



DEVELOPMENT OF A DArT MICROARRAY FOR COMPARATIVE STRUCTURAL GENOMICS AND MAPPING OF AGRICULTURALLY SIGNIFICANT GENES IN WILD POTATO SPECIES

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Introduction

For potato breeding programs, wild species are a valuable source of agriculturally significant genes. We specialized in the *Stellata* series encompassing 20 wild potato species sexually incompatible with cultivated potato. These are rich sources of disease resistance.



Series*	Species	EBN	Reported Disease Resistance
I. Miroblomia	<i>S. citrum</i>	7	Early blight, PMV, Verticillium
	<i>S. mirabilis</i>	7	
II. Bulbocastano	<i>S. bulbocastanum</i>	1	Blackleg, Bacterial wilt, Early blight, Late blight, PLRV, PMV, Ring rot, Verticillium, Wart
	<i>S. cardiophyllum</i>	1	Late blight, Ring rot, Verticillium, Wart
	<i>S. jamaei</i>	1	Blackleg, PLRV, PVY, Ring rot, Verticillium
III. Pinnatisecta	<i>S. pinnatisectum</i>	1	Black leg, Early blight, Late blight, PLRV, Ring rot
	<i>S. chacoense</i>	1	Blackleg, Early blight, Ring rot, Verticillium
	<i>S. andrei</i>	1	Late blight, Ring rot
	<i>S. inflatum</i>	1	Late blight, PLRV, Ring rot, Verticillium
	<i>S. jactator</i>	1	Ring rot, Verticillium wilt
IV. Polyadenia	<i>S. polyadenium</i>	1	Blackleg, Late blight, PLRV, PVX, PMV, Ring rot, Verticillium
	<i>S. commersonii</i>	1	Bacterial wilt, Blackleg, Early blight, Ring rot, Verticillium
V. Comersoniana	<i>S. commersonii</i>	1	Bacterial wilt, PMV, PVY, Rhizoctonia, Wart
VI. Circaeifolia	<i>S. circaeifolium</i>	1	Fusarium wilt, Rhizoctonia, Verticillium, Wart
VII. Ugricauda	<i>S. agricauda</i>	1	
VIII. Obovata	<i>S. obovata</i>	7	
IX. Yunguesia	<i>S. yunguesia</i>	2	PVX, PVY, PVX, PLRV, Verticillium
	<i>S. chacoense</i>	2	PLRV, Verticillium
	<i>S. argentea</i>	2	Verticillium
	<i>S. yunguesia</i>	2	
	<i>S. yunguesia</i>	2	

Tab. 1. *Classification scheme for series Stellata of Hawkes (1990) with modifications after Rodriguez and Spooner (1997), Lara-Cabrera and Spooner (2004), and Spooner et al. (2004).
*Resistance data from personal experience, published accounts (Austin et al. 1993; Helgeson et al. 1998; Micheletto et al. 2000; Naeess et al. 2000; Song et al. 2003; Vossen et al. 2003; Vossen et al. 2005), and USDA Potato Genebank website. Highlighted species are those used for DArT array construction.

To improve access to noteworthy traits from these wild potato species, we have initiated the development of a DArT (Diversity Array Technology, Fig. 1) microarray for comparative structural genomic studies.

Materials & Methods

Five different wild species were tested for intra-specific crossability (Table1). These species were also used for DArT microarray construction by A. Kilian (DArT Pty Ltd, Canberra, Australia).

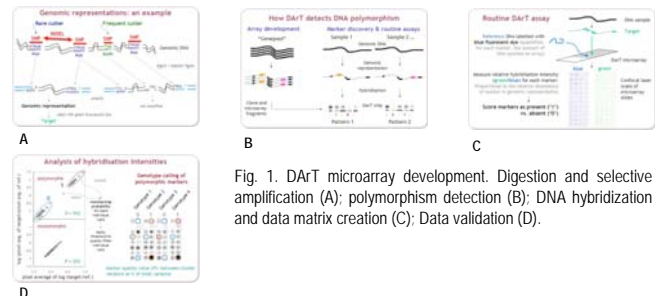


Fig. 1. DArT microarray development. Digestion and selective amplification (A); polymorphism detection (B); DNA hybridization and data matrix creation (C); Data validation (D).

Due to worldwide interest in resistance to late blight, different accessions of *S. commersonii*, *S. chacoense*, *S. polyadenium* and *S. pinnatisectum* were tested for foliar resistance to *Phytophthora infestans* US8 in field trials in Minnesota (fig. 3).

Results

F₁ mapping populations have been generated for each species used in array construction, except *S. polyadenium*. Validation of a first generation DArT array was done via phylogenetic analysis of these five species, *S. cardiophyllum* and *S. circaeifolium*.

Cluster analysis of DArT data (Fig. 2A) identified seven species-specific clusters, as expected. Comparison with previously reported phylogenies based on other molecular marker systems (Spooner et al., 2004; Fig. 2B) revealed general concordance. Thus, DArT data are reliable.

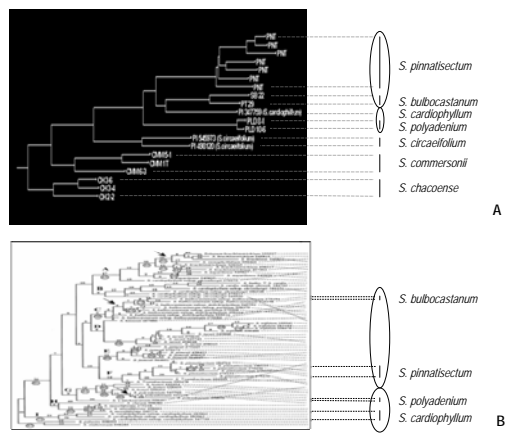


Fig. 2. Phylogenetic analyses of wild potato species using DArT markers (A) and AFLP markers (Spooner et al., 2004).

Late blight resistance screening (Table 2) demonstrated that many genotypes are resistant to *P. infestans* US8.

Tab. 2. Results of foliar resistance to *Phytophthora infestans* US8.

Species	Number of accessions tested	Resistant #	Susceptible #
CMM	63	13	50
CHC	133	11	122
PLD	2	2	0
PNT	2	2	0
Tot.	198	28	172

CMM: *S. commersonii*; CHC: *S. chacoense*; PLD: *S. polyadenium*; PNT: *S. pinnatisectum*. Disease index ranging from 1 to 9. Resistant ≤ 4.



Fig. 3. Field test for *P. infestans* resistance on *S. commersonii*, *S. chacoense*, *S. polyadenium*, *S. pinnatisectum*.

Future research

Stellata species will be systematically screened for new resistance traits. Ongoing molecular investigations focus on:

- *Stellata* series comparative mapping;
- wild potato-tomato genome comparison;
- mapping and cloning interesting resistance genes.

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