

TROPICAL SODA APPLE MANAGEMENT PLAN

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Recommendations from the Florida Tropical Soda Apple Implementation Management Team 2012 First Edition

The Tropical Soda Apple Management Plan was developed to provide information and make recommendations for the integrated management of tropical soda apple in Florida and other infested states. This is the first edition of the Tropical Soda Apple Management Plan for the USA. It will be updated periodically to reflect changes in management alternatives and implementation techniques.

Mention of a trade name or proprietary product does not constitute a guarantee or warranty of the product by the Tropical Soda Apple Management Team. There is no express or implied warranty as to the fitness of any product discussed. Any product trade names that are listed are for the benefit of the reader and the list may not contain all products available due to changes in the market.

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I Introduction

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Tropical soda apple (TSA), *Solanum viarum* Dunal (Solanaceae) is a weed native to southeastern Brazil, northeastern Argentina, Paraguay, and Uruguay that has invaded Florida grasslands and natural ecosystems. In 1988, TSA was first reported in the USA in Glades County, Florida (Coile 1993, Mullahey and Colvin 1993). The pathway of introduction is unknown, but it may have been accidentally introduced with cattle carrying undigested TSA seeds that were imported from Brazil. In 1993, a survey of beef cattle operations in south Florida estimated that there were 157,145 ha of infested pastureland, twice the infestation present in 1992 (Mullahey et al. 1994). The infested area increased to more than 303,000 ha in 1995 (Mullahey 1996, Mullahey et al. 1998). Currently, more than 404,000 ha are believed to be infested in Florida (Medal et al. 2010b). Due, at least in part, to favorable environmental conditions, a lack of natural enemies (herbivores and pathogens), and seed dispersal by wildlife and cattle feeding on the fruits, TSA has spread rapidly and has been observed in the majority of Florida counties and also in Alabama, Georgia, Louisiana, Mississippi, North Carolina, Pennsylvania, South Carolina, Tennessee, Texas, and Puerto Rico (Bryson & Byrd Jr. 1996, Dowler 1996, Mullahey et al. 1993, 1998, Medal et al. 2003, 2010a). Although TSA has been reported in Pennsylvania and Tennessee, it is highly probable that it does not overwinter in these states. Patterson (1996) studied the effects of temperatures and photoperiods on TSA in controlled environmental chambers and speculated that the range of TSA could expand northward into the midwestern US. TSA was placed on the Florida and Federal Noxious Weed Lists in 1995.

TSA typically invades improved pastures, where it reduces livestock carrying capacity. Foliage and stems are unpalatable to cattle; dense stands of the prickly shrub prevent cattle access to shaded areas, which results in summer heat stress (Mullahey et al. 1998). TSA control costs for Florida ranchers were estimated at \$6.5 to 16 million annually (Thomas 2007), and economic losses from cattle heat stress alone were estimated at \$2 million (Mullahey et al. 1998). TSA is a reservoir for at least six crop viruses (potato leaf-roll virus, potato virus Y, tomato mosaic virus, tomato mottle virus, tobacco etch virus, and cucumber mosaic virus) and the potato fungus *Alternaria solani* Sorauer (McGovern et al. 1994a, 1994b, McGovern et al. 1996). In addition, major insect pests utilize TSA as an alternate host; including Colorado potato beetle, *Leptinotarsa decemlineata* (Say); tomato hornworm *Manduca quinquemaculata* (Haworth); tobacco hornworm, *Manduca sexta* (L.); tobacco budworm, *Heliothis virescens* (Fabricius); tomato pinworm, *Keiferia lycopersicella* (Walsingham); green peach aphid, *Myzus persicae* (Sulzer); silverleaf whitefly *Bemisia tabaci* biotype B (Bellows & Perring); soybean looper, *Pseudoplusia includens* (Walker); and the southern green stink bug, *Nezara viridula* (L.) (Habeck et al. 1996, Medal et al. 1999, Sudbrink et al. 2000). TSA also reduces biodiversity in natural areas, ditch banks, and roadsides by displacing native vegetation (Langeland and Burks 1998). TSA interferes with restoration

efforts in Florida by invading area that are reclaimed following phosphate mining operations (Albin 1994).

TSA Management practices in Florida pastures primarily involve herbicide applications and mowing (Sturgis and Colvin 1996, Mislevy et al. 1996, 1997, Akanda et al. 1997). Herbicides or mowing provide temporary weed suppression at an estimated cost of \$61 and \$47 per ha, respectively (Thomas 2007). However, application of these control methods is not always feasible in rough terrain or inaccessible areas.

In June 1994, the first exploration for TSA natural enemies in South America was conducted by University of Florida and Brazilian researchers (Medal et al. 1996). Sixteen species of insects were found attacking the weed during this two-week survey. Host specificity tests were initiated in 1997 by J. Medal (University of Florida) in collaboration with the Universidade Estadual Paulista, Jaboticabal campus, Brazil, the USDA Biological Control Laboratory in Hurlingham, Buenos Aires Province, Argentina, and the USDA Biological Control Laboratory in Stoneville, MS. The South American leaf-feeder *Gratiana boliviana* (Chrysomelidae) was approved for field release in Florida in summer 2003. In 2004, The University of Florida – IFAS, the Florida Department of Agriculture and Consumer Services-Division of Plant Industry (FDACS-DPI), the USDA-APHIS, and the USDA-ARS convened a Biocontrol Implementation Team for TSA consisting of state, federal, and Florida cattle ranchers. The goal of the ‘TSA Biocontrol Team’ was to develop, and implement an effective management based on imported natural enemies of TSA. Based on the success achieved through the field release and establishment of the first biocontrol agent, this Tropical Soda Apple Management Plan compiles known information and provides recommendations for the integrated control of TSA in the affected states.

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II. Economics of Tropical Soda Apple in Florida

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Introduction

The economic effects of tropical soda apple (TSA), or for that matter any invasive exotic species, can be measured at several different levels and the appropriate level often depends on the goal of the analyst. At one level, the effects can involve simply measuring the direct expenditures made by parties trying to control TSA. At another level, it can be expanded to include the ripple effect of control expenditures and lost productivity across the economy-at-large. At yet another level, the effects could be expanded to include the indirect economic losses resulting from the reduction of ecological services in a TSA-dominated environment (Thomas 2011).

One of the more common forms of economic analyses is that of comparing a mitigation action's potential benefits and costs. These benefit-costs studies are traditional and effective means for carefully comparing mitigation options, and while they are considered effective analytical devices, practitioners must be careful to correctly classify costs and benefits. Costs are simply the amounts paid or charged for something and would include the costs of TSA control. However, costs would also include any lost opportunity experienced by foregoing potential TSA benefits (Table 1). In the first case, these costs would include obvious expenses such as controlling TSA in pasture land, but in the second case they could also include lost benefits from the removal of TSA (if any were known to exist). Benefits, on the other hand, are the improvement in human welfare resulting from the treatment and control of TSA (Table 2). These benefits can include both direct and indirect values and, in the case of TSA, would include the benefits to cattle producers and the economy at large from improved cattle production, and the less apparent benefits to humans from the recovery of ecological services from displaced native plant and animal species.

Table 1. : Costs of Tropical Soda Apple Control

| Direct Project Costs | Indirect Costs (opportunity cost) |
|---|--|
| 1. Treatment development costs -from finding/discovering and developing a control measure | 1. Lost marketable goods/services (from removal of invasive exotic) |
| 2. Project implementation costs - from applying the control factor(s) and follow up | 2. Lost non-marketable goods/services (from removal of invasive exotic) <ul style="list-style-type: none"> i. Use value -Directly measureable ii. Non-use value -Indirectly measurable |

Table 2. Benefits from Tropical Soda Apple Control

| Direct Project Benefits | Indirect Benefits |
|--|---|
| Restored marketable goods/services <ul style="list-style-type: none"> i. Direct effects (\$) – eg., cattle producers ii. Indirect effects (\$) – eg., economic sections linked to cattle producers iii. Induced effects (\$) – economy at large | Restored non-marketable goods/services <ul style="list-style-type: none"> i. Use value – from users of TSA improved environment -Directly measureable ii. Non-use value – from non-users of TSA improved environment (eg, ecological services) -Indirectly measurable |

This chapter will look at only one measure of costs associated with TSA infestation: control costs in Florida's affected pasturelands. By extension, some of these costs are potential benefits of a successful TSA control project and should be considered as such when measuring the cost effectiveness of a potential or on-going bio-control project. In addition to direct treatment costs incurred by cattle producers, the chapter will report both the indirect and induced effects of these costs to the overall economy; often termed the ripple effect.

Methods and Results

The balance of this chapter will focus on the cost of TSA control by Florida's cattle producers in 2006. While there are likely other TSA costs to consider, its impact on cattle production is likely the most significant impact on humans.

The fact that TSA can grow into dense stands, and even render entire pastures unusable for grazing, has had a significant economic impact on Florida's cattle producers (Salaudeen 2006). Cooke (1997) indirectly attributed the loss from TSA infested pastures at \$11 million, or about 1% of total Florida beef sales. In addition, Cooke (1997) estimated another \$2 million in economic losses due to heat stress as cattle are denied access to shaded areas crowded with TSA. Yet despite this and other strong evidence of substantial economic losses to Florida's cattle producers, the only direct measure of TSA losses are reported by Saluadeen (2006).

In 2006, Salaudeen developed and administered an economic survey to a sample of Florida's cattle owners (Salaudeen 2006). The questionnaire was designed to document the respondent's type of operation, their knowledge of TSA and the plant's effect on their ranch's operation, their method(s) of TSA control and costs and a series of questions on the history of their operation, their familiarity with weedy pests, and several demographic questions. A complete rendering of survey results can be found in Saluadeen (2006), and Saluadeen et. al. (2011).

A survey instrument was pre-tested and mailed to all 3,500 members of the Florida Cattleman's Association across the state of Florida (Saluadeen 2006). A total of 927 survey questionnaires were returned by cattle owners for an adjusted response rate of 27% (accounting for undeliverables).

Survey respondents claimed to follow four basic mitigation approaches to TSA infestations; reduced the rate of cattle stocking, increased the use of supplementary feed, increased the use of weed control, and removed pastureland from production (Table 3.) (Salaudeen 2006). The participation rates for each approach varied regionally. The most common control measure in all three regions of Florida was chemical control. Nearly all of Florida's cattle ranchers used a pesticide to protect their pastureland from pests, and this option added more to the cost of production (Michael and Norman 2002). The other three management responses were used sparingly by ranchers in all three regions. Cattle producers were willing to increase production cost by controlling TSA to maintain their constant herd size.

Table 3: Management Response to TSA by Region¹

| Types of response | North | Central ² | South |
|---|-------|----------------------|-------|
| Proportion who reduce stocking rate (%) | 2.4 | 6.1 | 7.4 |
| Proportion who control the weed (%) | 35 | 75 | 76 |
| Average stocking rate reduction (%) | 0.8 | 3 | 3.7 |
| Proportion who take pasture out of production (%) | 3.6 | 6 | 2 |

North Florida counties: Alachua, Baker, Bay, Bradford, Calhoun, Clay, Columbia, Dixie, Duval, Escambia, Flagler, Gadsden, Gilchrist, Gulf, Franklin, Hamilton, Holmes, Jackson, Jefferson, Lafayette, Leon, Levy, Liberty, Madison, Nassau, Okaloosa, Putnam, Santa Rosa, St. Johns, Suwannee, Taylor, Union, Wakulla, Walton, Washington.

Central Florida counties: Brevard, Citrus, Desoto, Hardee, Hernando, Highlands, Hillsborough, Indian River, Manatee, Marion, Okeechobee, Orange, Osceola, Pasco, Pinellas, Polk, Sarasota, St. Lucie, Sumter, Volusia

South Florida counties: Broward, Charlotte, Collier, Dade, Glades, Hendry, Lee, Martin, Monroe, Palm Beach

The most preferred methods of TSA control were chemical herbicides and mowing (Table 4). Remedy™ and Roundup™ were the two most commonly used herbicides, yet many had begun using Milestone™, a new herbicide specifically designed for TSA control.

Table 4: Tropical Soda Apple Control Costs and Methods by Region

| Category | North | Central | South |
|------------------------------------|--------------------------------|--------------------------------|--------------------------------|
| Top two methods of control. | Chemical (20%) Mowing (13%) | Chemical (45%) Mowing (29%) | Chemical (48%) Mowing (32%) |
| Top two chemicals used | Remedy (11%) Roundup (8%) | Remedy (38%) Roundup (22%) | Remedy (46%) Roundup (20%) |
| Average cost of control /acre (\$) | 25 | 19 | 18 |

Ranchers estimated their cost to control TSA on an acre basis, with the average cost for north Florida ranchers approximately a third higher than central and south ranchers.

To determine the extended economic impact or ripple effect of TSA control through the economy at large, the total expenditures associated with TSA control needed to be determined. The regional economic impact of TSA was estimated as a function of the total regional pastureland, the TSA infestation rate, the proportion of ranchers attempting to control TSA, and the cost of TSA control. More generally, the regional cost (RC_i) for controlling TSA can be expressed as:

$$RC_i = f(I_i, A_i, P_i, C_i) \quad [1]$$

Where:

I_i = TSA infestation rate for region i.

A_i = number of acres in commercial pastureland for region i.

