SCREENING A LARGE SET OF GRAPE ACCESSIONS FOR RESISTANCE AGAINST BLACK ROT (*GUIGNARDIA BIDWELLII /(ELL.)*)

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Beside grey rot, powdery and downy mildew, black rot is one of the most dangerous diseases of grapevine all over the world. In epidemic years it can cause crop losses up to 100 %. Black rot was first observed in Hungary, in 1999 thereafter favourable weather conditions enabled the development of a country-wide epidemic in 2010. Since that time epidemics appeared almost every year in Hungary. Newly developed grapevine varieties, highly resistant against powdery and downy mildew, cultivated without any chemical plant protection, are susceptible to black rot, so in order to avoid the increase of considerable economic losses plant protection is definitely necessary. Our aim is finding appropriate resistance sources for black rot resistance and integrating them into the newly developed fungus resistant grapevine assortment. The present work evaluates numerous grape accessions for black rot resistance: French-American hybrids and their backcrosses, *Vitis amurensis* hybrids and *Vitis vinifera* varieties indigenous in Georgia. The varieties 'Csillám', 'Seyval blanc' ('SV 5276') and two *Vitis amurensis* hybrids were identified as highly resistant against both leaf and berry infections.
Keywords: *Vitis vinifera* L., *Vitis* interspecific hybrids, fungal diseases, black rot, resistance breeding, phenotyping

Überprüfung einer großen Anzahl von Reb-Akzessionen auf Resistenz gegen Schwarzfäule *(Guignardia bidwellii /(Ell.))*. Schwarzfäule ist neben Grauschimmel, Echtem und Falschem Mehltau eine der bedeutendsten Rebkrankheiten weltweit. In epidemischen Jahren können die Ernteverluste bis zu 100 % betragen. In Ungarn wurde die Schwarzfäule das erste Mal im Jahr 1999 beobachtet, danach ermöglichte die günstige Witterung eine landesweite Epidemie im Jahr 2010. Seitdem treten Schwarzfäule-Epidemien fast jedes Jahr auf. Neugezüchtete Rebsorten, die hohe Resistenz gegen Echten und Falschen Mehltau besitzen und deswegen ohne Pflanzenschutzmaßnahmen kultiviert werden, sind sehr anfällig für Schwarzfäule und brauchen unbedingt chemische Behandlung, um beträchtliche ökonomische Verluste zu verhindern. Unser Ziel ist das Auffinden geeigneter Resistenzquellen gegen Schwarzfäule und diese Eigenschaft in neugezüchtete, mehltau-resistente Rebsorten zu integrieren. In der gegenständlichen Untersuchung wurden zahlreiche Reb-Akzessionen in Hinblick auf ihre Resistenz gegen Schwarzfäule evaluiert: franko-amerikanische Hybride und deren Rückkreuzungen, *Vitis amurensis*-Hybride und *Vitis vinifera*-Sorten, die in Georgien beheimatet sind. Die Sorten 'Csillám', 'Seyval blanc' (SV5276) und zwei *Vitis amurensis*-Hybride wurden als hoch-resistent identifiziert, sowohl gegen Blatt- als auch Beereninfektionen.
Schlagwörter: *Vitis vinifera* L., *Vitis*-interspezifische Hybride, Pilzerkrankungen, Schwarzfäule, Resistenzzüchtung, Phänotypisierung

Main diseases of grapevine, powdery (*Erysiphe necator* Schw.) and downy mildew (*Plasmopara viticola* (Berk. & M.A. Curtis) Berl. & de Toni), and black rot (*Guignardia bidwellii* (Ellis) Viala & Ravaz) were inadvertently introduced to Europe by contaminated propagation material from North-America in the second half of the 19th century. The European grapevine had not met these pathogens during its evolution, hence resistance is very rare among varieties and not present in economically important varieties. Environmental conditions were favourable for mildews in all winegrowing regions of Europe, thus they spread quickly through countries causing epidemics since the end of the 19th century. Breeding programs for creating resistant varieties were started shortly after the emergence of epidemics in the late 19th century and aimed to incorporate resistances to downy and powdery mildew into new varieties. However, besides mildews no other pathogen was in the target of European resistance breeding.

