estimated at 10% (Cramer, 1967 in EPPO).

BK is not easily controlled and requires a combination of fungicide applications and pruning of infected branches (Douglas, 2008). Re-infection can occur from wild *Prunus* species and poorly managed or abandoned plantings. Few commercial varieties of *P. domestica* plum are resistant to BK (Table 1) and new sources of high level resistance to this disease would be valuable for breeders and growers.

The USDA-ARS National Plant Germ-

Table 1: Relative susceptibility of plum cultivars to Black Knot (Babadoost, 2000; Carter et al., 2005; Cornell Univ., 2007; Douglas 2008)

<u>Very Susceptible</u>	
	Bluefre
	Damson
	Lombard
	Shropshire
	Stanley
	Veeblue
	Vision
<u>Moderately Resistant/Susceptible</u>	
	Early Italian (Early Fellenburg)
	Italian (Fellenburg)
	Valor
	Vanier
	Verity
	Voyageur
	Valerie
	Vanette
<u>Slightly Susceptible</u>	
	German Prune
<u>Resistant</u>	
	President

plasm System (NPGS) is an important source of germplasm collected world-wide presenting a wide range of potentially useful traits for plant breeders and growers. Currently there is no information on BK resistance associated with *P. domestica* accessions in the NPGS system. Minutes of the 2011 Prunus Crop Germplasm Committee meeting indicate that “Phenotyping data is critical and expensive to obtain. It can be performed while waiting for appropriate genomic technology..... For the next few years at least, we should focus our attentions on phenotypic evaluation of the collections.” (http://www.google.com/url?sa=t&rct=j&q=&esrc=s&source=web&cd=10&ved=0CF8QFjAJ&url=http%3A%2F%2Fwww.ars-grin.gov%2Fnpgs%2Fcgcr-reports%2Fapprul1.doc&ei=63oCVJfwB4y5ggSj-YDYDA&usg=AFQjCNFJaH_yaAu-Qa7jbBfEG-xkzKjK_1A&sig2=8FMjSLgvWta4p4HIyulaew, accessed 9-2014).

The research reported here was undertaken to provide data on resistance to BK in *P. domestica* and in particular in germplasm available through the U.S. NPGS.

Materials and Methods

Twenty-three *P. domestica* accessions from the U.S. NPGS-Davis, California (http://www.ars.usda.gov/main/site_main.htm?modecode=20-32-10-00, accessed 10-2014), were propagated onto ‘Brompton’ plum seedling rootstocks, and were planted in 1997 at the USDA-ARS Appalachian Fruit Research Station, Kearneysville, WV (Table 2). Trees were originally planted in a randomized block design in four blocks with a single tree replication per block, although over the years of observation dead trees were eliminated and additional genotypes were planted as they were received. The planting was nat-

Table 2: Plums (*P. domestica*) from the U.S. National Plant Germplasm System (US NPGS) evaluated for black knot resistance.

Genotype	NPGS Identifiers	Origin
Anna Spath	DPRU 1963	Romania
Burja	DPRU 1593; PI 506395	Bulgaria
Cacanska Rana	DPRU 1838; Q 22626	Yugoslavia
Fursts Fruhwetsche	PI 131179	Germany
Jojo ^z	Q 43772	Germany
Kinstendisliwa	DPRU 1594; PI 506396	Bulgaria
Kirkes	PI 133578, DPRU 0348	Australia
Obil'naja	DPRU 0734; PI 406688	USSR ^y
Ortenauer x Stanley #34 ^x	Q 43774	Germany
Pozegaca	DPRU 1616; PI536691	Yugoslavia
Pozegaca D-6	DPRU 1649, PI 543956	Yugoslavia
Pozegaca D-13	DPRU1613; PI 536688	Yugoslavia
Pozegaca P-24	DPRU1615; PI 536689	Yugoslavia
Pozegaca P-25	DPRU1614; PI 536690	Yugoslavia
C7	DPRU 2263	Yugoslavia
C8	DPRU 2264	Yugoslavia
C10	DPRU 2265	Yugoslavia
C11	DPRU 2266; Q 25826A3	Yugoslavia
V10	Q 25826A3	Yugoslavia
Reine Claude De Bavay	DPRU 0927	Belgium ^w
Ruth Gerstetter	DPRU 1630	Germany
Sans Noyau ^y	DPRU 2419	USA
Tamaioasa de Bristrita	Q 23698b	Romania