P_i = proportion of cattle ranchers controlling TSA in region i.

C_i = average TSA control cost for region i in US \$.

(For region i, ... i + 1 = 1, ..., 3: north, central, and south)

Alternatively, the regional cost (RC_i) can be expressed as:

$$RC_i = I_i * A_i * P_i * C_i \quad [2]$$

Regional mean infestation rates (I_i) were calculated from rancher responses (Salaudeen 2006). The number of acres in commercial pastureland for each region (A_i) was obtained from the National Agricultural Statistics Services (NASS 2002). Each county's pastureland estimates were used to get the total regional pastureland and the proportion of cattle ranchers controlling TSA in each region (P_i) was derived from the number of survey respondents that choose to control their TSA invasion (Table 3). Regional TSA control costs (C_i) per acre were also identified from respondents (Table 4).

Upper and lower bounds for regional cost estimates were then calculated for a 95% level of confidence. These estimates were the direct costs of TSA control by cattle producers and a direct measure of their lost profits (costs are assumed as losses to revenues). The 95% bounds represent the sampling error. In other words, these bounds on cost are the statistical estimate of the direct loss to cattle ranchers due to TSA invasion (Table 5).

Table 5: Estimates of Direct Regional Cost for Tropical Soda Apple Control

| Regions (pastureland acreage) (A_i) | 95% Statistical Bounds Lower & Upper | TSA Infestation rate (%) (I_i) | Ranchers that control TSA (%) (P_i) | Cost of TSA control/acre (\$) (C_i) | Regional cost of control (\$) ($RC_i$) |
|---|---|--|--|--|---|
| North (1,212,615) | Lower | 0.0133 | 0.29 | 23.56 | 110,191.44 |
| | Upper | 0.06 | 0.35 | 26.08 | 776,077.48 |
| Central (3,849,003) | Lower | 0.091 | 0.71 | 18.46 | 4,590,708.19 |
| | Upper | 0.1521 | 0.79 | 18.90 | 8,741,105.44 |
| South (1,002,726) | Lower | 0.01476 | 0.65 | 15.02 | 144,494.70 |
| | Upper | 0.1688 | 0.87 | 18.55 | 2,731,604.90 |

In the next step, the estimates of direct loss were used as input to the economic impact software, IMPLAN, an economic model that calculated the ripple effect of expenditures on the economy as whole. This was a two step process. IMPLAN first calculated the indirect impacts resulting from the direct costs and then it calculated the secondary or induced effects as the money moved from the ranches to their related support sectors and finally to the remainder of the economy; the ripple effect. Table 6 presents a summary of the economic impacts (direct, indirect, and induced) that resulted from controlling the infestation of TSA in cattle production in the three regions of Florida (north, central and south region).

Table 6: Estimated Economic Impact by Region

| Regions | Output | Direct (\$) | Indirect (\$) | Induced (\$) | Total Output (\$) |
|----------------|----------------|--------------------|----------------------|---------------------|--------------------------|
| North | Average | 368,387 | 186,913 | 48,145 | 603,445 |
| 95% C.I | Lower | 105,001 | 53,275 | 13,723 | 171,999 |
| | Upper | 739,522 | 375,219 | 96,650 | 1,211,391 |
| Central | Average | 6,248,361 | 1,862,488 | 699,358 | 8,810,207 |
| 95% C.I | Lower | 4,374,474 | 1,303,927 | 489,620 | 6,168,021 |
| | Upper | 8,329,376 | 2,401,830 | 915,511 | 11,646,717 |
| South | Average | 1,120,490 | 37,845 | 14,558 | 1,172,873 |
| 95% C.I | Lower | 137,688 | 1,712 | 633 | 140,063 |
| | Upper | 2,602,938 | 32,362 | 12,529 | 2,647,829 |

The cost of TSA control, money spent by cattle producers on chemicals and/or mowing, generated a total output loss of \$603,445, \$8.8 million and \$1.1 million annually for north, central and southern regions of the state respectively. These costs represent the revenues lost to all supportive business sectors resulting from reduced sales in supplies to cattle producers and reduced household incomes.

Generally, the largest economic impact from TSA infestation was in central Florida. This may be due to the high infestation rate and high proportion of ranchers that controlled the weed relative to other regions.

Conclusion

Allowed to spread, TSA will likely generate more economic losses to the cattle industry in terms of reduced pastureland and increased control cost. These losses to the cattle industry and its supporting sectors could significantly impact both the regional and state economies. It could also create economic inefficiency in cattle production as funds will be redirected from other factors of production to offset the cost of TSA control.

Based on control cost, smaller producers with less pastureland would likely feel the effect of TSA infestation more acutely due to smaller profit margin per animal. Larger operators with more expansive pasture tracts, as in the case with central and south Florida, may adapt to the pest better by simply moving their cattle to areas without TSA.

With statewide economic impacts ranging from \$6.5 million to \$16 million annually, TSA is a major concern for Florida and other southeastern states. This study only considered the impact to beef cattle producers, yet there was evidence that TSA was causing ecological damage in natural areas by displacing native plants and disrupting ecological integrity. The plant invades hammocks, ditch banks, and roadsides, where it out competes native plants (Langeland & Burks 1998).

At the time of this survey (2006), the preferred method of TSA control was chemical herbicide, with usage ranging from 20% to 48% for north and south Florida respectively. Remedy™ and Roundup™ were the two leading herbicides survey respondents reported using to control TSA. With continued TSA spread and the absence of alternative effective control measures, it is likely that the demand for these herbicides will continue to grow and this might have negative impact on the environment. Moreover, most of the chemical industries that produce these herbicides are located outside the state meaning that most of the money spent on herbicides will continue to leak away from the Florida economy.

The recent release of the South American leaf beetle, *Gratiana boliviana* as a method of TSA biocontrol has shown evidence of significantly reducing TSA on developed pasture land, particularly in the central and southern regions of Florida. In 2010, a follow up survey of Florida's cattle producers was administered to document the beetle's impact on TSA control on Florida's pastureland. While the survey is still being analyzed, preliminary results suggest a cost savings of approximately 50% statewide. If these savings are verified, it could lead to a state-wide savings of between \$3.25 to \$8 million annually, or assuming the savings are permanent, \$108 to \$266 million in total savings.

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III. Biology and Ecology of Tropical Soda Apple

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Introduction

Tropical soda apple (*Solanum viarum* Dunal) is a perennial weed that is a serious problem in many perennial grass pastures of Florida and throughout the southeastern United States. Tropical soda apple (TSA) is unpalatable to livestock and can infest a pasture or native area in 1-2 years resulting in loss of forage production and lower stocking rate. Geographically, the incidence of this plant in Florida has been highest in the southern part of the Florida peninsula though it occurs throughout the entire state and is present in Alabama, Georgia, Mississippi, South Carolina, Tennessee, Texas, Louisiana, and Puerto Rico. TSA escaped from Florida mainly from shipping cattle to other states but also from hay and sod sales, and wildlife movement. TSA was first collected in Florida from Glades County in 1988. In Florida, TSA infested acreage totaled 10,000 acres in 1990, increased to 400,000 acres in 1995, and presently infests over 1 million acres.

Tropical soda apple is a species belonging to the *Solanum* section Acanthophora of the prickly subgenus Leptostemonum, and is a common weed in South America, India, and West Indies. Recently, TSA was identified in Australia and could pose a threat to other Pacific Countries. In Brazil, TSA is primarily found in improved pastures that are not mowed. In Florida, TSA has been observed in pastures, hay fields, sod fields, landfills, ditch banks, citrus (*Citrus spp.*) groves, sugar cane (*Saccharum officinarum* L.) fields, vegetable fields, and native areas like oak hammocks (*Quercus spp.*) (Mullahey et al. 1993).

Biology

At maturity, TSA is from 3 to 6 ft tall. Stems, leaves, flower-stalks, and calyxes have broad-based white to yellowish prickles (Mullahey 1996). Leaves are pubescent (contain hairs), 4 to 8 in long and 2 to 6 in wide, and are moderately to deeply divided into broad pointed lobes (Wunderlin et al. 1993). The fruit is globular, about 1 in diameter and yellow when mature (Coile 1993). The immature fruit is green with white mottling like a watermelon and serves as a distinguishing characteristic. Seeds are moderately flattened and are covered in a mucilaginous layer which contains a glyco-alkaloid called solasodine (Mullahey et al. 1993). This plant was cultivated as a source of solasodine, a nitrogenous analogue of diosgenin, and used as a substitute for diosgenin in the synthesis of steroidal drugs (Sahoo and Dutta 1984).

Though TSA is an indeterminated plant, flowering and fruit production in Florida is concentrated from September through May. Throughout the year, this plant will have

immature and mature fruit present that ensures large numbers of viable seeds (approximately 40,000). Seedling emergence in south Florida primarily occurs from August through March. New plants can emerge from seed or from roots a few inches (3-6 in) long. The plant has an extensive root system which will extend 3 to 6 ft horizontally from the crown of the plant.

Seed germination has ranged from 30-100%, though average germination of seed from mature fruit is about 70%. Consequently, during one year, a single plant could supply enough viable seed to produce about 30,000 new TSA seedlings. Degradation of the fruit coat and subsequent release of seed on the soil surface takes about 2 months. Seed can remain dormant for months, if not years.

Ecology

Tropical soda apple has been observed as a weed in agricultural land and in natural areas. In Florida, TSA has been observed in pastures, ditch banks, citrus (*Citrus* spp.) groves, sugar cane (*Saccharum officinarum* L.) fields, vegetable fields, and native areas like oak hammocks (*Quercus* spp.). How TSA was introduced into Florida is not known. In Florida, it is an obligate weed mainly associated with human activities. The TSA seed is spread by cattle, wildlife, contaminated hay, grass seed, sod and moving water. Cattle and wildlife spread TSA by consuming the fruit and spreading the seed via feces.

The dung pile provides the perfect environment for seedling growth, by providing fertilizer, moisture, and resistance from grazing by cattle. Sampling cow manure from TSA infested pastures resulted in seed numbers ranging from 15,000 to 64,000 per ton of dry feces with an average seed viability of 84%. The average number of viable seed collected was 56,000 per ton of dry feces, or 56,000 viable seed per acre.

Cattle grazing TSA infested areas should be considered a major seed dispersal vector. Information is lacking on the amount and extent of seed dispersal from wildlife. Game animals like deer and feral hogs will consume the fruit and Sandhill Cranes have been observed to eat the seed from within the fruit. Disking a field, cattle congregating around a feeder, cleaning of ditch banks, or feral hogs rooting in a field appear to provide a favorable environment for TSA establishment and growth.

Standing water will stress the plant, even cause plant loss, but a new plant will emerge from seed once the area begins to dry. Cypress heads will have TSA in the center of the head until the summer rains completely flood the area, and then the TSA will die back to the outer regions (drier areas). As the water in the cypress head recedes during the winter, the TSA will begin to occupy (new plants from seed) the inner regions of the cypress head. During periods of drought, TSA will drop its leaves but regrowth will appear once soil moisture becomes available. Tropical soda apple can tolerate a frost or freeze but the foliage will suffer cold damage with some plant mortality. Many plants will begin regrowth about 2 weeks following a frost. Occasionally plants appeared

stressed and displayed symptoms of leaf mottling, inclusions, etc., associated with fungi, bacteria, and viruses

This plant has been identified as a host for 6 viruses that cause economic damage to vegetables. It also supports the reproduction and feeding by the Colorado potato beetle, green peach aphid, serpentine leafminer, tobacco horn worm, and the sweetpotato whitefly.

Implications of Biology and Ecology to Management of Tropical Soda Apple

Because populations of TSA continue to increase in the southeastern United States, it is imperative that land owners continue to prevent the spread of TSA within and across the Florida border. Although the movement of TSA seeds by wildlife cannot be prevented, seeds movement can be limited in some ways. One way to prevent the movement of TSA seeds is to clean off all equipment (remove TSA seeds and fruits) when leaving a pasture or area that is infested with TSA. Be sure to clean vehicles, mowers, tractors, and shoes.

Cattle can also transport TSA seeds. Therefore, it is important to ship cattle from an area that does not have TSA or is TSA-fruit free. Mowing a TSA-infested pasture prior to shipping will eliminate the fruit and the consumption of TSA seeds by cattle. The TSA seeds can remain viable in the digestive tract for up to six days. Therefore, when you buy cattle, hold them in one area for up to six days to avoid the spread of TSA to other areas on your ranch.

This plant spreads almost entirely from seeds and therefore it is a high priority to control seed production, “Dead plants don’t produce seeds”. Ranchers should have a zero tolerance for fruit and seed production. There are many control options for controlling TSA plants that include biological control, chemical control, and cultural control. Treated areas should be monitored monthly for at least 1 year to eliminate TSA.

Tropical soda apple is present throughout the entire state of Florida and probably will always be present in pastures and natural areas. Research on the biology and ecology has resulted in a better understanding of this invasive weed and management practices to prevent and control TSA. Weed control strategies continue to be updated as new information becomes available. Proactive TSA educational programs have heightened the awareness of agricultural producers and citizens of Florida. In addition, educators from other southern states have utilized TSA information from Florida to educate their agricultural producers. The University of Florida, IFAS, is committed to providing research and education programs to control TSA and to minimize the economic and ecological damage from TSA.

