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## ASSESSMENT OF THE RESISTANCE CONFERRED BY THE *bc-1* ALLELES TO *Bean common mosaic necrosis virus*

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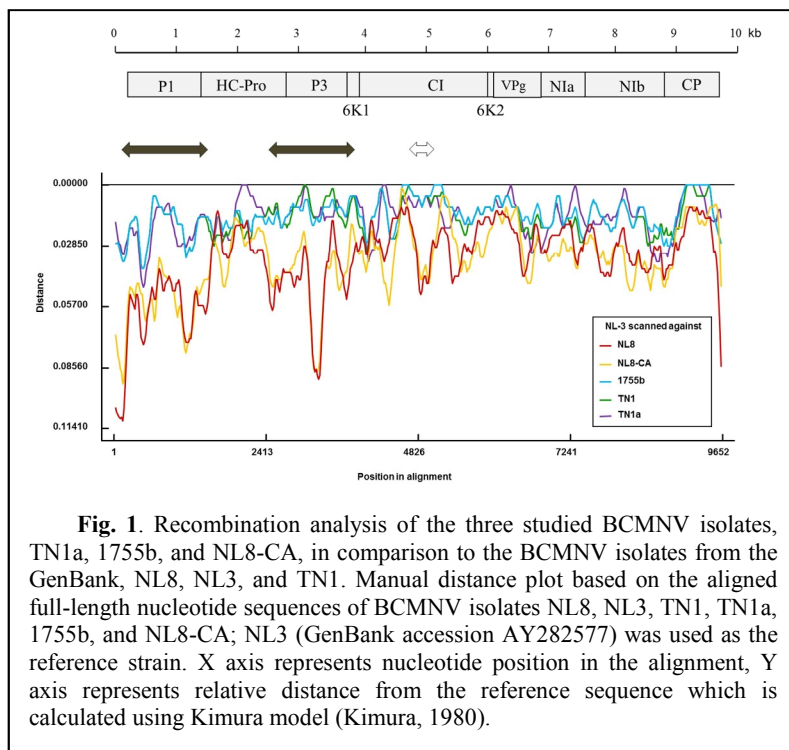
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**INTRODUCTION:** *Bean common mosaic necrosis virus* (BCMNV) is a potyvirus comprising several strains classified into two pathogroups according to the interactions with six recessive resistance alleles in common bean (1). These pathogroups (PGs), numbered III and VI, are defined by the ability (PG-VI) or inability (PG-III) of a BCMNV isolate to replicate in bean differential lines carrying *bc-1* or *bc-1<sup>2</sup>* resistance alleles. The biological and molecular basis for this differential response of BCMNV isolates to the presence of *bc-1* alleles is not known. Conversely, the genetic determinants involved in interactions of BCMNV strains with *bc-1* resistance alleles have not yet been identified either. We performed a complete biological and molecular study of three isolates of BCMNV belonging to PG-III and VI, collected in California and in Oregon. Particular attention was paid to BCMNV isolates' performance in common bean lines from host groups 2, 3, and 9, harboring *bc-1* and *bc-1<sup>2</sup>* alleles. The data obtained suggest that the *bc-1* alleles restricted systemic movement of PG-III isolates of BCMNV, while cell-to-cell movement of the virus in inoculated leaves did not seem to be affected.

**MATERIALS AND METHODS:** The reference BCMNV isolate TN1 used in this work was described previously (2). BCMNV isolate NL8-CA was collected in 2015 near Davis, CA, as a field sample from an heirloom common bean cultivar 'Zuni Gold' displaying mosaic and stunting. The TN1a isolate of BCMNV was found in a sample of common bean exhibiting mosaic found in Corvallis, OR, in 2015. BCMNV isolate 1755b was initially identified in a field sample 91-1755 collected in Willamette Valley, OR, in 2013 (3); this original sample contained mixed infection comprising a 1755a isolate of BCMV and a 1755b isolate of BCMNV. The 1755b isolate of BCMNV was biologically separated from BCMV by passaging it through common bean cultivar Redlands Greenleaf B, non-permissive for the PG-VIII isolate 1755a of BCMV (3). All virus isolates were propagated in the bean cultivar 'Dubbele Witte' using mechanical inoculation and then maintained under greenhouse conditions. The whole-genome cloning strategy, sequencing, and sequence analysis for BCMNV isolates 1755b, TN1a, and NL8-CA were conducted as described previously (2).

**RESULTS:** Of the three BCMNV isolates subjected to the biological typing on bean indicators, one (NL8-CA) was unable to systemically infect cultivars 'Redlands Greenleaf B' (HG-2) and 'Redlands Greenleaf C' (HG-3), and hence was typed as belonging to pathogroup III, while the other two (TN1a and 1755b) infected cultivars 'Redlands Greenleaf B' and 'Redlands Greenleaf C' systemically, and were typed as belonging to pathogroup VI. The two latter isolates, TN1a and 1755b had the pathogenicity profile identical to the control BCMNV isolate TN1 belonging to PG-VI. Isolate NL8-CA (PG-III) induced only local necrosis on inoculated leaves in cultivars 'Top Crop' and 'Jubila' harboring *I* gene protected with the *bc-1* allele, while isolates TN1, TN1a, and 1755b (all PG-VI) induced rapid whole plant necrosis (WPN) in 'Top Crop' 7-14 days

post-inoculation, and severe systemic necrosis, but not WPN, in ‘Jubila’ 3-5 weeks post-inoculation. In cultivars ‘Redland Greenleaf C’ expressing *bc-1* and ‘Redland Greenleaf B’ expressing *bc-1<sup>2</sup>* alleles, isolate NL8-CA was able to systemically infect only a small proportion of upper, uninoculated leaves, below 13% and below 3%, respectively. The whole genomes of BCMNV isolates NL8-CA, TN1a, and 1755b were cloned and sequenced, using the approach described previously (2). Upon sequence assembly, NL8-CA was found to be 9,627-nt long, excluding the poly (A). Based on conceptual translation, the BCMNV NL8-CA genome encoded a single polyprotein of 3,071 aa. Both TN1a and 1755b genomes were found to be 9,628-nt long, excluding the poly (A), and both genomes encoded a single polyprotein of the same size as NL8-CA (3,071 aa).



**CONCLUSIONS:** The whole genomes of isolates 1755b, TN1a, and NL8-CA were sequenced and sequence analysis revealed that despite the overall high nucleotide sequence identity between PG-III and PG-VI isolates (ca. 96%), two areas of the BCMNV genome in the P1/HC-Pro and HC-Pro/P3 cistrons appeared to be more divergent between these two pathotypes of BCMNV (Fig. 1). The data obtained suggest that the phenotypic differences among PG-III and PG-VI isolates of BCMNV in common bean cultivars from host resistance groups 2, 3, and 9, carrying *bc-1* alleles, were related to the impaired systemic movement of

the PG-III isolates to the upper, uninoculated leaves, and also suggest a role of the recessive *bc-1* gene in interfering with systemic spread of BCMNV.

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