The fungal pathogen *G. bidwellii* was first described in France in 1885 (Scribner and Viala, 1888), shortly after mildews, but climatic conditions enabled the development of disease symptoms only in Southern Europe. The occurrence of black rot remained sporadic until the latest decades in most of the European winegrowing regions. *G. bidwellii* is able to bring about infection only if leaf wetness is constant for several hours and the temperature is mild (Spotts, 1977). As long lasting humid periods became frequent in spring and summer also in continental areas like Hungary, black rot emerged also here as a major disease of grape.

The fungus is able to infect all expanding green parts of the plant (leaf, shoot, tendril, cluster). Symptoms appear 14 to 21 days after infection, as oval brown-reddish edged, light-tan centered necrotic lesions on the leaf and as larger and darker, elongated spots on the stem, cane and tendril. Inside the spots, pycnidia usually develop in regular circles around the perimeter (Scribner and Viala, 1888). The size and number of lesions determine the severity of the infection. In serious cases, spots can merge and necrotize the whole leaf. The infected berry shrivels from a soft brown rotten fruit to a small, black, hard mummy within 4 to 5 days (Wilcox, 2003). The economic loss is due to the devastation of the clusters and to the resulting yield losses (Ramsdell and Milholland, 1988).

Since 2010 epidemics are increasingly common in grape growing regions of Europe, including Hungary, causing severe crop losses. Traditional spray programs targeting mildews slow the appearance of black rot (Spotts, 1977a and 1977b; Dula, 2012), but vineyards of mildew-resistant varieties and plantations under organic cultivation regime suffered significant economic losses. The ideal solution from economic, environmental and human health aspects would be the chemical-free grape production on the basis of varieties with multiple resistances against black rot and mildews. As European breeding programs have not dealt with black rot disease before, identification of accessions with employable resistance are necessary at first. The aim of this study is to screen a large set of genotypes with wide genetic backgrounds to find source material for resistance breeding.

North American *Vitis* species (*Vitis cinerea*, *Vitis rupestris*, *Vitis lincecumii*) are known to possess resistances against black rot, because these grapes have co-evolved with the pathogen (Barett, 1953). French-American hybrids that merge the genomes of several North-American Vitis species, can be potential black rot resistance sources, as resistance loci could have been preserved through backcross generations. Resistance of these hybrids and varieties is inherited mainly from *Vitis rupestris* and *Vitis lincecumii* which possess high level resistance against black rot (Herbert and Barrett, 1953; Galet, 1988; Jabco et al., 1985). French-American hybrids are results of selections also for fruit quality, thus they are more valuable resistance sources than their wild progenitors (Galet, 1988; Skelton, 2010; Cattell, 2014).

There is evidence, that wild grape species indigenous in Asia can also possess high level resistances against fungal diseases like powdery and downy mildew, species which to our latest knowledge also did not co-evolve with these pathogens (Wan et al., 2007; Hoffmann et al., 2008; Riaz et al., 2013; Ramming et al., 2011; Pap et al., 2016). Black rot resistance among grape species native to Asia has not been investigated yet. Remarkable achievements in finding new sources of powdery and downy mildew resistances among unrelated germplasm urge us to seek also for black rot resistance in various gene pools.

MATERIALS AND METHODS

PLANT MATERIAL SCREENED FOR BLACK ROT RESISTANCE

Twelve accessions of French-American hybrids, 61 accessions of Seyve-Villard hybrids' progenies created in European breeding programs, 26 accessions of *Vitis amurensis* x *Vitis vinifera* F2, BC1 and BC2 hybrids created in Hungarian breeding programs, 7 accessions of *Muscadinia rotundifolia* x *V. vinifera* x *V. amurensis* x French-American complex hybrids created in Pécs, 57 accessions of Vitis vinifera varieties indigenous in subtropical areas of Georgia and 2 accessions of Vitis vinifera varieties indigenous in central Asia were screened for black rot resistance. The rootstock variety 'Börner' (*Vitis riparia* x *Vitis cinerea*) and 3 seedlings of Muscadinia rotundifolia from open pollination were used as resistant controls, two Vitis vinifera varieties ('Chardonnay' and 'Furmint') were used as sensitive controls.