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IV. Extent of the Tropical Soda Apple Infestation in the USA

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Tropical soda apple (TSA) was first reported in Florida in 1988 (Mullahey et al. 1993), and has since been reported in Georgia, Alabama, Mississippi, Tennessee, South Carolina, North Carolina, Texas, Pennsylvania and California (EDDMaps 2011, Figure 1). The population in Pennsylvania arrived in the summer of 1996 (Lingenfelter and Curran 1998) but did not become established (PISC 2011). Warm temperatures and high precipitation during the summer months in Florida are conducive to rapid growth of TSA, but the cool, dry conditions experienced during the winter are detrimental. Winter mortality probably limits the distribution of TSA in temperate regions suggesting that new infestations probably start from seeds.

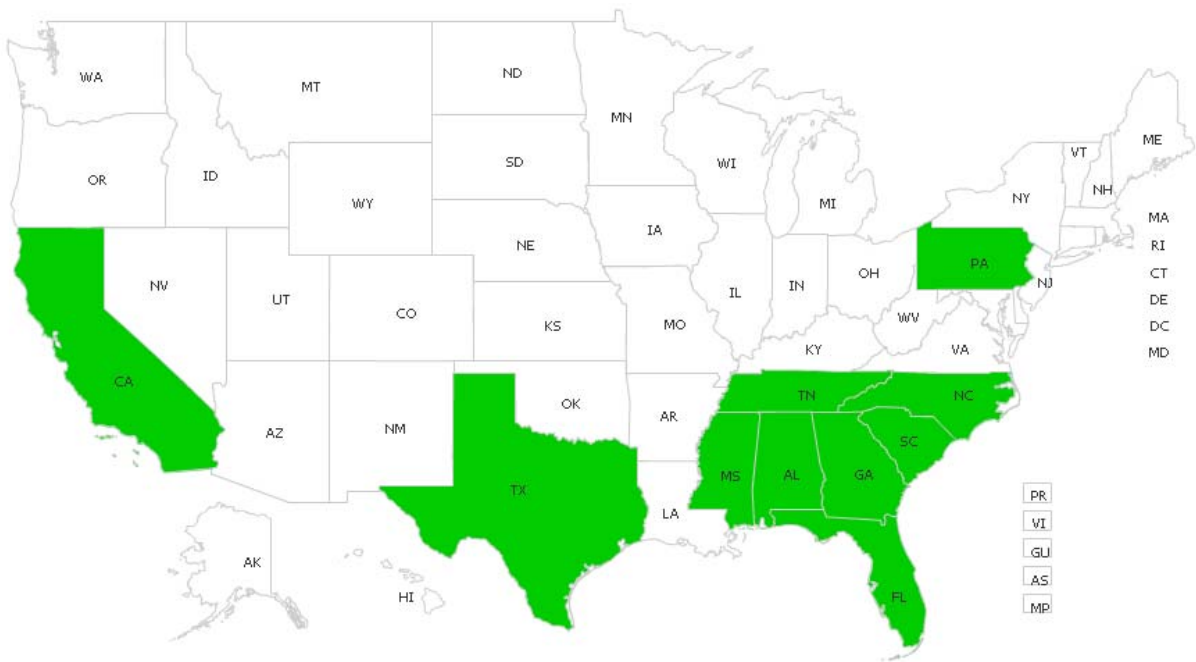


Figure 1. Distribution of tropical soda apple in The United States
(Source: www.eddmaps.org)

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V. Tropical Soda Apple Native Range Distribution

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Tropical soda apple (TSA) is native to southeastern Brazil, northeastern Argentina, Paraguay, and Uruguay (Figure 1) (Nee 1991), but has moved out of its native range to invade several other areas of the world in recent years. This weed has become naturalized in Honduras (Diaz et al. 2008), southern Mexico and Nicaragua (Medal unpublished data), the Caribbean, Africa, India, Nepal, China (Coile 1993, Chandra and Srivastava 1978), and the southeastern U.S. (Mullahey et al. 1998).

Tropical soda apple is found in scattered infestations in its native range, growing in grasslands, thickets, and disturbed areas (Bianco et al. 1991); but it is considered a major weed problem in introduced areas such as Florida (Mullahey et al. 1998). In Brazil, TSA is an occasional weed of field crops and its geographical distribution includes Goias, Minas Gerais, and the southern states (Costa et al. 1985). Genetic studies detected two haplotypes of TSA in Brazil, while only one of these haplotypes was found in the US populations (Kreiser et al. 2004). Therefore, additional surveys of natural enemies should focus in areas of Brazil where closely related TSA populations are present.

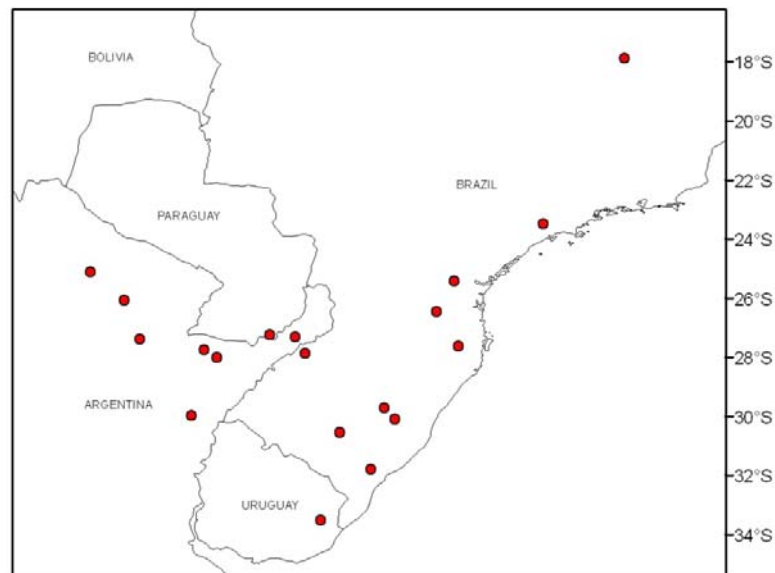


Figure 1. Distribution of tropical soda apple in the native range. Red dots represent voucher specimens.

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VI. Regulatory Strategies for Tropical Soda Apple

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Solanum viarum, commonly known as Tropical Soda Apple (TSA), is listed on both the Federal Noxious Weed List (1995) maintained by the United States Department of Agriculture (USDA) and the Florida Noxious Weed List (1994) maintained by the Florida Department of Agriculture and Consumer Services (FDACS). Language associated with the plants listed on the Florida and Federal noxious weed lists indicates that it is unlawful to introduce, possess, move or release *Solanum viarum* except under a permit by the USDA or FDACS. The term ‘possess’ within the context of FDACS regulation is intended to mean possession after willful introduction including intent to move or distribute. TSA can also be found on many other state noxious weed lists.

The introduction of TSA into Florida in the 1980’s went largely unnoticed until the early 1990’s when it was reported as displacing pasture grasses in south central Florida. The incursion of TSA into the Southeastern USA has resulted in some unique regulatory challenges. Primarily a rangeland weed pest, several interesting means of dissemination that had to be addressed were: 1) dispersal via livestock/wildlife TSA fruit consumption and ruminant seed deposits; 2) Bahia grass sod and seed harvest from pastures infested with TSA; and 3) compost manure operations. To effectively address these challenges, a TSA Task Force was formed by the Commissioner of Agriculture in 1993. The task force membership was composed of research, regulatory and industry stakeholders with interest and expertise in addressing TSA. The task force proved to be a very effective way to address all aspects of this new noxious weed threat in Florida including research, regulatory, outreach and educational issues. The task force expanded its scope of membership and influence to the Southeastern USA as TSA was discovered in other Gulf Coast States.

Using the task force, regulatory strategies were vetted through research and industry stakeholders with the ultimate goal of preventing the further artificial spread of TSA into areas not known to be infested. This mechanism proved successful in the development of regulations that were effective, science-based and compatible with the impacted agribusinesses. Research findings led by the University of Florida’s IFAS were keys in the development of creative regulations that allowed the movement of livestock, sod, and other regulated articles.

In the case of livestock it was determined that ingested TSA fruit containing seed passed through the animal’s digestive tract in less than one week, therefore best management practices were developed to create TSA fruit-free pastures wherein livestock could be held for the required time for TSA seed to pass from their digestive system. Other integrated management strategies were developed to eliminate TSA from sod fields, grass seed harvest and composed manure operations.

Communication between the state departments of agriculture and the USDA's APHIS in the southeastern USA through the task force, was instrumental in developing and clearly understanding the regulatory framework needed to address the intrastate and interstate movement of TSA-regulated articles. This same framework should be strongly considered for use in future invasive pest issues of this nature.

Over time the research community has developed more effective TSA controls, including some excellent selective herbicides and a very effective biological control insect brought in from South America. These new controls that are now available have helped mitigate the impact of TSA on agriculture and in natural areas making regulation less challenging and ultimately making this unwanted invader a less onerous weed pest.

VII. A. Management Techniques: Biological Control of Tropical Soda Apple Using Insects

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Classical biological control has been defined as ‘The intentional introduction or importation of exotic natural enemies, usually co-evolved, for permanent establishment and long-term pest control’ (Van Driesche and Bellows 1996). Plants which are introduced on purpose or accidentally into a new region without their co-evolved natural enemies (herbivores and pathogens) may become invasive. The goal of a biological control program is to re-establish the invasive plant/natural enemies associations that keep the plant in check in its area of origin. Tropical soda apple (TSA), *Solanum viarum* Dunal (Solanaceae) is an invasive spiny brush native to southeastern Brazil, northeastern Argentina, Paraguay, and Uruguay that has invaded several southeastern states but it is more troublesome in Florida pastures and conservation areas (Medal et al. 2008). This plant belongs to the Solanaceae, a family that includes very important economical crops such as eggplant, tomato, potato, tobacco, pepper, and approximately thirty native *Solanum* spp. Plants, like TSA, which have so many closely related economically important and native plants, are difficult targets for biological control because of the challenge of finding specialized herbivores that feed and reproduce only on the target plant. Despite the difficulties of dealing with an invasive plant in such a taxonomic group, biological control of TSA is one of the most successful projects and the first attempt using insects as biocontrol agents of a *Solanum* weed in North America (Díaz et al. 2009, Medal 2005, Overholt et al. 2009).

The first exploration for natural enemies of TSA in South America was conducted by University of Florida (R. Charudattan, J. Medal, J. Mullahey), and Brazilian (R. Pitelli) researcher in June 1994 (Medal et al. 1996). South American insects identified as potential biological control agents of TSA include the defoliating leaf beetles, *Gratiana boliviana* Spaeth (Medal et al. 2007, 2002, 1996), *Gratiana graminea* Klug (Medal et al. 2010c), *Metriona elatior* Klug (Gandolfo et al. 2008, Medal et al. 1996), and the flower bud weevil, *Anthonomus tenebrosus* Boheman (Coleoptera: Curculionidae) (Medal et al. 2011). Host range determination studies with *G. boliviana* adults and first instar larvae were conducted from January 1998 to April 2000 at the Quarantine of the Florida Biocontrol Laboratory in Gainesville. One hundred twenty-three plant species in 35 families were included in the feeding and oviposition preference tests (choice and no-choice). The plants tested included 53 species in the Solanaceae (Medal et al. 2002). Open-field host specificity test exposing *G. boliviana* to the eggplant cultivar ‘Black Beauty’ in Argentina (Gandolfo et al. 2007), and surveys of unsprayed eggplant fields in Argentina, Brazil, Paraguay, and Uruguay corroborated that *G. boliviana* is not a pest of eggplants in South America. *Gratiana boliviana* was unanimously approved by TAG committee members in 2002, and USDA-APHIS-PPQ granted a permit for field release in Florida in the summer of 2003. In total, at least 250,000 beetles have been released in

42 Florida counties since summer 2003. The beetle became established in central and south Florida, and it is causing significant defoliations (30-100%), and reducing the fruit production of the target weed from 40-60 fruits per plant to zero or few fruits per plant. However, the beetle has not been able to establish in north Florida (Medal and Cuda 2010, Medal et al. 2010a, 2010b, Overholt et al. 2009, 2010).

Additional explorations in Brazil for natural enemies revealed several potential insects that may be suitable as biocontrol agents of TSA. These include an undescribed *Platyphora* species (Coleoptera; Chrysomelidae), and a leaf-root feeder, *Epitrix* probably *parvula*. (Medal et al. 1996, Olckers et al. 2002, Medal et al. unpublished data). .

There is no doubt, that biological control of TSA is having an important role in stressing and reducing fruit production and plant density in central and south Florida. Post-release evaluations have indicated that biological control is a key management tool that can be integrated with other available management techniques including pathogens, chemical herbicides, mechanical removal, and cattle/hay/sod movement regulations to reduce the TSA populations.

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VII. B. Management Techniques: Impact of *Gratiana boliviana* on Tropical Soda Apple

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Five studies have been conducted to measure the effect of *G. boliviana* on performance and population dynamics of tropical soda apple (TSA), and all strongly show that the beetle has significant negative impacts to TSA. The first two studies were conducted in Polk and Sumter Counties, and in each study twenty marked plants were monitored during almost two years. Both studies found that an increase in defoliation of TSA by *G. boliviana* was associated with a significant decrease in fruit production. The third study examined the performance of TSA at 113 locations in 38 Florida counties and found that TSA performance, as measured by plant height, canopy diameter, cover and number of fruit declined as damage due to *G. boliviana* increased. The fourth study demonstrated that plants protected from herbivore feeding with an insecticide performed far better than plants exposed to *G. boliviana*, and the final study revealed that beetles were capable of decreasing TSA density by as much as 90% within 2-3 years of their release. Details of these studies are summarized below.

