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Crossability of wild potato species and advanced breeding lines resistant to late blight

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SUMMARY
In order to reduce the damage from *Phytophthora infestans* the use of new resistant cultivars is one of the viable approaches for late blight management. In crosses made at SLU, Alnarp (Sweden), wild and cultivated species, *Solanum tuberosum* L. cultivars and breeding lines possessing leaf or tuber resistance were used. The main aim of our study was to examine the crossability of several potato species and *S. tuberosum* breeding lines/cultivars resistant to *P. infestans*. This paper reports the results of crosses of two hexaploid (6x, 4EBN), one tetraploid (4x, 2n=48) and one diploid (2x, 2n=24) potato species with *S. tuberosum* cultivars and breeding lines (4x, 2n=48). Hybrid seeds were obtained in crosses where accessions of the Mexican species *S. demissum* Lindl. and *S. guerreroense* Corr. were pollinated with the Swedish cultivar Superb and *S. tuberosum* L. subsp. *andigenum* (Juz. and Bukasov) Hawkes. Hybrid seeds were also obtained from a direct cross between a selection from the cultivar Aurora (female) and the Bolivian diploid species *S. ruiz-ceballosii* Card. In several crosses within *S. tuberosum* genotypes the cultivars Ora, Kiva and Superb were found as effective pollinators. The crossability of the cultivars Kiva and Ora as well as of *S. tuberosum* breeding lines depended much on the choice of the other parental accession.

KEYWORDS
Phytophthora infestans, potato late blight, resistance, hybrid seedlings, wild species, breeding lines

INTRODUCTION

In the European Union almost 6 Mha of potatoes are grown, representing a value of close to €6,000,000,000. Late blight caused by *Phytophthora infestans* causes estimated annual losses (costs of control and damage) of more than €1,000,000,000 (Haverkort *et al.* 2008). Breeding achievements using large-scale approaches have not been able to significantly decrease yield losses caused by late blight. The most effective and environmentally friendly way to defeat *P. infestans* is by incorporation of resistance genes from new sources like wild potato species and advanced breeding lines.
Solanum demissum has been actively used in breeding programs targeting the development of cultivars resistant to *P. infestans*. Evaluation of diverse potato germplasm has been conducted already during nearly one century. Resistance genes of many species were successfully transferred to cultivated *S.*

PPO-Special Report no. 14 (2010), 209 - 212

tuberosum. In laboratory tests for resistance to *P. infestans* in accessions of wild potato species done in the late 1990s, extreme resistance was also found in plants of the Mexican species *S. guerreroense* (Zoteyeva, 1999). This species is phylogenetically close to *S. demissum*. In the same evaluation, an accession of *S. ruiz-ceballosii* (VIR-7370) was identified as highly resistant in both leaves and tubers (Zoteyeva, 1999, Zoteyeva *et al.*, 2004).

In the 1970s potato breeding with emphasis on vertical resistance was replaced with breeding for horizontal resistance (Wastie, 1991). Several authors have examined the relationship between race specific resistance and field resistance in potatoes and have found evidence for a beneficial effect of R-genes. There is also evidence that defeated R-genes may contribute to late blight resistance and a combination of R-genes and high levels of field resistance is therefore a desirable goal (Steward *et al.*, 2003). Another application of long-term resistance may be to make the optimal selection of R genes due to results of monitoring the *P. infestans* population for each potato growing area.

The main aim for the present study is to examine the crossability of wild species and breeding lines resistant to *P. infestans* and to obtain hybrids combining different types of resistance via inter-specific crosses.

Advanced breeding lines from the collection at the plant breeding program at SLU, Sweden, are promising parental material combining resistance to *P. infestans* and reasonably good consumer qualities. These lines were evaluated in the field under severe infection pressure as well as in laboratory tests.

Solanum demissum Lindl. and the phylogenetically close species *S. guerreroense* Corr. were used as genetic sources of extreme resistance to *P. infestans.* Accessions that showed a hypersensitivity reaction (HR) were identified in leaflet inoculation tests performed earlier. These accessions were pollinated with pollen of cultivar (cv.) Superb and *S. tuberosum* subsp. *andigena* Hawkes respectively. One accession from the species *S. ruiz-ceballosii* Card. (VIR-7370) was identified as highly resistant in both leaves and tubers (Zoteyeva, 1999). Plants from this accession were hybridized with selections of the cv. Aurora.