ARTIFICIAL INOCULATION

Woody cuttings were used to create rooted plants for artificial inoculation with the fungus. Cuttings were grown in greenhouse, shoots were cut back regularly to maintain the active growing stage. Infection was carried out on actively growing shoot tips and the 4 upper leaf stages of the shoot, in the three successive years of 2014, 2015 and 2016. Three repetitions per genotype were investigated at each evaluation. The average of 3 repetitions at a time presents a score for each genotype shown in Table 1. Young clusters developing on the potted cuttings were also infected in 2015 and 2016 around the 2-week-post-bloom stage. Conidia suspension was sprayed onto shoot tips and young clusters of potted test plants which were wetted with tap water before spraying. Damp was developed in the chamber until 100 % relative humidity was reached. The incubation took place under controlled conditions: temperature between 22 to 24 °C, a diurnal cycle of 12 h day/night and relative humidity just below saturation during the first 2 days post-inoculation, then reduced to 60 % relative humidity.

Table 1: List of grape varieties which gave moderate to high-level leaf resistance to black rot in artificial tests. Columns show five independent artificial testing time points between 2014 and 2016 for leaf infection and one for berry infection in 2016. Scores of leaf tests mean the following resistance levels: 1 = high-level susceptibility; 3 = moderate susceptibility; 5 = moderate resistance, 7 = high-level resistance; 9 = symptomless resistance. Berry resistance was classified into four categories: HS = highly susceptible, more than 50 % of berries turned into mummies; MS = moderately susceptible, less than 50 % of berries turned into mummies; R = resistant, intact berries with superficial spots without mummies; SR = symptomless resistance

Variety	Parentage/Origin		2014		2015		2016	
			I	II	I	II	I	
			Leaf	Leaf	Leaf	Leaf	Leaf	Berry
Seibel 7053	Seibel 5163	Seibel 880	9	9	9	9	9	HS
Seyve-Villard 5276	Seibel 5656	Seibel 4986	9	5, 9	9	7	7	R
Seyve-Villard 12375	Seibel 6468	Seibel 6905	3	5	4	3	4	MS
Seyve-Villard 18315	Seibel 7053	Seibel 6905		7	4		3	HS
Teréz	SV 12375	Olimpia		9	9	6	4	HS
Csillám (CSFT 194)	S 4643	Kékfrankos	9	9	9	9	9	SR
Felicia	Sirius (Bacchus x SV12375)	Vidal blanc	5		9		8	
Villaris	Sirius (Bacchus x SV12375)	SV12375	9	5, 7	9		5	
Merzling	SV5276	Riesling x Pinot gris	5		9		7	R
GM 318-57	Seibel 7053	Riesling Gm239	9	7	7	5	5	HS
Malverina	SV 12375 x Malvasia	Seibel 13666 x Merlot	7	9	5	7	3	HS
5-11-2	V. amurensis x Riesling	V. amurensis x Tramini	9		6-7	9	7	R
5-10-6	V. amurensis x Riesling	V. amurensis x Tramini	9		7	9	9	R
5-11-6	V. amurensis x Riesling	V. amurensis x Tramini	9		3		9	R
Muradouli	Georgian variety			4		9	5	
Ojaleshi	Georgian variety			5		5	4	
Bianca (control)	SV 12375	Bouvier	1	1	1		1	HS
Börner (control)	Vitis riparia Gm 183	Vitis cinerea Arnold	9	9	9	7	9	
Furmint (control)	V. vinifera variety		1	1	1	3	1	HS
Chardonnay	V. vinifera variety		1	1	1	3	1	HS

INFECTION MATERIAL

G. bidwellii isolates were collected in the vineyard of the Institute in Pécs and their identity was verified with molecular methods. Ribosomal DNA (LSU) was amplified with LR0R (5'- ACCCGCTGAACTTAAGC-3') and LR5 (5'- TCCTGAGGGAAACTTCG-3') primers and sequenced. The nucleotide sequence of the PCR products was established to be identical to the corresponding sequence of *G. bidwellii* as determined by sequence alignments using BLAST (Madden, 2002; Váczy et al., 2012). For production of inoculum used in artificial inoculations, the fungus was propagated *in vitro* on oatmeal and ½ PDA media. Spores were washed off with purified water from 14 to 16 days old cultures. The conidia suspension was diluted to 4 to 6 x 10^6 spores/ml.

DISEASE SYMPTOM EVALUATION

Disease severity on leaf was evaluated 21 days post-inoculation (dpi) and repeated after 7 days. As result the fully developed symptoms were recorded, for some accessions it was on the 21 dpi, for others on 28 dpi. Scoring was based on the number and size of leaf and shoot lesions and the presence of pycnidia, using the 1-to-9 classification scale developed by Barrett (1955). The classes of severity describe the following symptoms: 9 - completely symptomless, 7 - few small lesions without pycnidia, 5 - few lesions bigger in size with some undeveloped pycnidia, 3 - several lesions with many well-developed pycnidia, 1 - large or merging lesions on many leaves with large amounts of well-developed pycnidia.