Study 1. Medal and Cuda 2010

The first release of *Gratiana boliviana* adult beetles (F1 adult progeny born at the Gainesville quarantine facility from those collected in Paraguay) was made in three screened cages (40 adult beetles per cage) to allow them to build up their numbers. The cover of the cages were removed 13 weeks later (21 August 2003) to allow the natural dispersal of the 1,042 adult beetles that were recorded inside the three rearing cages in a densely infested 4 hectare patch of TSA in Polk County, Florida. Post release monitoring was conducted every 8-10 weeks from August 2003 until December, 2005. Twenty medium to large-sized TSA plants within 100 m of the release site were tagged and these plants were monitored for almost two years. The estimated visual defoliation increased from 46% in December 2003 to 94% in December 2004, and it was directly associated ($r = 0.75$) with the increase in number of adult and larvae beetles recorded on the TSA plants during the same period except from August to December 2004 when

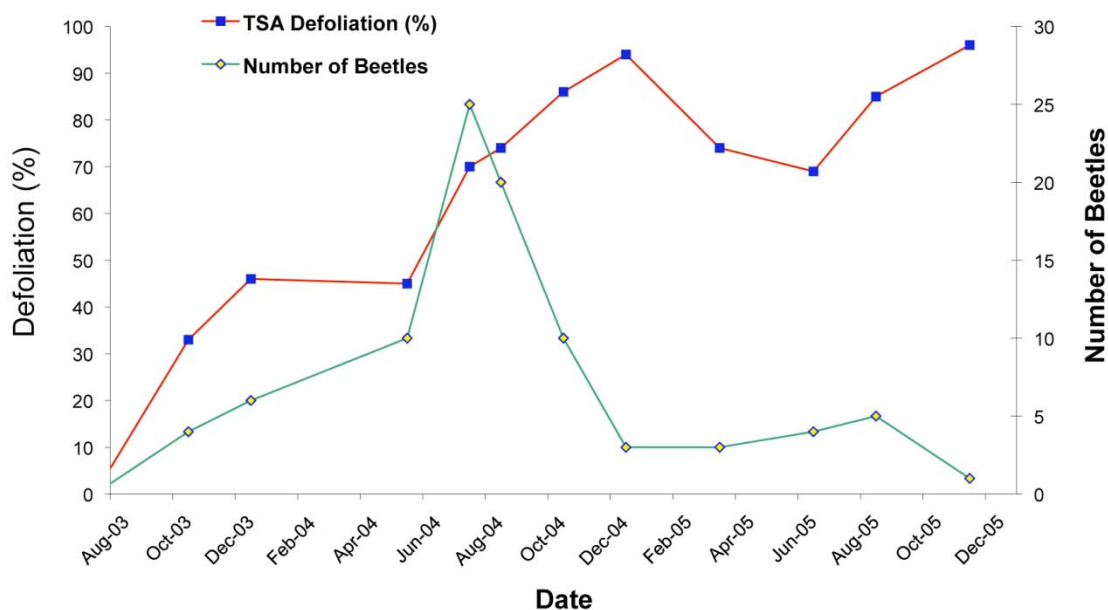


Figure 1. Mean defoliation (%) of 20 tropical soda apple plants and number of beetles per plant at the release site in Polk County, Florida (From: Medal and Cuda 2010).

the number of beetles per plant decreased (Figure 1). In 2005, the TSA defoliation averaged 69 to 96%. At least half of the 20 marked plants were not able to regrow after complete defoliation by the beetles in the previous year and also due to competition by other plant species growing nearby. There was a negative correlation ($r = -0.55$) between the level of TSA plant defoliation and number of beetles per plant during 2005 because the beetles had dispersed due to lack of food. Beetles showed a dispersal ability of approximately 1 mile per year during the evaluation period. The number of TSA fruit produced per beetle-defoliated plant significantly decreased, with none or very few small fruits produced compared with the large number of fruits observed during the summer of 2003 at the time the beetles were released (Figure 2).

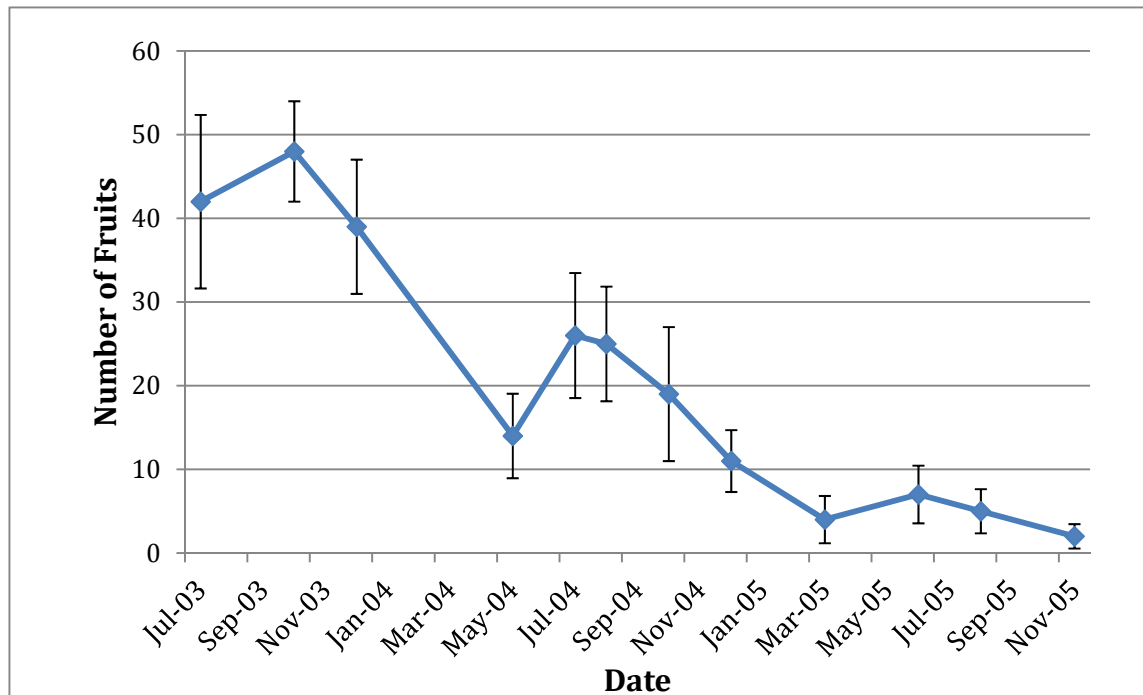


Figure 2. Mean number of fruits per plant on 20 tropical soda plants at the release site in Polk County, Florida following the release of *Gratiana boliviana* summer 2003.

Study 2. (Medal et al. 2010)

Gratiana boliviana adult beetles (500) were released in a densely infested 2 hectare patch of TSA in Sumter Co., Florida in the summer of 2005. The beetles were recently emerged from pupae (50% females, 50% males) at the University of Florida rearing colony located in Gainesville, Alachua County. These beetles were descendent from adults collected in Misiones province, Argentina in 2004. Post release monitoring was conducted every 8-10 weeks from February 2006 until October, 2007. On this release site, 20 medium to large-sized TSA plants within 100 m of the release site were tagged and these plants were monitored for 21 months. Defoliation increased from 20% in March 2006 to 75% in October of the same year, and from 10% in April 2007 to 90% four months later in August. Beetles numbers peaked in the early summers of 2006 and 2007, and declined during the winter (Figure 3). The number of fruit produced per TSA plant decreased from 36 in October 2006 to four in October 2007 (Figure 4).

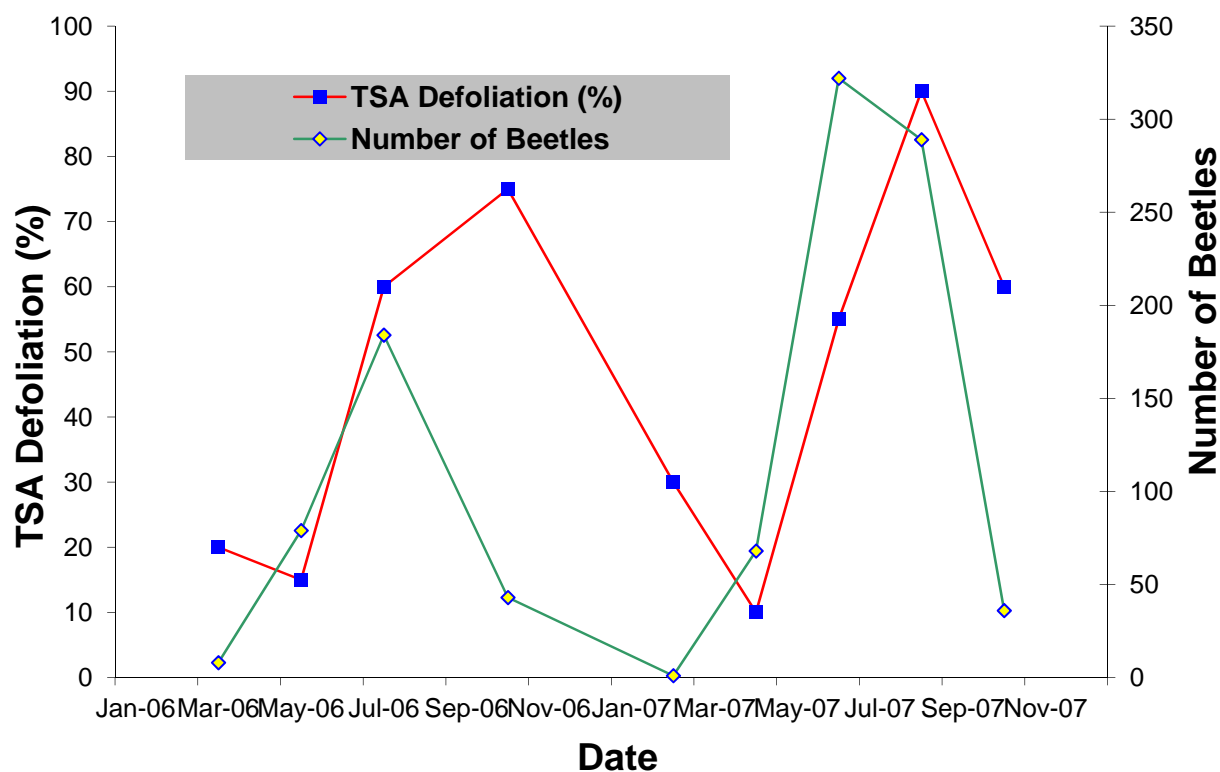


Figure 3. Mean defoliation (%) of 20 tropical soda apple plants and total number of *Gratiana boliviana* in Sumter County, Florida (From Medal et al. 2010).

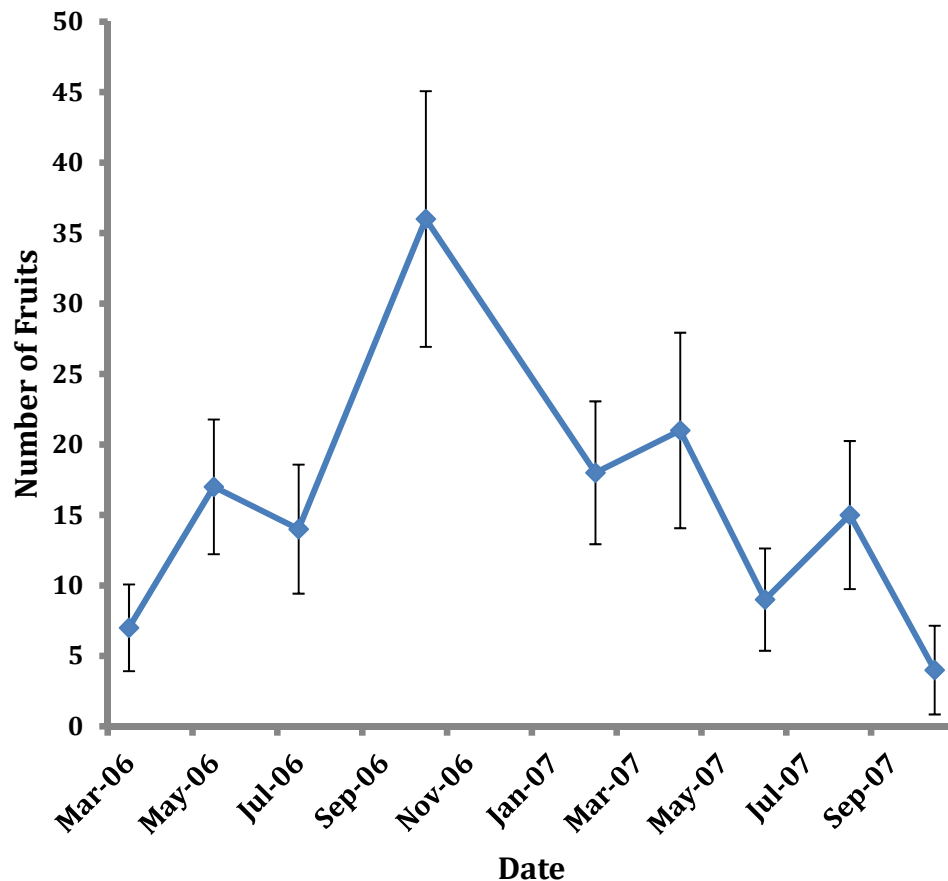


Figure 4. Mean number and standad deviations (bars) of fruits in 20 tropical soda apple plants in Sumter County, Florida (From: Medal et al. 2010).