^z Lichtenegger et al, 2012

^y <http://www.ars-grin.gov/cgi-bin/npgs/acc/display.pl?1306406>, accessed 9-14

^x Lichtenegger et al., 2012

^w <http://www.ars-grin.gov/cgi-bin/npgs/acc/display.pl?1010096>, accessed 9-14

^v Callahan et al., 2009, 2013

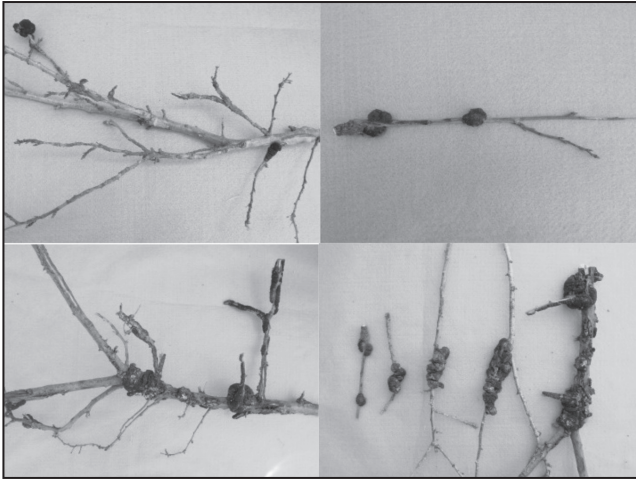


Fig 1: Black knot (BK) severity ratings based on 1-4 scales of “knot” size and incidence of infection on a tree: Upper left: size 2 frequency 1; upper right: size 2, frequency 2; lower-left: size 3 frequency 3; lower right size 4 frequency 4. Size and frequency ratings were averaged for a 1-4 scale of susceptibility.

urally exposed to BK which is endemic to the U.S. Mid-Atlantic area where the planting is located. Additionally, *P. domestica* plantings that had been planted in 1979 on the research station grounds provided local inoculum to the test site. BK infection was rated on a 1-4 scale of symptoms and a 1-4 scale of incidence including a no symptom (zero) rating (Fig. 1). Included in the evaluation were standard varieties of known susceptibility to BK including the highly susceptible and widely grown fresh market cultivar ‘Stanley’ (Fig. 2). ‘President’, also a popular variety, has been rated as highly resistant (Carter et al., 2005; Douglas, 2008; Univ. of Illinois, 2000) (Table 1). ‘Bluebyrd’ (Scorza and Fogle, 1999) and ‘HoneySweet’ (Scorza et al., 2013) have also been found to be highly resistant in our long-term observations, and ‘Orablu’ highly susceptible. Included in this report are evaluations of commercial fresh market cultivars from the U.S. and Canada that were planted in the test plot. Descriptions of many of these cultivars can be found in Andersen et al. (2006), Brooks and Olmo (1997), and Okie and Ramming (1999).

The test planting was generally only lightly pruned, if at all, in most years except in

2012 when a more extensive pruning was carried out. BK infection was rated in 2011, 2012 and 2013. In 2014, re-infection of new growth that followed the 2012 pruning was rated. All ratings of BK infection were made by the same evaluators in the spring of each year prior to leafing-out.

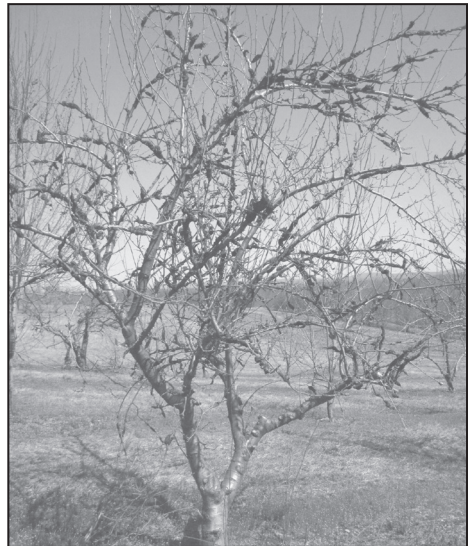


Fig 2: ‘Stanley’ tree (15 years) severely infected with black knot ([*Apiosporina morbosa* (Schw.)]).