Disease severity on berries was evaluated 21 days post-inoculation for the first time, thereafter resistant phenotypes were pursued until full ripening. Classes of resistance were developed based on the experience of the tests, as no rating for berry resistance was available from publications. Four categories were distinguished for evaluation: symptomlessly resistant (SR): not any symptoms appeared; resistant (R): symptoms appeared as superficial spots; moderately susceptible (MS): less than 50 % of berries developed into mummies; highly susceptible (HS): most of the berries developed into mummies.

RESULTS

Results from molecular analysis of the *G. bidwellii* isolate that was used for artificial infections revealed identity with the *G. bidwellii* (accession CBS 237.48) sequence in GenBank. This proves, that infections were carried out with the real causal agent of black rot.

From the French-American group of varieties 1 Seibel and 11 Seyve-Villard hybrids were evaluated for resistance to black rot. Results of the artificial infections are demonstrated in Table 1, including only accessions which were evaluated tolerant to resistant. 'Chancellor' ('Seibel 7053') showed symptomless leaf resistance at each test. 'Seyval blanc' ('Seyve-Villard 5276') also had outstanding resistance, it was symptomless in 3 of the 4 tests. 'Villard noir' ('SV18315') was moderately resistant, all the other tested accessions were classified susceptible or highly susceptible. Resistance of 'Villard blanc' ('Seyve-Villard 12375') was found inconsequent, and this variety was placed between categories moderately sensitive and moderately resistant. 61 descendants of French-American hybrids were screened for black rot resistance, which were grouped based on their resistant progenitors (SV 12375, Seibel 7053, Seibel 4986, Seibel 4643) as can be seen in table 2.

'Villard blanc' was the most commonly used resistance source in breeding programs due to its excellent combination ability with several partners, good resistance to mildews and high fruit quality (Csepregi and Zilai, 1988). 37 descendants of 'Villard blanc' were screened in our experiments, the majority of which grouped into the susceptible or highly susceptible class. 'Villaris' and 'Malverina' showed moderate resistance, 'Teréz' was the only variety with high-level resistance. Of the descendants of 'Chancellor', four accessions were tested. 'GM318-57' showed high-level resistance in 3 of the 4 independent tests and had moderate symptoms at one occasion. 'GM 78-10-8' was moderately susceptible, 'Hibernal', 'GM322-58' and 'MM32'were highly susceptible. 'Seyval blanc' ('SV5276') from the 'Rayon d'or' ('Seibel 4986') descendants' group showed outstanding black rot resistance, while 'Merzling' and 'Felicia' were classified as moderately resistant. The Hungarian bred interspecific variety 'Csillám', originating from a cross of 'Roi des noirs' and 'Kékfrankos' (Kiss et al., 2017;) was the only variety in the screening which did not show any symptoms in four independent assays.

Table 2: Black rot resistance level of descendants of French-American hybrids classified in groups based on their genetic origin

Resistance groups	French-American Hybrid Progenitors			
	SV 12375	Seibel 7053	Seibel 4986	Seibel 4643
Symptomless resistance (9)				Csillám (CSFT 194)
High-level resistance (7)	Teréz		SV5276	
Moderate resistance (5)	Villaris, Malverina	GM318-57	Felicia, Merzling	
Moderate susceptibility (3)	Suzy, Moldova, E338, Nero, V25/20, Ecs18, Ecs20, MM31, Ecs26, Ecs28, E313, Ecs44, E411, E346,Viktória gyöngye(CSFT 195)	GM78-10-8		
High-level susceptibility (1)	SV20365, Ecs16, Ecs22, Zalán, Bianca, Ecs38, Ecs46, Ecs48, Isaura, E345, E352, Esther, E350, E344, MM2, MM21, MM44, MM42, MM57	Gm322-58 MM32		

Among *Vitis amurensis* x *V. vinifera* hybrids 2 accessions of the F2 generation ('5-11-2', '5-10-6') showed high-level resistance, a third accession ('5-11-6') had medium level resistance, while the other accessions were classified susceptible or highly susceptible.

M. rotundifolia x *V. vinifera* x *V. amurensis* x French-American derived complex hybrids which are the latest candidates for state approval in the breeding program, and which possess high-level mildews resistances and excellent fruit quality, were equally highly susceptible to black rot disease.

