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SolviNix® LC: A Plant Virus Based Bioherbicide

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On December 11, 2014, a historic milestone was set when the U.S. Environmental Protection Agency granted FIFRA Section 3 registration for a bioherbicide containing a plant virus as the active ingredient. The registration marked the first time a plant virus was registered as an herbicide active ingredient anywhere in the world.

The long process of registration, which began in 2005, was finally successful thanks to the guidance and able navigation of the registration effort by Dr. Michael Braverman, Manager, IR-4 Biopesticide and Organic Support Program. Collaboration of several UF-IFAS research and extension faculty, staff, and students as well as the support and encouragement from the Florida Cattlemen's Association, the Tropical Soda Apple Taskforce, and the Florida Department of Agriculture and Consumer Services-Division of Plant Industry were also key to our success.

Left: Thorny leaves of tropical soda apple plant. Right: Flowers.

Tropical soda apple fruit. Left: Partially mature, green fruit (this picture is from Bugwood, U. Ga. collection). Right: Fully mature, yellow fruit.

The bioherbicide, with the trade named *SolviNix®* LC, is labeled for use as a post-emergent foliar herbicide to control *Solanum viarum* (tropical soda apple), a South American plant that has become invasive in pastures and conservation areas in the southeastern United States.

Tropical soda apple is a Noxious Weed in the United States and a Class 2 Regionally Prohibited Weed across New South Wales, Australia. It is as well an invasive or problematic weed in several other countries, including Brazil where it is native. Besides pastures and conservation areas, it is reported to affect crops in some countries.

The active ingredient of *SolviNix®* LC is a strain of Tobacco mild green mosaic virus (TMGMV). Known for nearly 70 years, TMGMV is a pathogen of tobaccos (*Nicotiana* spp.), peppers (*Capsicum spp.*), and about 20 other species in the Solanaceae. It was first described as a mild strain of Tobacco mosaic virus (TMV) and later named variously as Green tomato atypical mosaic virus, Para-tobacco mosaic virus, Tobacco mosaic virus-South Carolina mild mottling strain, Tomato atypical mosaic green mottling strain, Tobacco mosaic virus strain U2, and Tobacco mosaic virus strain U5. It is now classified as a distinct Tobamovirus species, *Tobacco mild green mosaic tobamovirus*, with two naturally occurring strains, U2 and U5. An isolate of the U2 strain is used as the active ingredient in *SolviNix®* LC.
TMGMV is believed to occur worldwide in tropical and subtropical regions where *Nicotiana glauca* (tree tobacco), a

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natural host to this virus, is distributed. Normally, TMGMV is found infrequently in *N. glauca* and some cultivars of tobacco and pepper but is not known to cause serious economic losses. Typically, it causes a mild, green, systemic mosaic symptom in susceptible hosts but in tropical soda apple it elicits a lethal hypersensitive response expressed as systemic necrosis and plant death. Just one application of the virus (e.g., as high-pressure foliar spray) to a few physiologically active leaves on a plant is sufficient to infect and kill the entire plant, including the root system. *SolviNix®* LC has performed consistently in field trials yielding > 85% weed kill in about 3-6 weeks following a high-pressure foliar application. So far, no natural resistance to TMGMV U2 has been found among tropical soda apple plants in the United States. Also, several tropical soda apple accessions from New South Wales, Australia, have been found to be equally susceptible as the Florida plants to the virus.

The discovery that TMGMV-mediated lethal hypersensitive response could be used as a novel method of weed control was made in 1999 in Dr. Charudattan's biological control of weeds program at the University of Florida-Institute of Food and Agricultural Sciences (UF-IFAS). The project was subsequently moved forward and industrially developed by BioProdex, Inc., the *SolviNix®* LC registrant. With funding from USDA-National Institute of Food and Agriculture-Small Business Innovation Research (USDA-NIFA-SBIR) Phase I and II grants, the company developed a scalable industrial process to mass produce the virus and formulate it into a commercial product, and assembled a registration data package.

Registration of *SolviNix®* LC exemplifies an effective collaboration in research and technology transfer involving a land-grant university (UF-IFAS), USDA-NIFA-SBIR, IR-4, and a private enterprise (BioProdex, Inc.).

It can be reasoned that the time it took to register *SolviNix®* LC, was nearly 10 years from our pre-registration consultation with the EPA in 2005, is due to the fact that this was the first proposal to register a virus as a bioherbicide agent. With no prior example to draw from, much effort went into data gathering, particularly nontarget plant host range, field efficacy, and label parameter data. In the end, it is the uniqueness of the tropical soda apple-TMGMV U2 system coupled with several well-known features of the virus that assured that the virus can be used safety as a bioherbicide.

First and foremost, TMGMV U2 kills the tropical soda apple plant quickly, completely, and consistently, which is a rare feature among plant viruses. Typically, plant response to virus infection is expressed as immunity (no visible plant response), resistance (necrotic local lesions in infected leaves only), or susceptibility (systemic mosaic, foliar mottling, plant stunting, and other debilitating yet nonlethal symptoms). Relatively rarely, as in the TMGMV U2-tropical soda apple system, the resistance response is expressed as lethal, hypersensitive, systemic necrosis.

Since infected tropical soda apple plants are completely killed, no infected but still living plants are

Tropical soda apple infestation in a pasture (left) and a conservation area

Reaction of TSA to SolviNix® LC treatment. Left to right: An untreated TSA plant and a SolviNix® LC treated plant on days 13, 23, and 33 after treatment.

left in the field to serve as a virus reservoir. Moreover, as the virus is mechanically transmitted and has no known, confirmed, vector capable of disseminating it, it can be used in targeted applications without the risk of secondary spread.

Despite its worldwide occurrence, TMGMV is genetically stable, as evidenced by the low frequency of emergence of new strains in nature. Furthermore, in nature TMGMV has a restricted host range compared to the moderately broad host range reported from artificial manual inoculations in the laboratory/greenhouse.

Unlike fungal foliar bioherbicides that require optimum moisture and humidity for performance, the virus infectivity and disease development are not constrained by microclimatic conditions. Consequently, field application of *SolviNix®* LC is generally unencumbered by the weather.

Finally, from the literature it seemed possible to mass produce the virus on an industrial scale to meet the market needs. We have now confirmed this through our production process. The manufactured virus end-product, when stored properly, is stable for many years, which makes the industrial production cost-effective and expedient. So, in retrospect, TMGMV U2 is an ideal viral agent for development as a bioherbicide.