Study 3. Overholt et al. 2009

In the fall of 2008, an extensive survey was conducted to estimate the distribution and abundance of *G. boliviana* in Florida, and to measure the beetle impact on TSA. A total of 113 randomly selected sites with TSA were surveyed in 38 counties (Figure 5). At each site, 10 plants were sampled by walking a straight line through the center of the infested area and selecting a plant every 5 steps. *Gratiana boliviana* was found at 48% of the surveyed sites, with average density of 3.2 beetles/plant. Beetles were present at 77% of sites between 26° and 27° latitude, 79% of sites between 27° and 28° and 54% of sites between 28° and 29°. No beetles were found at 32 sites surveyed north of 29° latitude. The northernmost occurrence of *G. boliviana* was at 28.8° in Seminole County near the town of Sanford. Plant height, diameter, cover and the number of fruit all decreased as damage due to beetles increased. Assuming that beetles arrived at survey sites from the nearest release site, the average distance beetles traveled per year since their release was

about 3 miles. An interpolated surface of beetle density indicates highest densities along a band running from approximately Naples on the west coast to Fort Pierce on the east coast, with a few areas of high density further north (Figure 6). The survey demonstrated that *G. boliviana* was firmly established in south/central Florida, and has spread from release sites. The lack of beetles north of 29° latitude may be due to asynchrony in seasonal phenologies of TSA and *G. boliviana*. Freezing temperatures do not usually kill TSA plants, but all above-ground parts die back (Mullahey et al. 1998). *Gratiana boliviana* enters diapause in the fall as day length decreases. If a freeze arrives prior to beetles entering diapause, the beetles would starve due to a lack of food. Similarly, increased abundance of TSA in the spring, through regrowth from root tissue and seed germination, occurs later in more northern areas of Florida than in the south, where plants may continue growing throughout the winter in some years. If *G. boliviana* diapause terminates before TSA increases in abundance in the spring, food would be scarce and population increase of beetles would be negatively affected.

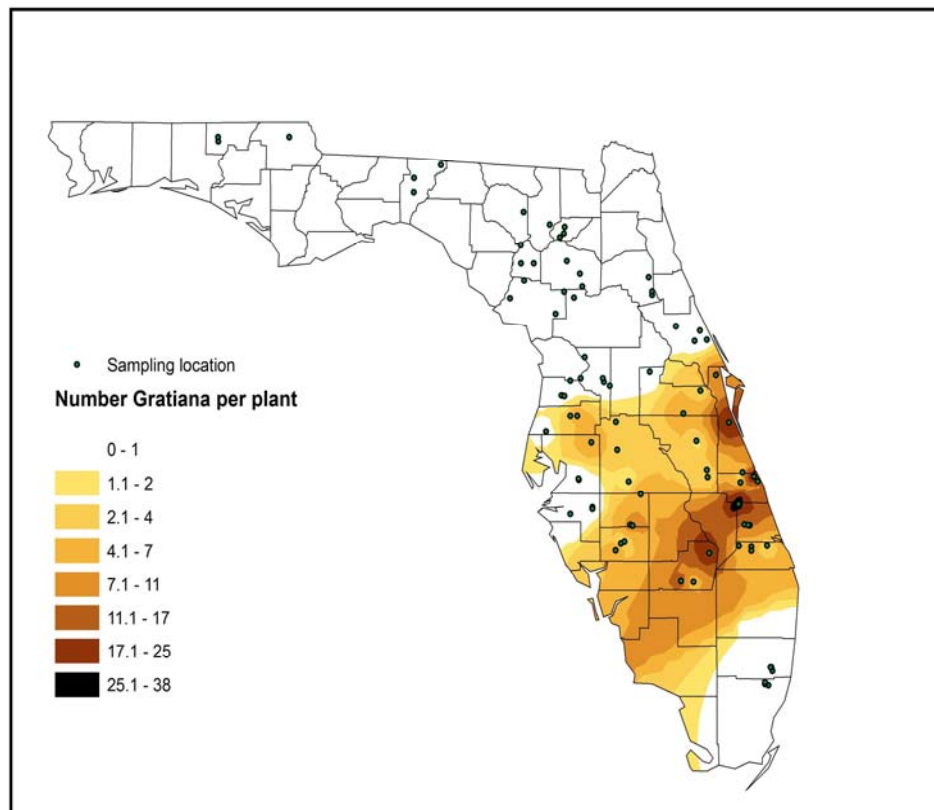


Figure 5. Sampling sites and interpolated density surface of *Gratiana boliviana* from data collected during a statewide survey in fall 2008.

Study 4. Overholt et al. 2010

A 2-year exclusion study was conducted on a ranch in Saint Lucie County, Florida from March to October in 2008 and 2009. Exclusion studies are designed to measure the performance of plants protected from insect herbivores and unprotected plants. Plots of TSA were in a partially shaded hammock area of the ranch, and divided into sub-plots which were either protected with a soil drench application of the insecticide imidacloprid (Admire® 2F), or left unprotected. In both years of the study, TSA plants protected by insecticide were taller, wider, and had greater canopy cover than unprotected plants. The number of fruits on protected plants was higher on most sampling dates than on unprotected plants in 2008 and 2009 (Figure 6). Survival of plants was higher in plots protected with insecticide than in unprotected plots during both years of the study (Figure 7).

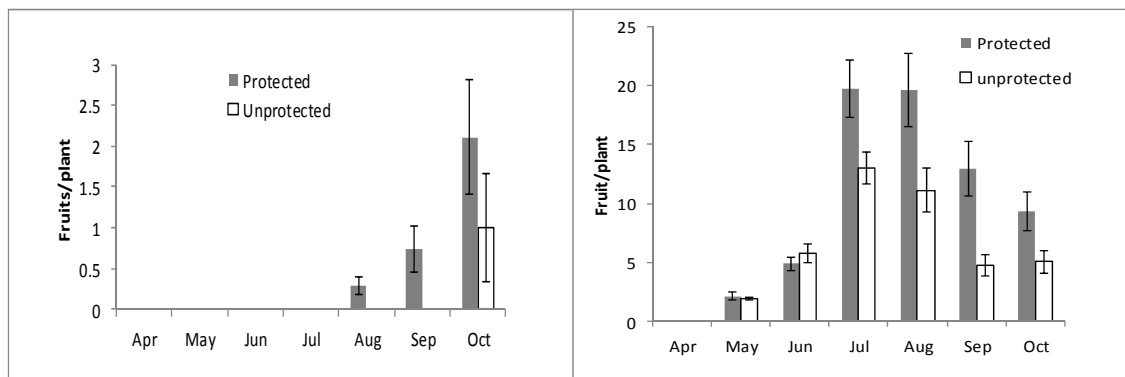


Figure 6. Fruit production of unprotected and insecticide protected TSA plants in 2008 (left) and 2009 (right).

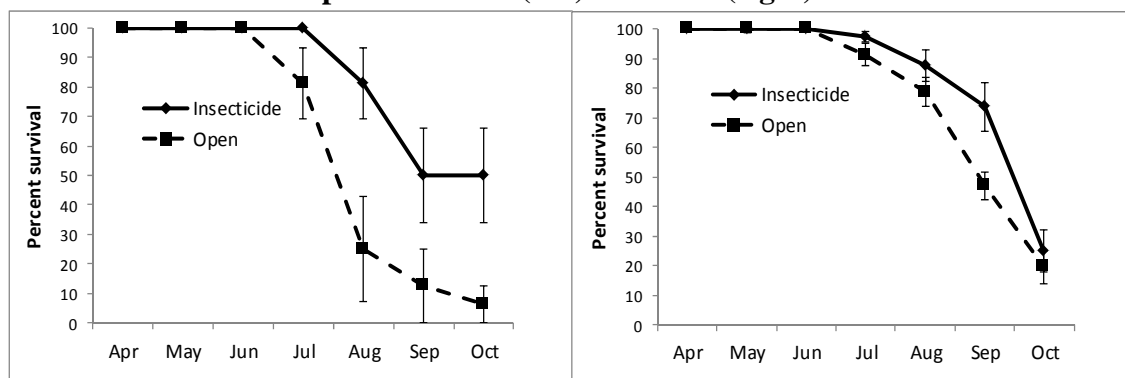


Figure 7. Survival of unprotected and insecticide protected TSA plants in 2008 (left) and 2009 (right).

Study 5. Overholt et al. 2010

The population dynamics of *G. boliviana* and TSA were monitored for 40 months at four locations on a ranch in St. Lucie County, Florida (figure 8). A preliminary survey

found no *G. boliviana*, or plants which exhibited characteristic signs of *G. boliviana* feeding, on the ranch. TSA was almost entirely restricted to hammock areas due to control by mowing in open pastures. Thus, four hammock areas along a northwest to southeast axis were selected as study sites. On 7 June 2006, 800 adult *G. boliviana* were liberated in Release site 1 and 350 in Release site 2, with the number of beetles released proportional to the area of each hammock. Two other hammock areas (Control sites 1 and 2) were selected as controls, with the closest being 1,640 m from the nearest release site, and the second control site 764 m further away. Every three months from July 2006 to October 2009, the four areas were sampled. The two control sites were quickly compromised as beetles were found at both sites during the second sampling in October 2006, indicating that the beetles had moved at least 1.64 km in 4 months. If we assume that all long distance movement occurs during an eight month period between March and October when beetles are active, then the annual distance beetles could have moved was at least 3.28 km. The dynamics of TSA populations differed between the four sites (figure 8). At Release site 1, which had a much higher density of TSA at the beginning of the study than the other three sites, the TSA population declined approximately 90%, with most of the decrease occurring during the first 2 years. At the other three sites, the TSA density was initially low, and remained more or less stable during 3 years. These results show that *G. boliviana* was able to regulate TSA at an average density of approximately 1 plant/4 m², and that the beetles were able to reduce TSA to this density in about 2 years. TSA density tended to be lowest in October, increased during the winter, and then declined during summer months. We hypothesize that this seasonal dynamic is due to a release of plants from herbivore pressure during the winter when *G. boliviana* is in diapause. *Gratiana boliviana* population density increased in the spring, and was typically highest during the summer.

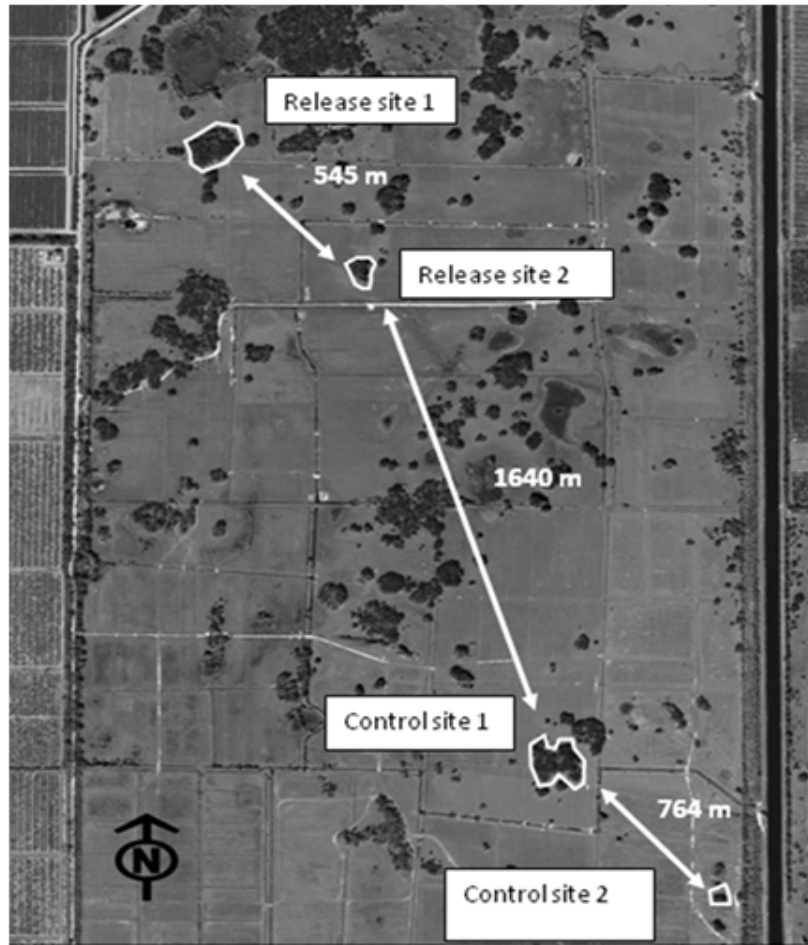


Figure 8. Aerial photograph of the area where the population dynamics study was conducted in western St. Lucie County Florida.