The *V. vinifera* varieties originating from Georgian and Uzbek germplasm were in majority susceptible or highly susceptible to black rot. Two varieties, 'Muradouli' and 'Ojaleshi', had moderate resistance to the disease.

Varieties, showing moderate to high-level resistance in leaf tests in 2014, were selected in 2015 for screening for berry resistance to establish the relation between the two parameters. Table 1 shows the scores for clusters in comparison with scores for leaves. Symptoms recorded on leaves are not always in agreement with symptoms observed on berries. 'Csillám' was the only variety for which berries were symptomless, and it was put into a category named Symptomlessly Resistant (SR). In case of 'Csillám' leaf and berry resistance were coherently symptomless. In case of five accessions, 'Seyval', 'Merzling', '5-11-2', '5-10-6' and '5-11-6', spots appeared on the berry, but symptoms were uncharacteristic for black rot. Small, black, superficial, scabby lesions appeared on the berry skin which did not cause the decay of the entire berry as occurs on sensitive varieties. Few pycnidia developed in the spots. As the berry grew in size the scabs detached from the berry in some cases, leaving intact skin tissue behind. Accessions with this symptom were called Resistant (R). 'Chancellor' showed symptomless leaf resistance at each test, however 50 to 80 % of the berries turned into mummies. Despite high-level leaf resistance in 'Teréz' and 'GM318-57', 80 to 90 % of berries in the clusters of these varieties were destroyed by black rot, thus they were categorized Highly Sensitive (HS). 'Malverina' had moderate level of leaf resistance coupled with severe berry symptoms. The control varieties ('Furmint' and 'Bianca') showed almost complete yield loss.

DISCUSSION

Wild grape species that are indigenous at the North-American habitat of *Guignardia bidwellii* are certain resistance sources to black rot due to the co-evolution process between host and pathogen. Among North-American *Vitis* species, *Vitis cinerea, Vitis rupestris, Vitis lincecumii* are crucial sources of resistance, but having a wider range of sources would be of major importance. For breeding purposes it is more convenient to use donors, that have undergone selection for fruit quality than using wild accessions with undesirable traits. This way more backcross generations and a long period of work can be spared in breeding black rot resistant varieties. French-American hybrids merge the genomes of numerous North-American grape species, besides these are cultivated vines with acceptable fruit quality, so they represent a potential group for finding appropriate resistance sources.

In spite of standardized inoculation conditions and the similar development stages of plants, black rot infections often resulted in differing results for the same genotype. It is even more apparent for evaluations accomplished by different researchers: resistance levels of some varieties showed inconsistencies between our evaluations and results gained by other research groups. 'Villard blanc' ('Seyve-Villard 12375') was described susceptible in the first publications (Hartman and Hershman, 1988). Jabco et al. (1985) evaluated this variety to be tolerant, and Rex (2012) found it resistant. In our experiments, typical symptoms appeared in all four tests, though with different severity. As a result, we could not determine its class, and described 'Villard blanc' as moderately sensitive/moderately resistant.

Further discrepancies were detected in the case of 'Chancellor', which showed symptomless resistance in two successive years in 4 tests. Jabco et al. (1985) found the same result, while other researchers evaluated 'Chancellor' susceptible (Ries, 1999; Hartman and Hershman, 1988) or moderately susceptible (Ellis and Nita, 2004).

Tests on 'Seyval blanc' also produced inconsistent results. Based on field observations, Ellis and Nita (2004) found it moderately susceptible, while Hartman and Hershman (1988) described it susceptible. In artificial tests, it proved to be moderately (Jabco, 1985) or highly resistant (Rex, 2012). In only one test did 'Seyval blanc' produce small lesions without pycnidia, in three other tests it appeared to be symptomless.

