

Title: Field Testing Of Resistant Tomato Lines To Control Late Blight And Early Blight In Conventional And Organic Growing Systems.

Lead investigator: Martha Mutscher, Professor, Dept. of Plant Breeding and Genetics,
303 Bradfield Hall, Cornell University, Ithaca, NY 14853
Phone: 607-255-1660, Fax: 605-255-6683, E-mail: mam13@cornell.edu

Team members Tom Zitter, Professor, Dept. of Plant Pathology Cornell University
Majid R. Foolad, Professor, Department of Horticulture, Penn State Univ.
Alan A. MacNab Professor, Dept. of Plant Pathology Penn State Univ.

State(s) involved: NY and PA

Years funded: 3 years, 2005/2006 through 2008/2009

Funding amount: \$122,364

B. Nontechnical Summary.

The recent creation of tomato inbred lines possessing LB resistance and/or EB resistance created the opportunity to restructure integrated strategies for the control of these diseases. Prior work funded by USDA CAR has defined some of the parameters for a coordinated control strategy using the available resistance and control compounds. Integration of genetic resistance and reduce risk fungicides and biological controls are urgently needed. For the NE growers to use this approach, these resistant lines and strategies must also fulfill the needs of the shorter, cooler season characteristic of the NE growing regions. Our goal is to finalize the development of the tomato lines possessing resistance to LB and EB, then to test the best of these lines under traditional and organic growing systems. This work will result in the development of a coordinated total disease control program for NE tomato production using the genetic resistance to provide more reliable disease control and also significantly reduce reliance on chemical control.

C. Introduction.

Tomatoes are an important part of a diverse and balanced diet. One medium fresh tomato (135g) provides 47% RDA of Vitamin C, 22% RDA Vitamin A and 25 calories (135g). Tomatoes are rich in the anti-oxidant lycopene, a compound that protects cells from oxidants that have been linked to cancer.

Although greater production occurs in Fl and Ca, tomato is also an important vegetable crop in NE states, including NY, NJ and PA. The acreage in PA, NJ, and NY was 11,000 A with a value in excess of \$68 million during 2002 (USDA Statistics, 2002).

LB and EB have resulted in numerous epidemics of tomatoes throughout North America, causing considerable loss of yield and crop quality. In the NE in both 2003 and 2004, LB and EB were major problems for tomato production, and heavy losses were sustained. These diseases are a high priority for both NY and PA (see letters of support from NY and PA Vegetable Grower's Associations, and from individual growers and IPM Educators).

Presently, NE growers rely on multiple fungicide applications to achieve the current levels of disease control. New immigrant forms of *P. infestans* have introduced both metalaxyl-resistance and the capability of oospore survival, which will make LB disease management even more

difficult. Chemical control measures can be an effective means of managing LB, and are usually guided by forecasts of blight-favorable weather conditions, but chemical applications are both time- and cost-dependent, and may be of reduced efficacy if weather conditions are particularly conducive for the disease. Similar fungicides used to control LB also control EB. Copper fungicides are used in organic fresh market tomato production; however, copper has been shown to only suppress late blight. Fungicide sprays add a great financial burden to growers. In PA, for example, the average cost of each spray is about \$20 per acre, for a total of \$200 per season (average of 10 sprays per season). That adds up to over \$1,000,000 per year in just one state, which when extrapolated over the entire county, is a tremendous expense to growers, consumers, and to the economy. Recent research has also demonstrated the greater challenge of controlling EB disease caused by the light variant of *A. tomatophila*. All of these factors have generated considerable interest in alternative methods for LB and EB control.

The recent creation of tomato inbred lines possessing LB resistance and/or EB resistance created the opportunity to restructure integrated strategies for the control of these diseases. Prior work funded by USDA CAR has defined some of the parameters for a coordinated control strategy using the available resistance and control compounds. Integration of genetic resistance and reduce risk fungicides and biological controls are urgently needed.

D. Objectives.

Objective 1. Identify appropriate LB and EB resistant lines for use in regional trials and possible release.