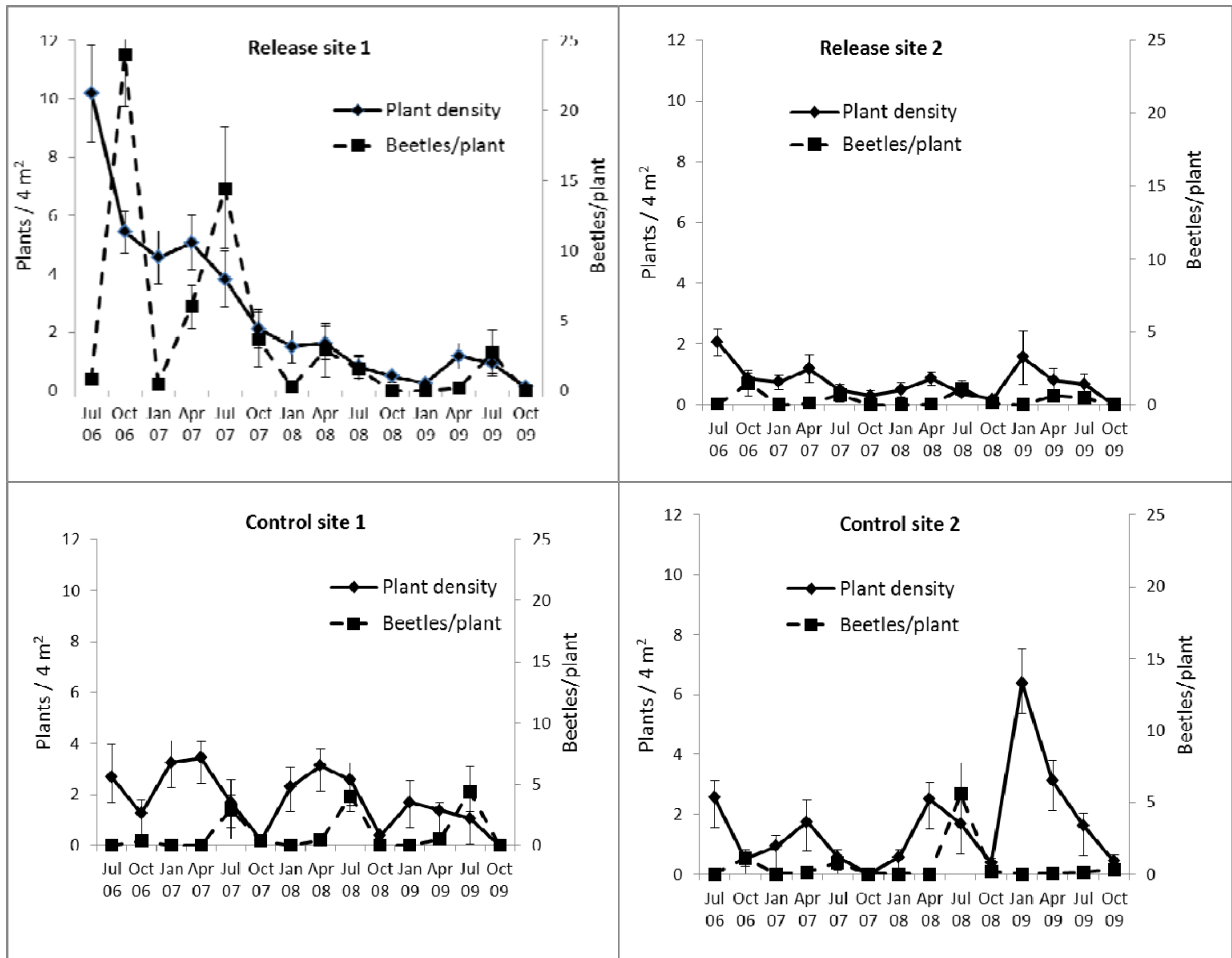


Figure 9. Density of tropical soda apple and *Gratiana boliviana* during a 40 month period in four hammocks in western St. Lucie County, Florida.

VII. C. Management Techniques:

Biological Control of Tropical Soda Apple North of Florida using *Gratiana boliviana*

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Tropical soda apple (*Solanum viarum* Dunal) is a noxious weed that is known to infest pastures and natural areas throughout the southeastern United States. From the first known collection site in central Florida, the weed has spread throughout Florida and into Alabama, Georgia, Mississippi, North Carolina, Pennsylvania, Puerto Rico, South Carolina, Tennessee, Texas, and West Virginia (Figure 1) (NAPIS 2011). TSA is most invasive in open pastures (Mullahey et al 1998). In Mississippi, Alabama, and Georgia, TSA invades oak hammocks, cypress heads, and wooded areas. In Texas, the weed occurs in pastures, native grasslands and forested areas. To date, little has been published about TSA's plant associations in the other states.

Introduction of TSA was most likely from Brazil and is thought to have occurred only into Florida (Kreiser et al. 2004; Mullahey et al. 1993). Various avenues of dispersal have spread TSA throughout Florida and into other states (Mullahey et al. 1998). Cattle are known to consume the fruit when feeding in contaminated areas and the movement of such livestock likely resulted in new infestation sites. Hay harvested from areas infested with TSA can spread the fruit and seed when sold and moved to new areas. Also, other types of seed (such as Bahiagrass, Bermudagrass, clover, etc.) harvested from TSA infested locations, can be contaminated and may have moved the weed to new areas. Once the weed is established in a new area, additional movement of the weed seed is facilitated via the digestive systems of livestock and wildlife that consume the fruit, contaminated equipment, hay, seed, sod, and composted manure.

In tropical climates, TSA is a perennial shrub that blooms and produces fruits throughout the year (Patterson et al. 1991). In Florida, peak flowering and fruit production occurs from September through May (Mullahey et al. 1993). In contrast, TSA bloom and fruit production in other southern states is limited by frost. North of frost free zones, TSA starts flower and fruit production in June and peaks in late August (Bryson and Byrd 2007). Similarly plant growth and survival of overwintering seed is affected by the severity of winter conditions. Field tests indicated that TSA can survive as a perennial at winter temperatures above 10 °C and may survive colder temperatures but as an annual (Bryson and Byrd 2007). In Mississippi, TSA was found to persist through mild winters, resulting in higher population levels the following spring. With colder winters, TSA can function as a facultative annual when seedling emergence is early enough for plants to produce fruit before frost. Unlike central Florida, the thick stands that can be impenetrable to livestock, large wildlife, and humans are rare in northern locations. Dense stands of TSA have appeared after the soil has been disturbed through clear cutting

TSA infested wood lots and introducing livestock. However, when dense stands appear, they tend to be short lived and the plants disappear from the patch within a year. Small patches of TSA will often persist in small patches in the neighboring area.

The impact of weather conditions on the persistence of the weed and density of the stands likely influences current management practices. In most states outside of Florida, eradication programs are being actively pursued (Figure 1). Management of the weed in these areas focuses on prevention, mechanical, and chemical practices. To prevent the interstate movement of TSA, southern states have developed voluntary compliance agreements that outline regulations to insure products from areas with TSA are free of weed seeds. These agreements cover the movement of livestock, seed, plant products (including seed, sod and hay), soil, and manure. Mowing is regularly used to prevent seed production by destroying flowers and early fruit development. Plants with mature fruits are cut, piled, and burned to destroy seed viability, or buried. Herbicides that possess post-emergence control of existing plants and pre-emergence control of germinating seeds are used on both large stands as well as sparse infestations. In contrast, central Florida management strategies have migrated towards biological control. The TSA leaf beetle, *Gratiana boliviana* Spaeth, has been found to be an effective biological control agent in reducing stand density and fruit production (see previous chapter).

Some states, such as Georgia, have stopped wide-scale herbicide applications due to funding restrictions, while other states, such as Alabama, have continued a vigorous herbicide campaign and are now looking at ways to reduce the weed in areas difficult to spray. Because biological control showed promise in reducing plant growth and fruit production, as well as providing control in natural or non-chemically treated areas, a program was initiated in 2004 to test the beetles' effectiveness in Alabama, Georgia, and Texas. Several areas were identified in each of the three states to serve as initial release sites of *G. boliviana* to determine if the beetle would overwinter and potentially serve as field insectaries. The selected stands of TSA occurred in various habitats, including pastures and woodlots, in order to determine the types of habitats in which the beetle could become established. At the release sites, stand density and fruit production were documented to determine the impact of the beetle. Beetles were produced in Miami, Florida, shipped to the states on a regular basis, and released from 2004-2008 (Table 1). Beetles reproduced at all locations, caused feeding damage on TSA plants, and overwintered (Table 1). Two years after the last release, beetles were recovered at sites in Alabama and Georgia. Despite surviving multiple winters where temperatures reached below freezing, beetle numbers failed to increase during subsequent warmer months after the two-year initial establishment period. Possible explanations for lack of persistent establishment include sparse and widely distributed patches of TSA, low winter temperatures, prolonged low temperatures, and predation pressures. Laboratory studies on the effects of temperature on the development of *G. boliviana* revealed that states north of Florida were beyond the limit of the insect's range (Díaz et al. 2008). However, TSA stand density and fruit production decreased at all study sites where the biological control agent was released. Although TSA plants could be found in adjacent areas, there

were no signs of beetle presence. To date, it is not known if the beetles had direct influence on causing the decline of TSA in the release areas. Perhaps the stress of beetle feeding reduced the ability of the plants from regenerating from roots. However, cooler winter temperatures also could have prevented root regeneration.

TSA continues to be a major weed affecting pasture lands throughout southern United States. Additionally, eradication programs will always face the continuous threat of seed coming from areas with established populations of the weed. The effect of winter temperatures on both the plant and the current biological control agent will likely influence management strategies. Best management practices need to account for the potential that root stock and seed will survive winters north of the frost-free areas. The benefit of using biological control agents on this weed is well documented. However, the effective agent, *G. boliviana*, may not be suitable for areas experiencing prolonged below freezing temperatures. Foreign exploration is needed that focuses on areas that are climatically similar to areas north of the frost free zone in order to find potential biological control agents that can survive these environmental conditions. Future efforts could also explore if control temperature rearing programs could be used to select for *G. boliviana* that would be more tolerant of conditions found north of central Florida.

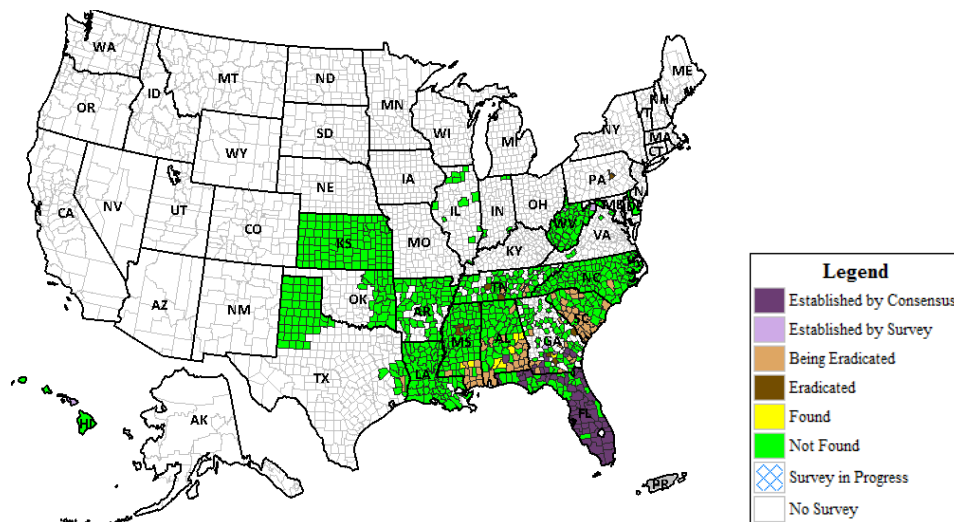


Fig. 1: Current distribution of tropical soda apple in the United States (National Agriculture Pest Information System assessed Aug. 2011).

Table 1: The number of tropical soda apple leaf beetles (*Gratiana boliviana*) released outside of Florida and the duration that the beetles were found at the locations.

| Site | Location | GPS | Habitat Type | Number Released | | | | | Establishment Duration |
|------------------------|-----------------|--------------------------|--------------------|-----------------|------|------|------|------|------------------------|
| | | | | 2005 | 2006 | 2007 | 2008 | 2009 | |
| Kenny's Farm | Coffee Co, AL | N31.23815 W86.09565 | small pasture | 2650 | 2350 | 0 | 0 | 0 | 2005-2007 |
| Brook's Farm | Geneva Co., AL | N31.15618 W86.06380 | over grown pasture | 2650 | 2350 | 1850 | 0 | 0 | 2005-2007 |
| Maplesville Site | Chilton Co., AL | N32.71036 W8691058 | small pasture | 0 | 3300 | 3000 | 0 | 0 | 2006-2009 |
| Herd Farm - Trail Site | Decatur Co., GA | N31.064133 W84.518983 | woodlot edge | 2600 | 1000 | 0 | 0 | 0 | 2005-2006 |
| Herd Farm - Pond Site | Decatur Co., GA | N31.06711 W84.51784 | Pasture | 0 | 1000 | 5250 | 0 | 0 | 2006-2007 |
| 40-Acre Private Ranch | Jasper Co., TX | N30.92019 W93.99660 | pasture with woods | 0 | 0 | 175 | 450 | 0 | 2007-2009 |

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VII. D. Management Techniques: Biology and Ecology of *Gratiana boliviana*

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Gratiana boliviana was introduced from South America into the southeastern United States in 2003. *Gratiana boliviana* has four developmental stages (egg, larva, pupa, adult) (Figure 1). Eggs are brown and enclosed in a membranous envelope, larvae are spiny and pale green, pupae are spiny, flattened and immobile, and adults are about ¼” long and nearly as wide, and a deep green color. Adults and larvae usually feed on the upper side TSA leaves, while pupae and eggs can be found on the underside of leaves, eggs can be found also on the leaf petiole/stems. The life cycle of the beetle begins when a female lays an egg.