MATERIALS AND METHODS

Pathogen material

In the evaluation of resistance of accessions from different wild species, two Polish *P. infestans* isolates from the collection of pathogens of IHAR-Mlochow Research Center (Poland) were used. The concentration of the inoculum comprised 75 sporangia/mm^3. The resistance of Swedish cultivars and *S. tuberosum* breeding lines were tested using an aggressive Swedish isolate. The concentration of the inoculum comprised 25 000 zoospores/ml. Tests were performed on detached leaflets and decapitated tubers.

Plant material

Hybridizations were performed on cut branches in the summer of 2009. The branches were kept in jars with water in a greenhouse. Plant material used in the crosses is represented in Table 1.

Table 1. *Parental accessions used in the crosses. Levels of resistance to Phytophthora infestans in leaves and tubers and consumer qualities.*

Parental accessions	Resistance to *Phytophthora infestans**)		Consumer qualities
	leaf	tuber	
Kiva	S*	R	Good
Ora	M	R	Good
Rosamunda**	not tested	R	Good
Superb	S	M	Very high
08-9-Aurora	R	M	High
05-A3 adg	M	R	Satisfying
93-1015	ER	S	Satisfying
04-2081	M	R	Satisfying
04-2662	R	R	Satisfying
04-3262	M	R	Satisfying
S. demissum L.	R	M	Wild type
S. guerreroense Corr.	R	M	Wild type
S. ruiz-ceballosii Card.	R	R	Wild type

**Levels of resistance, ER - extreme, R - high, M - medium, S – susceptible*
***Immune to wart, resistant to Globodera rost, race 5*

RESULTS AND DISCUSSION

In crosses performed the Swedish cultivar Superb was found as an effective pollinator and yielded a high number of seeds in all crosses performed. A high number of seeds were also obtained in one single cross between the cultivar Rosamunda (female) and the breeding line 04-3262 (Table 2). In reciprocal crosses between cv. Ora and the breeding line 04-3262 higher number of seeds was found when Ora was used as female parent. In combination where Ora was used as pollinator the number of seeds was two times lower. Two breeding lines, 04-2081 and 04-2662 (both female parents), were crossed with the cultivars Kiva and Ora. With both breeding lines seed production was lower in combinations with Ora compared to Kiva. In opposite, in combinations of these two cultivars with the breeding line 93-1015, used also as female parent, number of seeds was higher in crosses with Ora compared to with Kiva. Obtained results showed that seed formation in crosses using the same cultivars or breeding lines depends much on the other parental accession.

Table 2. *Results from crosses performed with accessions of wild species and S. tuberosum genotypes.*

Successful crosses	Nr of seeds per fruit	Unsuccessful crosses
04-2081 × Kiva	**110,3**	04-2081 × 04-3262
04-2081 × Ora	**80**	04-3262 × 04-2662
04-2662 × Kiva	**65,7**	04-2662 × 04-3262
04-2662 × Ora	**47,5**	04-2662 × Superb
04-2662 × Superb	**102**	93-1015 × 04-3262
04-3262 × Ora	**68,8**	Kiva × 04-3262
Ora × 04-3262	**120**	Ora × 04-3262
Ora × Superb	**144,7**	Kiva × 08-A3 adg
Rosamunda × 04-3262	**170**	08-A3 adg × 04-3262
93-1015 × Kiva	**94**	08-A3 adg × 04-2662

Successful crosses	Nr of seeds per fruit	Unsuccessful crosses
93-1015 × Ora	**120**	08-A3 adg × Ora
08-A3 adg* × Kiva	**102**	grr VIR-18407 × 05-A3 adg
08-A3 adg × Superb	**206**	dms VIR-3355 × Superb
08- 9-Aurora × rcb* VIR-7370	**29**	
dms* VIR-3355 × Superb	**45,5**	
grr* VIR-18407 × Superb	**40**	
dms VIR-3355 × 05-A3-adg	**33,7**	
grr VIR-18407 × 05-A3- adg	**54**	

**Abbreviations of Solanum species: adg – S. tuberosum subsp. andigenum, rcb – S. ruiz-ceballosii, dms – S. demissum, grr – S. guerreroense.*

Crosses done on different genotypes of *S. guerreroense* pollinated with different sets of pollen from *S. tuberosum subsp. andigenum* resulted in three unsuccessful (nine pollinated flowers) and three successful (seven pollinated flowers) crosses. One successful and one unsuccessful cross were noted in combinations where *S. guerreroense* and *S. demissum* were pollinated with *S. tuberosum* (cv. Superb). Obtained results confirmed the data obtained by Hermsen and E. Sawicka (1979) who found differences in ability to cross among different genotypes within species.
Plants of the diploid species *S. ruiz-ceballosii* were used as male parents in crosses with a selection of cv. Aurora. Regardless the difference in chromosome number of parental accessions this combination resulted in seed production (Table 2).

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