The NY breeding program has recently created 32 LB and EB resistant Freshmarket breeding lines, and 17 LB resistant processing tomato lines. The PSU tomato breeding program has also been developing fresh market as well as processing lines with improved EB resistance. Although these are not as advanced as the NY lines, they also should be evaluated for use in the Northeast. This abundance of material is advantageous, since it allows selection of the materials with optimal horticultural characteristics for the NE, but there are more than we could include in regional trials. Therefore a limited number of large trials will compare these lines to determine the best subset of lines for extended regional testing in year 2.

Objective 2. Test EB and LB resistant lines in regional trials to determine their performance and utility in traditional and organic systems.

The trials of the lines selected in year 1 are still ongoing as of 8/28/06. One alteration in the plans is that for the trials in 2007 will expand to focus on hybrids that possess LB/EB resistance, and even better horticultural type through combination of the parent of the hybrids. Seed for the year 3 hybrid trial will be generated in year 2.

Objective 3. Release materials and information to enable NE growers to use the new strategy.

The information generated by this work will be provided to extension personnel and growers through a variety of channels including publication or articles, and the web site <http://vegetablemdonline.ppath.cornell.edu/Home.htm>, presentations at demonstration days held at the research plots, and at stakeholder meetings in both states. The tomato lines will be released through appropriate channels to promote their use. Complete information will be provided to seed companies along with the releases to guide their use of the materials in developing and releasing varieties for use in NE. This objective will be accomplished year 3.

E. Approach. The approach is to use tomato lines or hybrids combining LB and EB resistance, and adaptation to conditions in the North East to determine the best lines and the best use of these lines in control of LB and EB in North East tomato production under standard or organic practices.

F. Progress.

Year 1: In the first field season, all of the lines available were evaluated in trials in NY under standard and organic practice and PA under standard practice to determine the best lines to include in more extensive trials in the second field season. The results of the Early Blight trial confirmed that the Cornell lines were fixed for the early blight resistance. The results of this early blight trial also demonstrated the difference in the degree of disease control the resistance provides on stems vs. on the foliage. The stem ratings on all of the resistant lines are uniformly very low (from 0.0 to 0.2), in contrast to the ratings of the susceptible controls (>4.0). However foliar symptoms, as measured by the % defoliation on the last reading date (19 Sep) or by area under the disease progress curve (AUDPC), were not as well controlled as the stem symptoms. There was also considerable variation among the resistant lines for foliar disease development. This could have been due to differences among these lines for maturity and fruit load/development, rather than any true difference for resistance.

The results of the trials allowed the selection of lines with confirmed resistance to LB and EB and with the best horticultural characteristics in addition to disease resistance. The better inbreds in terms of production, fruit size, as well as fruit characters such as shape, smoothness, blossom end scar have high production levels, though fruit size was smaller than that of the hybrids. The best subset of lines were selected for testing in year 2, and for creating hybrids to be tested in year 3. Therefore this objective has been accomplished

Year 2: A subset of LB/EB lines selected in 2005 was retested in 2006 in expanded trials in NY and PA summer 2006.

The establishment and initial growth of the trial plots in Ithaca was very favorable. Lines selected in 2005 were retested in 2006 in expanded trials. We also tested these lines in disease trials with modest input of chemical controls, in addition to the resistance, to extend control of foliar symptoms of early blight. The Freeville plot was inoculated with the light phenotype of the early blight pathogen, *A. tomatophila*, which is the most severe isolate of this pathogen. Data collection proceeded with readings starting with observation of first disease. The plots were later inoculated with the late blight pathogen as well. The disease screens resulted in the elimination of one line that was either contaminated or was still segregating for LB resistance. The trial confirmed the resistance of the other lines.

The 2006 season proved to be one of the wettest summers on record for Freeville, NY, with 19” recorded rainfall during the experiment. Early in the season, a heavy rain caused a nearby stream to start to overflow. The plot was not flooded, but the water table was nearly at ground level in parts of the plot. Wet soil conditions resulted in stunted plants, which especially occurred in small field depressions. Therefore we were not able to use the Ithaca trial to test for fruit characteristics and yield. Since plants were obviously severely stunted in major portions of the field, fruit and yield data would not have given valid results.

Based upon the prior two years results, several of the lines were selected for production of experimental hybrids that will be homozygous for late blight and early blight resistance.