Temperature-dependent development and survival studies revealed that *G. boliviana* can complete development at temperatures between 16°C and 34°C. The number of degree-days required to complete one generation was 341 and the estimated lower developmental threshold was 13°C. The upper lethal threshold was estimated to be 34-35°C. Cold tolerance studies revealed that the lethal time for 90% of adults (LT90) was 13 days at 5°C and 9 days at 0°C. Based on the developmental and cold tolerance data, a map predicting the areas of establishment and number of generations per year was generated, which suggests that the northern extent of the *G. boliviana* range in the USA will be near 32-33° north latitude (Figure 2). (Diaz et al. 2008). However, evidence from the field indicates poor establishment north of 29° (see chapters VII B and VII C).

In central Florida, the beetles actively feed and reproduce in the field from around March/April until October/November, which is probably sufficient time to complete about 7-8 generations. However, for 4-5 months during the winter, the beetles enter an adult resting state called ‘diapause’. The timing of diapause allows the synchronization of insect herbivores with the phenology of their host plant. During diapause, the beetles feed very little, and do not reproduce (Diaz et al. 2011). They are difficult to find at this time of year, as they hide in leaf litter beneath plants. Diapausing beetles are brown from November to March and remain mostly in the leaf litter (Fig. 3). Once the days become longer and warmer, adults turn green and move from the leaf litter to TSA leaves (Figure 3). Eggs are found from late March through October. Larvae and pupae are common from April to November (Figure 4).

Several natural enemies of *G. boliviana* have been identified in Florida, but there is no information on the distribution and impact of these enemies on the population of the beetle. While their impact in large field infestations might be negligible, natural enemies could have greater impact on concentrated populations found in rearing facilities. A preliminary list of predators, parasitoids and diseases is presented in Table 1.



a



b



c



d



e



f

Figure 1. Life cycle of *Gratiana boliviana*. a) egg, b) larva, c) pupa, d) adult, e) male (note orange testis) and f) reproductive female.

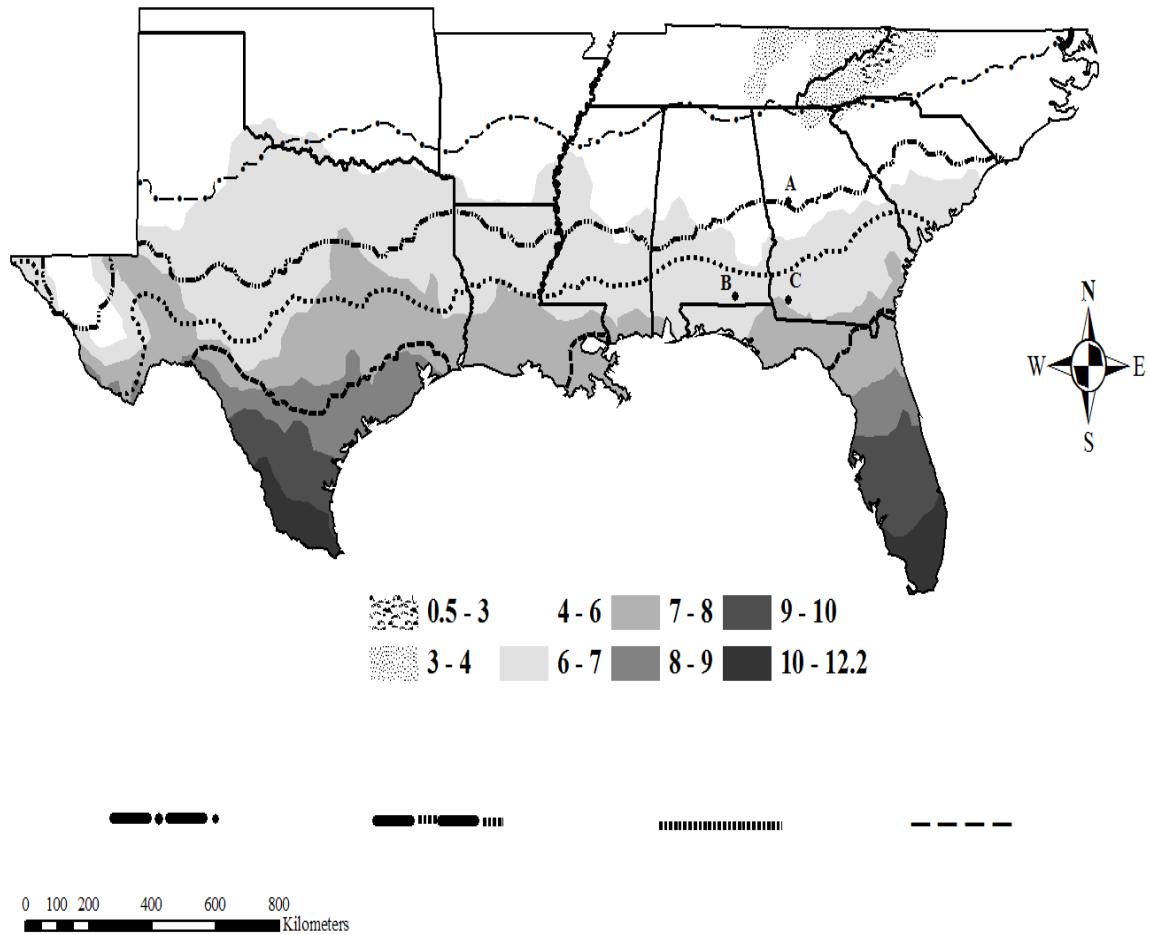


Figure 2. Map showing the predicted number of generations of *Gratiana boliviana* in the southeastern United States, and LT50 and 90 lines at 0 and 5°C. Points A, B, and D are release sites in Chilton, Alabama, Coffee Co., Alabama and Decatur Co., Georgia, respectively, where *G. boliviana* is known to have overwintered. Point C is Meriwether Co., Georgia where *G. boliviana* failed to establish.

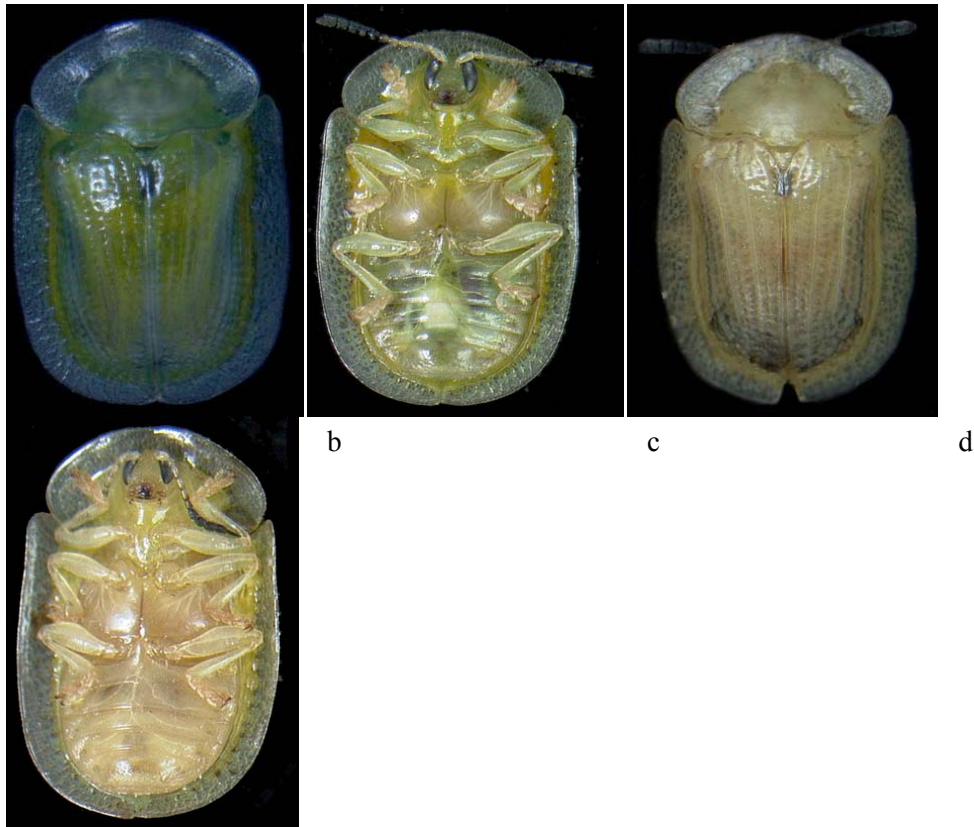


Figure 3. *Gratiana boliviana* adults: a) reproductive dorsal b) reproductive ventral, c) diapause dorsal, d) diapause ventral.

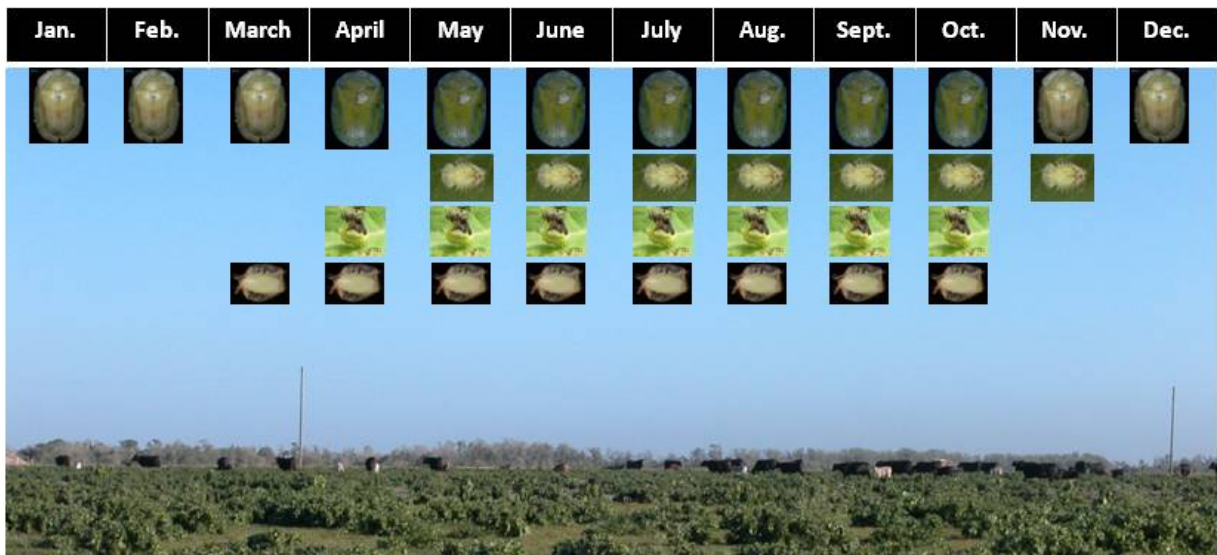


Figure 4. Schematic representation of the phenology of *Gratiana boliviana* in Florida.

Table 1. Natural enemies of *Gratiana boliviana* found in Florida.

| Organism | Type |
|--|------------|
| Spiders | |
| <i>Coleosoma acutiventer</i> | predator |
| <i>Peucetia viridans</i> | predator |
| <i>Acanthepeira</i> sp. | predator |
| <i>Misumenops celer</i> | predator |
| <i>Acanthepeira stellata</i> | predator |
| <i>Cheiracanthium inclusum</i> | predator |
| <i>Misumenops</i> sp. | predator |
| <i>Oxyopes salticus</i> | predator |
| Insects | |
| Orthoptera: | |
| <i>Oecanthus</i> sp. | predator |
| Hymenoptera: | |
| <i>Crematogaster</i> sp. | predator |
| <i>Eupelmus</i> sp. | parasitoid |
| <i>Aprostocetus</i> near <i>cassidis</i> | parasitoid |
| <i>Conura side</i> | parasitoid |
| Hemiptera: | |
| <i>Sinea</i> sp. | predator |
| <i>Stiretrus anchorago</i> | predator |
| Entomopathogens | |
| <i>Nosema</i> sp. | disease |
| <i>Mattesia oryzaephili</i> | disease |
| Short gram negative bacteria | disease |

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VII. E. Management Techniques: Field Release Techniques for *Gratiana boliviana*

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Objective

The objective of the beetle release program was to get the beetle, as quickly and with as little trauma to the beetle as is practical, to a site infested with tropical soda apple (TSA) and to get the beetle well distributed in the parts of Florida where TSA is present. The most practical method is to collect active adults, usually from a rearing facility, transport them as quickly as practical to a TSA infested site and release them in the shortest time possible. If the release site is fairly close to the rearing facility the beetles can be transported by vehicle, but if that is not practical the beetles are usually sent via commercial overnight shipment to the person who will be doing the release.