Table 3: Overall black knot ratings on *Prunus domestica* trees in 2011, 2012, and 2013 under natural infection pressure. Ratings are a combination of 1-4 severity and 1-4 incidence ratings, summed and divided by 2. No visible infection was rated as zero. Average for 3 years is presented and forms the basis for ordering the genotypes. Following pruning to remove overhanging and dead limbs in 2012 infection of new shoots was rated in 2014. Average ratings of zero are considered highly resistant, greater than zero and less than 2.2 are moderately resistant, greater than 2.2 and less than 2.8 are susceptible, 2.8 and greater are highly susceptible. Blank cells indicate data not recorded.

Genotype	2011	2012	2013	3 year average	2014 post pruning
Bluebyrd	0	0	0	0	0
Burja	0	0	0	0	0
Early Italian (Early Fellenburg)	0		0	0	0
Green Gage		0	0	0	
HoneySweet	0	0	0	0	0
Obil'naja	0		0	0	0
UC Pitless ^z	0		0	0	0
USDA Pitless ^y	0		0	0	0
<i>Pozegaca types</i>					
C7	0		0	0	0
C8	0		0	0	0
C10	0		0	0	0
C11	0		0	0	0
D6	0		0	0	0
D13	0		0	0	0
P24	0		0	0	0
P25	0		0	0	0.6
Pozegaca	0		0	0	0
V10	0		0	0	0
Sans Noyau	0		0	0	0
President	0.5	0	0	0.2	0
Ruth Gerstetter	0	0.8	0	0.3	0
CA Stanley	0.3	0	0.7	0.3	0
Vision	0		0.5	0.3	0
Kinstendsliva	0		0.7	0.4	1.1
Tamaioasa de Bistrita	0	1.0	1.0	0.7	1.0
Italian (Fellenberg)		1.5	0	0.8	
Long John	2.0		0	1.0	0
Bluefre	1.5		1.0	1.3	0
Prune 707	1.0	3.0	0	1.3	
Reine Claude de Bavay	1.5	2.0	0.7	1.4	0
Jojo	1.9	1.7	1.2	1.6	0.8
Kirkes	1.5	2.3	1.8	1.9	1.4
Seneca		1.3	2.5	1.9	0
Fursts Fruhwetsche	1.5 ^x	2.0 ^w	3.0	2.2	
Grand Prize	1.3	3.0	2.3	2.2	0
Ortenauer x Stanley #34	2.0	2.8	2.0	2.3	1.6
Anna Spath	4.0		1.0	2.5	
Cacanska lepotica	4.0	2.5	1.5	2.7	0
Cacanska Rana	3.0 ^x	3.0 ^w	2.5	2.8	
Castelton	4.0	4.0	1.3	3.1	0
Stanley	3.5	3.0	3.5	3.3	0.9
Orablue	3.8	3.3	3.5	3.5	1.3
Blue Ribbon	3.5	3.5	3.8	3.6	0

^z Obtained from University of California, Davis, CA.
^y Obtained from USDA, Parlier, CA.
^x 2007 rating, no rating in 2011.
^w 2008 rating, no rating in 2012.

Results and Discussion

Of the 43 genotypes evaluated (including 23 accessions from NPGS-Davis) (Table 3) 33 (77%) can be classified as highly or

moderately resistant, and of these, 19 (44%) were without visible symptoms. Nineteen of the highly or moderately resistant genotypes (58%) were accessions from the NP-

GS-Davis collection and 13 (68%) of the highly resistant (no visible infection) were also NPGS-Davis germplasm accessions. The average of the ratings for genotypes in the susceptible and highly susceptible classes (average rating 2.2 and greater) was 3.1 in 2011, 3.0 in 2012, and 2.8 in 2013 suggesting a year to year uniformity in the infection ratings. Pruning in 2012 led to the development of vigorous new shoots which were evaluated for new infections in 2014. The generally low level of new infections observed in 2014 suggests that except for the most susceptible genotypes there was insufficient time for symptom development. The rapid re-infection of highly susceptible genotypes points to the value of resistance in disease management.