The trial of the lines in PA also provided information on line performance. The trial performed under field conditions at Rock Spring, PA in the summer of 2006 tested a total of 14 fresh market breeding lines or cultivars, including 8 Cornell lines, 4 Penn State lines and two controls (LB-susceptible cv. Mt. Fresh Plus and LB-resistant breeding line NC96LB). The entries were grown in three replicates of 12 plants each. Throughout the season no inoculations were used, but there was uniform presence of EB throughout the 12-acre tomato plot. There was no incidence of LB. During September 2006, plants were visually evaluated in three different dates for the incidence of EB (% defoliation) as well as other horticultural characteristics. The AUDPC for the EB disease progression was calculated. Overall and comparatively, the materials developed at Cornell and Penn State exhibit good levels of EB resistance. The AUDPC tended to be lower indicating less disease in most of the Cornell lines. As is commonly observed, the intensity of EB development is highly dependent on fruit load and plant maturity, and greater disease expression could also be an indication of plant with high yield and/or concentration of set for plant vine size. Higher AUDPC and disease could also be an indication of residual segregation for disease resistance, so uniformity of disease level must be considered.

The lines selected based on the results summer of 2005 and 2006 were used in the winter of 2006/2007 for the production of hybrid seed. Crosses were generally successful, and the resulting seed will be used in trials summer of 2007. Most of these hybrids were generated in cooperation with Dr. R. Gardner of NCSU, and are the crosses of a Late Blight/Early Blight line from the Cornell program crossed with a Late Blight/Early Blight line from the NC program. Both sets of lines possess Late Blight/Early Blight resistances, but the two sets of lines complement each other for other important traits.

Year 3: The Late Blight/Early Blight resistant hybrids produced in the winter of 2006/2007 were trialed in replicated trials in Ithaca, NY and near Albany NY, in PA, and in NC. The NY trials included CU plots in Ithaca, and demonstration plots are also in Freeville and at Geneva. Plants were also distributed to organic farmers who are grower-evaluators in the organic seed initiative administered at Cornell.

The majority of the hybrids being tested are fresh market determinate types. All of these hybrids were generated in cooperation with Dr. R. Gardner of NCSU, and are the crosses of a Late Blight/Early Blight line from the Cornell program crossed with a Late Blight/Early Blight line from the NC program. Since both parent were fixed for the resistances, these hybrids are also fixed for Late Blight/Early Blight resistance, and should have the highest level of control for both diseases.

The trial in Ithaca were established in RCBD, with one row per genotype per rep and three reps. Productivity of the experimental hybrids were not significantly different from that of the control hybrids when considering either fruit number or total weight of fruit on the two harvest dates separately or combined for total production. The experimental hybrids have number of fruit similar or in a very few cases sign higher than that of control hybrids. Similarly the experimental hybrids have fruit weights similar to that of the control hybrids.

A consistent issue with freshmarket tomato lines carrying the EB tolerance is tendency for reduced fruit size. It is not clear whether this is a linkage issue, since the tolerance was derived from a wild species, or if it is a pleiotropic affect of the EB tolerance. This tendency for smaller fruit is reflected in the comparison of the two NC parents. The line NC33EB1 has somewhat larger fruit than NC45EB2., and hybrids with NC33EB1 as the male parent tend to have larger fruit than parallel hybrids with the smaller fruited line NC45EB2 as the male parent. Therefore it is possible to create EB tolerant lines with larger fruit size. The hybrids best for fruit size in this trial were those with 071002 or 071008 as the female parent and NC33EB1 as the male parent.

The trial in Roxbury was established in RCBD, with one row per genotype per rep and three reps. The plants in this trial were grown trellised on raised beds on an organic farm, hosted by an experienced organic grower, data were collected by an extension agent, Chuck Bornt. The entries in this trial were a subset of the experimental hybrids tested in the larger Varna trial. The results of the Roxbury trial were similar to those of Varna trial for the common entries. Again the experimental hybrids with 07-1008 as the female parent produced larger fruit than most of the other experimental hybrids.

The trial in PA included 4 fresh-market hybrids, which were grown in three replications of 12 plants each, and which was exposed to EB by natural infection. There was no incidence of LB. Of the 4 hybrids tested, the two with NC33EB1 as a parent, crossed to a CU line, were better, with larger fruit. This location did not have hybrids with 07-1008 as a parent.

