Apparent Immunity and Tolerance to Tomato Big Bud Disease in *Lycopersicon peruvianum* and in Two of Its Tomato Hybrids

P. E. THOMAS, Research Plant Pathologist, Agricultural Research Service, U.S. Department of Agriculture, Irrigated Agriculture Research and Extension Center, Route 2, Box 2953A, Prosser, WA 99350, and SHER HASSAN, Professor and Chairman, Plant Pathology Department, NWFP Agricultural University, Peshawar, Pakistan

**ABSTRACT**


All plants in a seed lot of *Lycopersicon peruvianum*, U.S. Department of Agriculture Plant Introduction (PI) No. 128655, and in seed lots of two F2 hybrid progenies of PI 128655 × tomato either did not become infected (apparent immunity) or were infected without symptoms (tolerance) after graft inoculation using tomato tissue infected with tomato big bud disease. The same germ plasm was previously shown to contain the same expressions of resistance and tolerance against three phloem-limited viruses—beet curly top, tomato yellow top, and potato leafroll.

A number of aster yellows type diseases of tomato (*Lycopersicon esculentum* Mill.) and potato (*Solanum tuberosum* L.) reported worldwide under a variety of names are believed to be caused by members of a group of mycoplasmalike organisms (MLOs) (3,9,12,21). These include tomato big bud diseases reported in the United States (5,6,8), Australia (1,13), and the Middle East (21); the stolbur diseases of tomato and potato in Europe and Russia (4,12,19); mal azul in Portugal (2); potato purple top wilt in India and Australia (1,9,13); and potato haywire in the United States (20).

The exact relationships among the MLOs associated with these diseases are not known (4) but all are transmitted by one or more leafflower species that vary in transmission efficiency for different strains of the pathogens in different areas of the world (4). Transmission from tomato plants with big bud or stolbur symptoms produced the potato syndrome called haywire, purple top wilt, or stolbur in different areas of the world (1,20).

Symptoms of tomato big bud in the western United States are identical to those described in detail by Samuel et al (13) in South Australia. They are characterized by teratological changes, particularly in the flowering structure. Calyx segments on flowers remain united almost to the tips, and the whole calyx enlarges to form a bladderlike structure 2-5 cm in diameter with a toothed opening at the top. Anther and ovary development stop and petals become virecent. Purple coloration develops, particularly along veins of the bladder structures and on the undersides of young leaves. Auxiliary buds proliferate, stems gradually thicken, and the plant finally assumes a tufted, rosette growth habit.

Golino (7) showed that the leafflower-transmitted virencence agent (BLTVA), an MLO, has caused big bud symptoms in experimentally infected tomato. Furthermore, biological and molecular evidence has established that field-collected big bud symptomatic tomatoes from California are infected with BLTVA rather than other MLOs (14,15). Although it is not yet clear what relationship the BLTVA-MLO may have with the MLO that causes the disease in other parts of the world, BLTVA has become a prime suspect as the tomato big bud causal agent in the western United States within the geographic range of its vector, *Cicurifer tenellus* (Baker).

Field infection rates approaching 100% in both tomatoes and potatoes have been reported for stolbur in Europe (4,19), and serious losses to purple top wilt in potato have been reported in Australia (1), but there is no record of serious economic losses to any MLO-induced disease of potato or tomato in the United States.

MLOs are phloem-limited pathogens. We previously discovered apparently complete tolerance and immunity to three phloem-limited viruses (beet curly top virus [BCTV], tomato yellow top virus [TYTV], and potato leafroll virus [PRLV]) in plants of *Lycopersicon peruvianum* (L.) Mill., USDA Plant Introduction (PI) No. 128655, and two selected F2 hybrid progenies of PI 128655 × *L. esculentum* (10,11,16-18). The basis for resistance to the viruses appeared to be related to translocation in the phloem. This study was performed to determine whether PI 128655 was also resistant to a completely different but phloem-limited pathogen, the tomato big bud MLO.

**MATERIALS AND METHODS**

Source of germ plasm, *L. peruvianum*, PI 128655, was collected at Charanilla, Tampaca, Peru, in 1938 by L. H. Blood (from original record of L. H. Blood) and increased by open-pollination at Ogden, UT, and later at Prosser, WA. Hybrid progenies were produced from interspecific crosses between a selected PI 128655 plant with immunity to BCTV (16,17) and tomato cv. Bonnie Best. F1 and F2 generations of the hybrids were increased in the field by open-pollination. Plants with immunity and complete tolerance to BCTV were selected in the F1 generation, and plants with resistance to TYTV were selected in the F2 generation. The F3 hybrid progenies used in this study were derived from individual, open-pollinated F1 plants selected for resistance to TYTV in the preliminary studies (11).

Inoculum source. A tomato plant with big bud symptoms typical of those described by Samuel et al (13) was selected in the field. Cuttings were rooted in a greenhouse, and the resulting plants were used as a source for graft inoculations. A second tomato plant from the field with typical big bud symptoms was used when the initial experiments were repeated. These two MLO lines both hybridized with a DNA probe specific for the BLTVA and did not hybridize with a probe specific for the aster yellows MLO, indicating that the tomato big bud symptomatic tomatoes used as the inoculum source for these experiments were infected by the BLTVA (hybridization assays conducted by Mary Shaw, Department of Plant Pathology, University of California, Davis, CA) (14,15). By ELISA, both sources of inoculum assayed negative for BCTV; beet western yellows virus (BWWV); PRLV; potato viruses A, M, S, X, and Y; and tobacco mosaic virus. Young tomato plants remained symptomless when rub-inoculated with

---

Cooperative investigation of the Agricultural Research Service, U.S. Department of Agriculture, and the Washington State University Agricultural Research Center, Prosser, WA.