Our results for the varieties of known susceptibility are confirmatory of other sources (Table 1) with the exception of ‘Vision’ which is generally rated as highly susceptible but was rated as moderately resistant in our tests, and ‘Italian’ (‘Fellenberg’) and ‘Early Italian’ (‘Early Fellenberg’) which we rated as more resistant than generally reported (Table 1).

A group of U.S. NPGS-Davis accessions that were highly resistant with no symptoms were the Pozegaca types obtained from the former Yugoslavia as part of a U.S. PL480 project in which the first author participated. This landrace of plums has an ancient origin. Emperor Gaius Aurelius Valerius Diocletianus (245-313) established large plum orchards of the cultivar ‘De Bosnia’ on the banks of the Drava and Sava rivers in Croatia. ‘Pozegaca’ was developed from these plantings (Ramming and Cociu, 1990; Faust and Surányi, 1999). ‘Pozegaca’ trees have been seed propagated in the past and may continue to be seed propagated in some localities. Elite seedlings have been selected over the centuries and vegetatively propagated from root suckers, producing a wide range of types within the same general group (Rozpara et al., 1998). Based on phenotype it appears that the “common prune”

which is known as ‘Pozegaca’ in Bosnia-Herzegovina, Croatia, Serbia, and Bulgaria, known as ‘Wegierka’ in Poland, ‘Bystricka’ in Romania, ‘Domaci Svestka’ in Czech Republic, and ‘Hauszwetsche’ in Germany is, as a group of the same ancient origin. These are small sweet plums highly appreciated and utilized for fresh consumption, baking, confections, and the distillation of spirits. The production of “common prune” has been severely affected by *Plum pox virus* to which this group of plums is highly susceptible (Drkenda and Kurtović, 2012). In our experimental orchard the Pozegaca types are typically among the latest blooming and latest in fruit ripening. Our observations of fruit phenotype, flowering date, and resistance to BK suggest that ‘Burja’ may also be a “common prune” type. Several “pitless” plums including ‘Sans Noyau’ that were likely imported from France by Luther Burbank (Callahan et al., 2009, 2013) also appear to be highly resistant to BK. In addition to U.S. NPGS-Davis accessions, ‘Bluebyrd’, a seedling from open pollination of NY H4, a selection from the New York State Agricultural Experiment Station, Geneva, NY (Scorza and Fogle, 1999), and ‘HoneySweet’, an open-pollinated seedling of ‘Bluebyrd’ have never shown BK infection over the course of the present study and for many years prior to this study. ‘HoneySweet’ is a cultivar genetically engineered for high level resistance to *Plum pox virus* (Scorza et al., 2013). ‘Bluebyrd’ and ‘HoneySweet’ are both fresh market types developed from genotypes adapted to north and eastern U.S., and eastern Canada.

Norton and Boyhan (1991) studied the inheritance of resistance to BK in a number of interspecific crosses, none of which included *P. domestica* which is a hexaploid species that is sexually incompatible with most other *Prunus* species. These authors concluded that resistance was controlled by a single recessive gene (*bk*). In our study a number of U.S. NPGS accessions show a high degree of resistance to BK as do several domestic cultivars. It is not known if the resistance in

P. domestica is distinct from that reported by Norton and Boyhan (1991) or if there are different alleles for resistance in *P. domestica*. Further genetic and molecular studies would be important in clarifying the genetic nature of BK resistance in *P. domestica* and verifying the value of this resistance in the U.S. NPGS collection.

The utilization of rapid cycle crop-breeding ('FasTrack' breeding) (Srinivasan et al., 2012; <http://ucanr.edu/sites/fastrack/Approach/>, accessed 10-2014) can be used to accelerate the development of new BK resistant plum varieties utilizing the U.S. NPGS germplasm identified in this study.

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