In NC, Randy Gardner also tested the hybrids of the Cornell lines 071001, 071002, 071006, 071007, 071008, or 071009 or 048143-7 crossed to the male parent NC33EB1 in North Carolina. He reported that all of the hybrids withstood late blight and early blight in his fields. He had a preference for the crosses 071009 x NC33EB1 and 048143-7 x NC33EB1 for the fruit size in his location, although they did not produce the largest fruit in the NY trials. He also noted that naturally occurring SLS moved into the plot near the edges of the planting.

G. Results.

The goals of all objectives were met. Lines fixed for resistance to early blight and late blight were confirmed, selected for best horticultural type, and used, in conjunction with similar lines from NC, to create hybrids that were also homozygous for these resistances. Trials of the resulting hybrids determined which had the best combination of resistance and horticultural type.

In conjunction with the year 3 Ithaca trials were shown to seed company representatives in field day. Company representatives showed considerable interest in the hybrids. The Cornell lines 07-1002, 07-1008, 07-1009 and 048143-7. were released to the seed companies under MTA, and the companies are using them for production of experimental hybrids. EB/LB lines NC33EB1 and NC45EB2 have also been released by the NC program. Companies are testing experimental hybrid with LB and EB resistance, some of which will be on the market within the next 2 years.

During the course of this project, fungicides consisting of convention, materials that induce systemic acquired resistance (SAR), and those suitable for organic production were trialed in both greenhouse and field experiments. The best subset of products were tested in field trials on lines NC33EB-2 and NC45EB-1 in 2006 and on hybrid NC06115 in 2007 and 2008, the latter providing the highest level of resistance for EB in a hybrid. Our results showed that genetic

resistance provided excellent control of EB when compared with susceptible (Supersonic) and intermediate resistance (Mountain Fresh). Although the conventional treatment (chlorothalonil) provided the best control in 2006 and 2007, acceptable control was also achieved with biological fungicides (*Bacillus subtilis*) combined with Actigard (a SAR product) or fixed copper (Champion).

In the 2008 experiments, we extended our work of pyramiding genetic resistance and fungicide inputs by following a spray schedule that used reduced-risk products with the lowest environmental impact quotient (EIQ) available. Outstanding control of EB in the hybrids NC06115 (which has EB resistance) was achieved with the lowest EIQs tested (50 and 113) compared with the standard chlorothalonil treatment (300 EQ) needed to control the disease in susceptible hybrids.

In addition to the work planned, the experiment led to two unexpected discoveries. The most noteworthy discovery in the cooperator fields was the occurrence of Septoria leaf spot (SLS) in materials resistant to EB and LB, as well as susceptible controls, in both 2006 and 2007. This finding emphasized the need for the addition of SLS resistance into the current EB/LB resistant materials to be fully acceptable in the Northeast region and other adjoining regions, and to fully achieve reduction in fungicide usage. Sources of stable resistance for SLS have subsequently been identified and progress is being made to combine this resistance along with the previous EB/LB resistant lines we studied in this project.

A second discovery, by chance in the conventional cooperator fields, was the identification of resistance to strobilurin fungicides in local strains of EB pathogen. Since strobilurin fungicides are widely utilized in most tomato growing areas, this development further emphasized the need for quick development and release of resistant varieties to minimize this problem.

Did this grant serve as seed money for obtaining additional, related grants? If so, please describe.

This project provided the impetus for continued funding of formula funds (Hatch) and allowed us to submit proposal for additional funding to state and federal agencies. The proposal is currently pending

Has your project or study enhanced collaboration among stakeholders interested in the development and implementation of improved IPM strategies and systems?

Knowledge of our work has been followed in other states in the NE region and beyond. The materials with combined LB/EB resistance were requested by most US and international seed companies, who are using them in their varietal development. New hybrids with LB/EB resistance are in the commercialization process, and should be commercially available for the 2010 season. We have also witnessed improvement in the interest in disease resistant varieties for commercial and organic growers, and with the added interest by High Tunnel vegetable growers. The interest with the latter group is now underway within the region in the form of additional grant proposals to foster this work. We have made presentations at key vegetable conferences in New York, Pennsylvania, Ohio, North Carolina, Virginia and Mississippi to cover

both the breeding efforts and the reduced use of pesticides. States have also expressed interest in learning more about the use of low EIQ products.