College of Agriculture and Home Economics Research Center, Plant Pathology PPNS 0047, Pullman, WA 99164.

Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the USDA and does not imply its approval to the exclusion of other products that may also be suitable.

Accepted for publication 24 July 1991 (submitted for electronic processing).

This article is in the public domain and not copyrightable. It may be freely reprinted with customary crediting of the source, The American Phytopathological Society, 1992.
buffered extracts of the inoculum sources.

**Graft inoculation and indexing.** Test plants and index plants (tomato cv. VF 145) were graft-inoculated by inserting grafts into the sides of the stem and by cleft grafts on excised terminal shoots. The grafts were wrapped with Parafilm, held in a moist chamber for 1 wk, and returned to the greenhouse.

**RESULTS**

Plants of *L. peruvianum*, PI 128655, the two hybrid progeny, and cultivated tomato were graft-inoculated with tissue from cuttings taken from the first field source of tomato big bud. Nine uniform plants from each category with successful graft unions were selected for use in these experiments.

The tomato scions on all of the selected plants grew and continued to express typical big bud symptoms. The inoculated tomato control plants developed typical big bud symptoms within 4–5 wk. However, all of the *L. peruvianum* and hybrid plants remained completely symptomless until discarded 24 wk after inoculation.

At 12 wk and again at 18 wk after inoculation, both the inoculated test and tomato control plants were indexed for the presence of the big bud agent by graft inoculation to healthy tomatoes. All of the tomato control plants, three of nine *L. peruvianum*, five of nine hybrid 1, and four of nine hybrid 2 plants produced positive indices (Table 1). The same indexing results were obtained with the same plants 18 wk after inoculation.

The resistance of *L. peruvianum*, PI 128655, and the two hybrid progenies was tested again 2 yr later. Groups of test plants from the same seed lots were graft-inoculated with a second field isolate of tomato big bud. In the second test, 18 plants in each test category (*L. peruvianum*, hybrid 1, and hybrid 2) and six cultivated tomato control plants with successful grafts were used.

Results were essentially identical to those obtained in the first test. Again, all tomato plants developed typical big bud symptoms within 6 wk, whereas all *L. peruvianum* and hybrid plants remained symptomless. At 12 wk after inoculation, big bud was detected by graft transmission to cultivated tomato in eight of 18 *L. peruvianum*, five of 17 hybrid 1, and seven of 18 hybrid 2, and in all inoculated tomato control plants (Table 1).

The plants from the second test were used in further experimentation to determine whether the big bud agent actually multiplied in the symptomless *L. peruvianum* and hybrid plants or, alternatively, whether it arrived there by passive transport from the infected scions. To do this, the infected graft scions were removed after the 12-wk indexing. The plants were maintained an additional 18 mo in a greenhouse and indexed again.

<table>
<thead>
<tr>
<th>Assessment</th>
<th>Number infected/number inoculateda</th>
</tr>
</thead>
<tbody>
<tr>
<td>L. peruvianum</td>
<td>Hybrid 1</td>
</tr>
<tr>
<td>1</td>
<td>3/9</td>
</tr>
<tr>
<td>2</td>
<td>8/18</td>
</tr>
</tbody>
</table>

*a U.S. Department of Agriculture Plant Introduction Number.

*b Fifth generation open-pollinated progeny of interspecific F1 hybrids selected in the third generation for resistance to BCTV and in the fourth generation for resistance to TYTV. The *L. peruvianum* parent was resistant to BCTV.

*c All infected *L. peruvianum* and hybrid plants remained symptomless, and all tomato plants developed big bud symptoms. Plants were indexed for infection 12 wk after graft inoculation.

During this interval, the plants were pruned several times to force growth of new vegetative stems. Results of the 18-mo index were identical to the 12-wk index. Plants that had indexed positive at 12 wk retained the big bud agent and remained symptomless.

**DISCUSSION**

About 60% of the *L. peruvianum*, PI 128655, and the hybrid progeny germs plant populations assessed for resistance to tomato big bud were apparently immune, because they could not be infected by graft-inoculation methods that routinely infected susceptible plants. The remaining plants were infected with the big bud agent and were completely tolerant under our experimental conditions.

The question of whether the big bud agent failed to move from infected scions into the immune plants or, alternatively, whether it moved into but failed to multiply in the immune plants was not tested.

The expressions of resistance against tomato big bud obtained in the experiments reported here are essentially the same as those obtained with the same germ plant populations when inoculated with BCTV, TYTV, and PLRV (10, 16). Immunity to the viruses appeared to be based on resistance to virus translocation in the phloem. Because the tomato big bud MLO is phloem-limited, it seems possible that a basic mechanism in the phloem could account for resistance to movement of both the viruses and the MLO. However, neither these experiments nor the earlier experiments with viruses provided any information concerning the mechanisms responsible for tolerance (infection without symptoms) of plants that became infected. It seems highly improbable that tolerance to the viruses and to the MLO could be conditioned by the same mechanism.

We are not aware of any previous reports concerning resistance in tomato to big bud or similar MLO diseases. Because the MLO agents that cause tomato big bud, stolbur, potato purple top wilt, haywire, and similar diseases around the world are believed to be closely related, the resistances to big bud reported here may be effective against the other diseases. The reisolation in the hybrid progenies of the resistances identified in *L. peruvianum* suggests that the resistances may be incorporated into tomato by traditional breeding methods. These resistances may also be available for use in potato through advanced gene transfer methods.

**ACKNOWLEDGMENT**

We thank Mary Shaw for conducting the hybridization assays that associated the big bud disease of inoculum sources with the beet leafhopper-transmitted virescence agent.

**LITERATURE CITED**