Based on information in breeders' descriptions 'Csillám' was regarded as an offspring of 'Villard blanc'. Therefore the symptomless resistance of 'Csillám', an attribute which is not present in the putative parents, turned up as a surprise. High-level resistance to black rot in case of this variety was also confirmed by Kellner et al. (2014). The question arises whether it is possible, that 'Csillám' is a real progeny of 'Villard blanc'. Molecular analysis carried out at the Szent István University-Gödöllő, revealed the parentage of 'Csillám'. 40 SSR loci verified the real parents as 'Kékfrankos' ('Blaufränkisch') and 'Roi des noirs' ('Seibel 4643'). (Kiss et al., 2017;) Presumably, 'Seibel 4643' passed down the resistance trait to 'Csillám'. 'Seibel 4643' originates from the second backcross generation of 'Jaeger 70' (*V. lincecumii x V. rupestris*), so 'Csillám' can be regarded as a BC2 hybrid of these species. In the same experiment 'Teréz', another Hungarian-bred interspecific variety, was proved to be the real offspring of its documented parents 'Villard blanc' and 'Olimpia'. In the case of this variety the origin of this high-level black rot resistance in leaves is not clear. Phenotypic evaluations for black rot resistance were carried out at more time points in each experiment, which showed that symptoms appear at different days post-inoculation (dpi), for various varieties. Symptoms appeared later than 21 dpi on 'Felicia' and 'Merzling' which is the commonly used time-period for evaluation. This slow symptom development could be the reason that 'Felicia' and 'Merzling' were classified as highly resistant in other experiments (Rex, 2012). The varying length of the period after which symptoms appeared, do not influence the severity of the disease, and therefore we evaluated disease severity based on the fully developed symptoms independent of time passed since inoculation.

The scabby lesions symptom on the berry skin was not described for the *Vitis* genus before, but it is known to be typical for muscadine grapes (Wilcox et al., 2015). Varieties possessing this kind of berry resistance are of high economic importance, because even if slight leaf symptoms appear under high disease-pressure conditions, no crop loss will occur in the field.

Screening grape germplasm for black rot resistance, that according to our present knowledge have not co-evolved with this pathogen may also lead to success. The example of finding valuable resistance sources against mildews in unrelated germplasm, suggested to make a quest also for black rot resistance among Central-Asian and Chinese grapes. The evaluation of a large set of Georgian *V. vinifera* varieties supported our assumption. Most of the accessions were highly susceptible, but 2 varieties showed tolerance.

V. amurensis x *V. vinifera* hybrids represented another group of accessions, which could not have co-evolved with *G. bidwellii*, according to the latest idea of grape fungal pathogens incidence on continents. Surprisingly among the 11 F2 hybrids 3 showed medium to high-level leaf resistance and the same peeling spot symptom on the berries, that was observed only on two other Seyve-Villard hybrids. Herbert and Barrett (1953) described *V. amurensis* very susceptible based on the evaluation of two clones, and presumed that this species is uniformly susceptible.

The information about other non-co-evolved resistances in Asian grape species against powdery mildew and downy mildew have been expanding in the last decades. Powdery mildew resistant grapevine varieties have been discovered recently in Central Asia (Hoffmann et al., 2008; Coleman et al., 2009). Till now 10 Vitis vinifera varieties and 2 *Vitis sylvestris* accessions are proved to possess the same monogenic loci responsible for high-level powdery mildew resistance. Molecular data suggest, that cultivated varieties inherited this trait from the wild progenitor (*V. sylvestris*) of grape. (Riaz et al., 2013). Numerous wild grape species native to China were screened for downy and powdery mildew resistance, and several species proved to have accessions resistant to one or both diseases. Variation of resistance was present within a species among genotypes without correlation to the geographic distribution (Wan et al., 2007). Genetic determinism and phenotypic aspects of resistance were studied in the case of some Chinese species, but the understanding of the whole process is failing yet. However it is conceived that different resistance mechanisms play a role in the case of species, investigated till now. Defense mechanisms in Vitis romanetii and Vitis piasezkii against powdery mildew proved to be even stronger and quicker in activation than in other North-American grape species that have developed under co-evolution with the pathogens (Ramming et al., 2011; Pap et al., 2016). These results support the conviction that it is worth screening additional germplasm from Asia for black rot, too.

CONCLUSIONS

Symptom development of black rot greatly depends on the growth intensity of the tissue being infected. Young leaves on the shoot tip often stay symptomless in case of failing growth. Thus, besides the well-defined climatic conditions that determine the success of black rot infection, the stage of plant tissue is also of great importance at the artificial testings.

'Roi des noirs' ('Seibel 4643') was not included in our evaluations for black rot resistance, however 'Csillám', the only accession with symptomless resistance is offspring of this variety. Testing of 'Roi des noirs' and its other descendants is the next logical step to find further accessions which could be potential genetic resources for black rot resistance.

So far, 'Csillám', 'Seyval blanc' and *V. amurensis* F2 hybrids '5-11-2', '5-10-6' have proved to possess the highest level of resistance against black rot.

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