H. Impacts.

1. Safeguarding human health and the environment:

As a result of this project, we have addressed the use of genetic resistance in conjunction with use of reduced-risk fungicides, lower EIQ fungicides, and organic products. We have demonstrated that this approach is feasible for both conventional and organic growers. Further, we have shown that multiple-resistance can also be adopted by home gardeners, to the point of not having to spray their individual gardens.

2. Economic benefits:

There are distinct savings to be realized from this work. First, there are obvious cost savings for growers since they do not need to rely so heavily on the use of pesticides. Secondly, we demonstrated that when using the lower EIQ fungicides, one can not only improved disease control when combined with genetic resistance, but that by following the lower EIQ program there are cost savings to the grower (lower fungicide costs) when compared with a conventional program.

3. Implementation of IPM:

We have made oral presentations of our work to grower conferences in New York and Pennsylvania (3). We have also made six oral presentations at tomato disease workshops in different parts of the US, encompassing IPM regions in the Northeast, Southern and North Central regions. (see Appendices for web sites and publications)

I. Appendices. Please attach to your report any of the following that will enhance our understanding of your project and its impacts:

ONLINE INFORMATION

WOOSTER, OH Meeting of the 20th Tomato Disease Workshop, Oct 20, 2005

Online: <http://www.oardc.ohio-state.edu/millerlab/TDW%20Proceedings/TDW%202005%20Proceedings.pdf>

Mutschler, M. A., Zitter, T. A., Bornt, C. 2005. Tomatoes for the Northeast combining early blight and late blight resistance.

ASHEVILLE, NC Meeting of the 21st Tomato Disease Workshop, Nov 9, 2006

Online: <http://cherokeereservation.ces.ncsu.edu/fletcher/programs/plantpath/2006-11-tomato-disease/TDW06proceedings.pdf>

Mutschler, M. A., Zitter, T. A., and Bornt, C. 2006. Effectiveness of genetic resistance and lite fungicides for disease control.

Zitter, T. A., and Drennan, J. L. 2006. Efficacy of fungicides for early blight and late blight control.

WILLIAMSBURG, VA meeting of the 22nd Tomato Disease Workshop, Oct 25, 2007
<http://www.cpe.vt.edu/tdw/>

Zitter, T. A., Zitter, S. M., and Drennan, J. L. 2007. Evaluation of lite fungicides for early blight and Septoria leaf spot control and pyramiding fungicides with genetic resistance.

RAYMOND, MS meeting of the 23rd Tomato Disease Workshop, Oct 16, 2008
http://conference.ext.msstate.edu/tomato_disease_workshop/

Zitter, T. A., and Drennan, J. L. 2008. Using an Environmental Impact Quotient (EIQ) driven fungicide schedule to maximize control of early blight and Septoria leaf spot of tomato. (in preparation)

Zitter, S. M., Zitter, T. A., Southwick, S., and Mutschler, M. A. 2008. Expression and genetic control of resistance to Septoria leaf spot in tomato. (in preparation)

OTHER PUBLICATIONS

Zitter, T. A., Drennan, J. L., Mutschler, M. A., and Kim, M. J. 2005. Control of early blight of tomato with genetic resistance and conventional and biological sprays. *Acta Hort (ISHS)* 695:181-190. Proc. of the First International Symposium on Tomato Diseases. M. T. Momol and J. P Jones, eds., Orlando, Florida, USA.

Zitter, T. A., and Drennan, J. L. 2008a. Using host resistance and lite fungicides to control early blight on tomato, 2006. *Plant Disease Management Reports (online)* Report 2:V060. DOI: 10.1094/PDMR02. The American Phytopathological Society, St. Paul, MN.

Zitter, T. A., and Drennan, J. L. 2008b. Using host resistance and lite fungicides to control early blight and Septoria leaf spot on tomato, 2007. *Plant Disease Management Reports (online)* Report 2:V061. DOI: 10.1094/PDMR02. The American Phytopathological Society, St. Paul, MN.

Zitter, T. A., and Drennan, J. L. 2009. Using host resistance and reduced EIQ fungicides to control early blight and Septoria leaf spot on tomato, 2008. (for submission to *Plant Disease Management Reports [online]* Report 3) The American Phytopathological Society, St. Paul, MN.