

# DEVELOPMENT OF INTEGRATED *Solanum bulbocastanum* GENETIC AND PHYSICAL MAPS AS A COMMUNAL RESOURCE FOR MAPPING AND ISOLATION OF R GENES

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Potato Pathology and  
Potato Pathology and Genomics  
Genomics



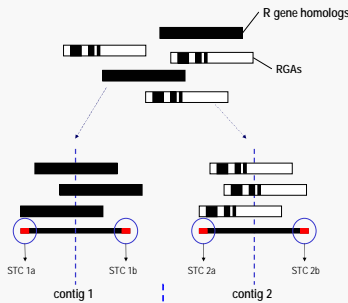
This poster can be found at: <http://ppg.coafes.umn.edu/>

## Introduction

We are developing integrated resources of genetic and resistance (R) gene physical maps for the primitive potatoes using *S. bulbocastanum* as a model. This resource would enable efficient and rapid R gene mapping and isolation. The steps of this project are diagrammed below.

1. Isolating BACs corresponding to R gene fragments

2. Assembling recovered BACs into contigs



3. Markers derived from BAC end sequence are used to integrate genetic and physical (BAC contig) maps. Markers may be subsequently applied to phenotypically segregating populations of other *Solanum* species, allowing users to genetically map resistance traits and immediately identify candidate genes

## 1

Toward this goal we isolated an extensive collection of R gene homologs, designing PCR primers from 13 previously cloned R genes from both tomato and potato (tab.1) and we recovered RGAs by degenerate PCR primers (see poster P095).

Tab.1 *Solanum* R genes used for BAC hybridization

R gene	Donor Species	Pathogen	Structural Class
Cf-9	<i>S. pimpinellifolium</i>	<i>Cladosporium fulvum</i>	LRR-Transmembrane
Gpa2	<i>S. tuberosum ssp. andigena</i>	<i>Globodera pallida</i>	NBS-LRR
Gro1.2	Cultivated potato	<i>Globodera rostochiensis</i>	NBS-LRR
Hero	Tomato	Potato cyst nematode	NBS-LRR
I2	Tomato	<i>Fusarium oxysporum f.sp. lycopersici</i>	NBS-LRR
Mi-1	Tomato	Root knot nematode, aphids, <i>P. infestans</i> *	NBS-LRR
Prf	Tomato	<i>Pseudomonas syringae pv. tomat</i>	NBS-LRR
Pto	Tomato	<i>Pseudomonas syringae pv. tomat</i>	Rolein Kinase
R1	<i>S. demissum</i>	<i>Phytophthora infestans</i>	NBS-LRR
RB	<i>S. bulbocastanum</i>	<i>Phytophthora infestans</i>	NBS-LRR
Rx2	<i>S. acule</i>	Potato virus X	NBS-LRR
Sw5-d	Tomato	Tomato spotted wilt virus	NBS-LRR
Ve	<i>S.aethiopicum</i>	<i>Verticillium spp.</i>	Surface receptor

\* (homolog Rpi-b1b2, Van der Vossen et al, 1<sup>st</sup> Solanaceae Genome Workshop, 2004)



We have used fragments from several R gene homologs including the late blight resistance genes R1, RB, and Mi-1 as hybridization probes to screen a 5x portion of our *S. bulbocastanum* BAC library. Seven to ~ 50 clones were identified for each probe, suggesting homologs of many R genes map to multiple genome locations (fig. 1).

Fig.1 A high density blot of *S. bulbocastanum* BAC library (blot represent 2.4 genome equivalent) hybridized with Mi-1 gene homolog.

## 2

BACs are assembled into contigs by BAC end hybridization (Bradeen, MMG 2003, 269: 603-611).

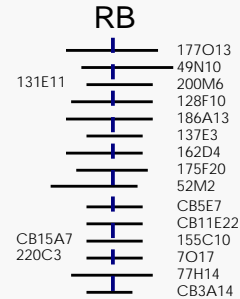


Fig 2. BAC contig of the RB region on chromosome 8 assembled via BAC end hybridization

In addition to chromosome 8 RB BACs, we have recovered >13 clones with homology to RB, suggesting RB homologs map to at least 2 additional locations in the *S. bulbocastanum* genome. Consistently, RB homologs map to 3 genome locations in tomato, including chromosome 8 (Pan, 2000). Mapping efforts in *S. bulbocastanum* are ongoing.

## 3

The genus *Solanum* includes important cultivated species including potato, tomato, and eggplant. Phylogenetic relationships are well established (fig. 3) and overall genome organization is known to be similar.

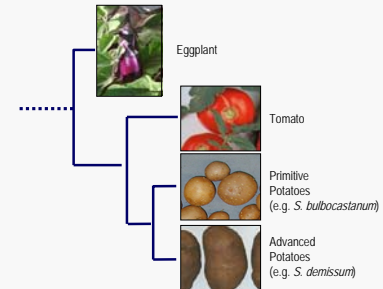


Fig.3 Phylogeny of the genus *Solanum* (Adapted from D. Spooner, USDA-ARS)

Potentially, BAC end sequence from *S. bulbocastanum* R gene contigs can be used to map phenotypic traits in other *Solanum* species. Here we test two BAC derived PCR primer pairs on genotypes of primitive potato (*S. bulbocastanum*), advanced potato (*S. demissum*), tomato, and eggplant (fig. 4).

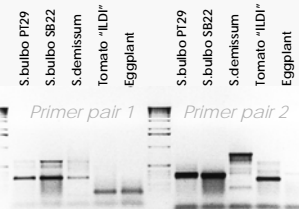


Fig.4 Amplification from two primer pairs designed from Mi-1 BAC clones amplify fragments from additional *Solanum* species. Amplification of intended target sequences might be most reliable for species closely related to *S. bulbocastanum* and primers must be carefully tested over a phylogenetic range prior to use. Sequence analysis of generated fragments is ongoing.

The presence of bands for each species tested and the well established synteny within the genus *Solanum*, suggest *S. bulbocastanum* BAC end sequence associated with putative R gene might be useful for mapping throughout the genus.