Release Site Selection

Selection of a release site is important. Sites should have a good amount of healthy TSA so there will be an adequate food supply for the beetles. Whenever possible releases should be made near the center of the TSA infestation. Don't release only one or a few beetles per plant; rather release 10-20 beetles per plant on plants that are fairly close together. The objective is to establish a breeding, growing population in a particular area that can then spread to nearby TSA. Avoid releasing beetles on TSA plants that will be disturbed in the near future by mowing or application of herbicide. Also avoid placing beetles on TSA plants that could be damaged by flooding or drought. While TSA grows in both shaded and open areas, and the beetle will feed on the plant in both conditions, it is recommended to release the beetles in shaded areas if possible. Shaded areas are cooler and newly released beetles can adjust better in these conditions. As they become acclimated to the area they will move out to TSA in sunny areas. Also, TSA in shaded areas such as under trees and in hammock areas is less likely to be disturbed by mowing, spraying or damaged by frost and will therefore be a better long term food source for beetle establishment.

Time of year to release

Beetles can be released any time during their active period (April to October), but earlier in the year is better, provided there is good re-growth of TSA plants after winter cold damage, as it will give the beetles more time to reproduce and establish a good population before entering diapause (a resting stage) in the late fall.

Moving beetles to release site

Beetles are often shipped in 32 oz. plastic containers with screened tops. Normally there are 100 to 150 beetles per container. Some fresh TSA leaves should be

placed in each container to serve as a food and moisture source and a place for the beetles to cling to during transport. A small container of ice is often placed in the base of the insulated shipping container to keep the beetles from becoming over heated. When the beetles arrive at the release site most of the beetles will be either clustered on the top of the container clinging to the screen or down below clinging to the leaves. If the beetles are warm, those on top they will often fly out on their own when the container top is removed. Those clinging to the leaves can be released by placing the leaves on TSA plants in the field. This is less traumatic to the beetles and they will disburse on their own from there.

Documentation of Release

In order to monitor the distribution of the releases of the beetle in the state of Florida, some basic information is taken at each release site and placed on a standard form developed for the project (shown below). This information is later entered into a state-wide data base maintained by the University of Florida to help record and coordinate the field releases as well as give an overall view of distribution of the beetle.

Gratiana Release Record

| | | |
|---|--|---|
| <p>Date of Release: _____</p> <p>Name of Releaser: _____</p> <p>Title: _____</p> <p>Organization: _____</p> <p>Work Phone: _____</p> <p>E-Mail: _____</p> <p>Property Owner: _____</p> | | |
| Origin of insects: DPI, Ft. Pierce | | Life stage(s) released: |
| Number released : | | |
| <p>Location of release:</p> <p>County: _____</p> <p>Latitude _____</p> <p>Longitude _____</p> | | Directions to site: |
| <p>Land use:</p> <p><input type="checkbox"/> ranch</p> <p><input type="checkbox"/> park</p> <p><input type="checkbox"/> conservation area</p> <p><input type="checkbox"/> residential</p> <p><input type="checkbox"/> other</p> | | |
| <p>Habitat type:</p> <p><input type="checkbox"/> open pasture</p> <p><input type="checkbox"/> pine flatlands</p> <p><input type="checkbox"/> hardwood hammock</p> <p><input type="checkbox"/> other _____</p> | | <p>Tropical soda apple density:</p> <p><input type="checkbox"/> very high (81-100% cover)</p> <p><input type="checkbox"/> high (61-80% cover)</p> <p><input type="checkbox"/> moderate (41-60% cover)</p> <p><input type="checkbox"/> low (21-40% cover)</p> <p><input type="checkbox"/> very low (0-20% cover)</p> |

VII. F. Management Techniques: Control of Tropical Soda Apple by Using a Plant Virus as a Bioherbicide

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Introduction

Tropical soda apple (*Solanum viarum* Dunal, Solanaceae, abbreviated as TSA) is extremely susceptible to Tobacco mild green mosaic tobamovirus (common name: tobacco mild green mosaic virus, TMGMV), a mechanically transmitted virus with no known insect vectors or other natural modes of transmission. TSA plants inoculated with TMGMV are killed quickly and consistently. Through carefully planned and executed research at the University of Florida, it has been determined that TMGMV could be developed and used as a biological herbicide to control TSA (Charudattan et al. 2003, 2004, 2009; Charudattan and Hiebert 2007). TMGMV kills TSA by eliciting a lethal hypersensitive response from the plant. This response is highly host-specific as shown by the fact that among plants that are susceptible to TMGMV, only TSA is killed totally and in a characteristic and predictable manner with the appearance of hypersensitive necrotic lesions on the leaves (Figure 1) followed by wilting of the plant and death (Figures 2 and 3). TSA plants of all ages are killed in about two to five weeks following inoculation with the virus. Younger plants are killed sooner than older plants but the final level of weed kill is generally the same (Pettersen et al. 2000). All parts of the plant are killed excluding mature fruits but including roots; the root death precludes any regrowth.

TMGMV is a member of the plant virus Family Tobamoviridae, Genus *Tobamovirus* and occurs naturally in Florida and many other U.S. states. It occurs worldwide in susceptible *Nicotiana* species (wild and cultivated tobaccos), particularly *N. glauca* (tree tobacco) but is not known to cause significant economic losses in susceptible crops such as tobacco. Through greenhouse and field studies conducted over a decade, it has been established that TMGMV could be used as an effective and safe bioherbicide to control TSA.

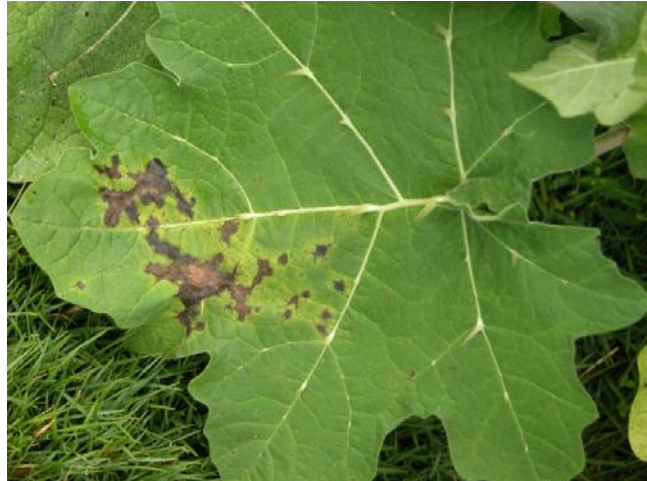


Figure 1. Hypersensitive necrotic lesions elicited by TMGMV in an inoculated TSA leaf. These lesions are often the clear sign of TSA's response to TMGMV infection.

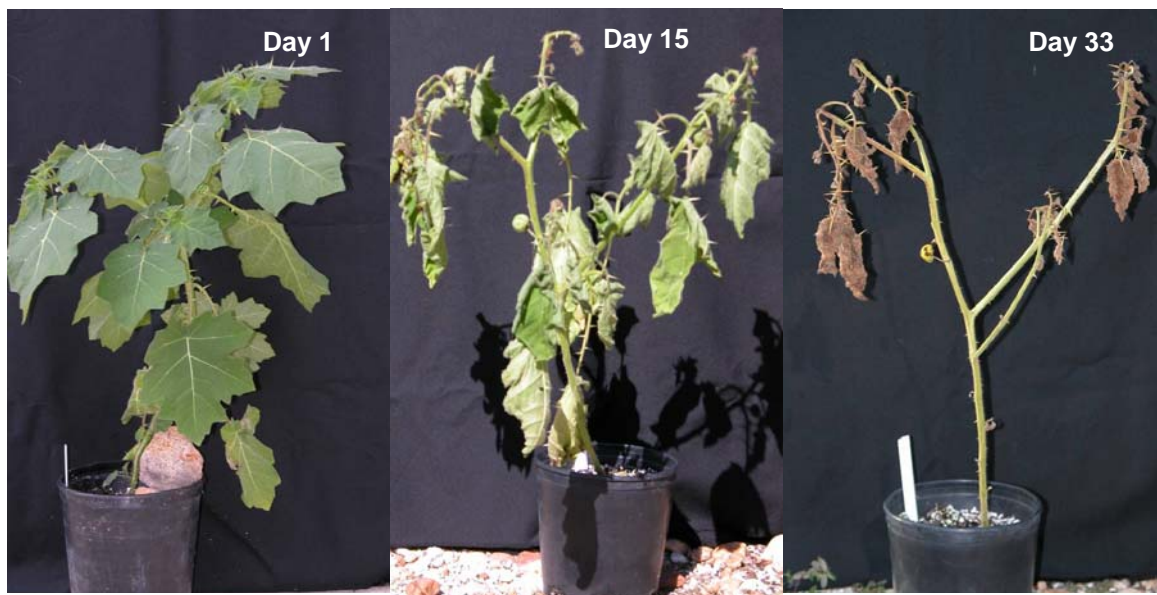


Figure 2. Progression of reaction of a TSA plant manually inoculated by rubbing five young leaves with an aqueous preparation of TMGMV. Left: inoculated plant on day 1 and the same plant on day 15 (center) and day 33 (right) following inoculation.



Figure 3. Top row: TMGMV-inoculated and non-inoculated healthy TSA plants in the field. The inoculated plants are dead in the picture on the left; they are yellow, wilting, and dying in the picture on the right. The lack of virus spread from the infected to the nearby healthy plants is evident in these pictures. Bottom row, left: Field plots planted with TSA, before inoculation with TMGMV. Bottom row, right: The same plots with TMGMV-inoculated (dead) and control plants (green and living) from a field study. TMGMV provided nearly 100% TSA control; the green areas in the two plots with dead TSA are grasses and other weeds.

Tobacco mild green mosaic virus

Discovered nearly 60 years ago, TMGMV is a well-studied pathogen of tobaccos (*Nicotiana* spp.) and about 25 other plants. TMGMV was first described as a mild strain of Tobacco mosaic tobamovirus (tobacco mosaic virus, TMV) that is distinguished by its mild mosaic symptoms in tobacco compared to the common TMV strains. It is now classified as a separate *Tobamovirus* species, Tobacco mild green mosaic tobamovirus (ICTV Decimal Code 71.0.1.0.011) consisting of two naturally occurring strains, U2 and U5, that are distinguished by the size of their 3' n-terminal regions. The Florida isolate of TMGMV that is pending registration as the bioherbicide has biological properties (host range) and genomic sequence (NTR) that are consistent with those of the U2 strain (ICTVdB Descriptions, 2011, available online: <<http://www.ictvdb.rothamsted.ac.uk/ICTVdB/00.071.0.01.011.htm>>).

TMGMV is a pathogen principally of plants in the Solanaceae because the majority of the TMGMV-susceptible plants are in this family while a few are in other unrelated families. Tomato, an economically important member of the Solanaceae, is not susceptible to TMGMV, but to a different *Tobamovirus*, namely Tomato mosaic tobamovirus (tomato mosaic virus, ToMV), which is common in tomato worldwide. Typically, even species that are susceptible to a plant virus may consist of resistant and immune varieties, as is the case with TMGMV. For example, the cultivated tobacco (*Nicotiana tabacum*) and pepper (*Capsicum* spp.) that are considered susceptible to TMGMV include susceptible, resistant, and immune varieties. TMGMV, like its closely related TMV, can be a common contaminant in vegetatively propagated plants, such as ornamental plants grown from cuttings in a nursery setting where the plants become infected from repeated handling by workers who consume tobacco products, use contaminated equipment, etc. However, the number of natural hosts to TMGMV is much smaller than the number of hosts reported from artificial inoculations and situations.

Safety features

Host-range and negligible risks to nontarget plants:

To determine the potential risks to nontarget plants, the host reaction of 435 plants in 61 families representing 183 genera, 311 species combinations, and 435 cultivars were tested. As required for bioherbicide registration, these data were gathered from repeated, replicated trials done under controlled conditions (Charudattan et al., unpublished). The data thus obtained corroborated and expanded previous reports (Plant Viruses Online, 2011) that TMGMV is a pathogen adapted to the Solanaceae; nearly all of the susceptible plants are in this family whereas about 98% of non-solanaceous plants tested were insusceptible to TMGMV. Only two hosts are of concern: peppers (*Capsicum* spp.) and tobacco (*N. tabacum*) but because TMGMV has no natural means of dispersal other than by chance physical contact, the probability of risk to these crop plants is remote. Furthermore, Charudattan et al. (unpublished) have established that susceptible tobacco and pepper cultivars could be grown next to TMGMV-infected TSA plants or in the same field site that had a crop of TMGMV-infected tobacco used for TMGMV mass-production without any adverse effects.

Lack of risks to humans and the environment:

As a plant virus, TMGMV does not infect humans, animals, or insects and does not pose any risks to these higher life forms. TMGMV does not pose any toxicological concerns regarding aquatic organisms nor does infection trigger toxin production in plants. It is fair to say that humans routinely consume fruits and vegetables carrying plant viruses with no ill effects.

Limited environmental persistence:

In greenhouse and field studies, TMGMV could not be detected in test soils or roots of TMGMV-killed TSA plants 6 months after completing a study by using a sensitive biochemical assay (enzyme linked immunosorbant assay, ELISA), highly host-specific plant-inoculation tests, and spectrophotometric measurements with a detection limit of 0.1 µg per ml of the virus. Hence, it is likely that TMGMV degrades to a level below the detection limit (Charudattan et al. unpublished).

Infective TMGMV was recovered from recently fallen, completely desiccated TSA leaves but not from TSA stems that had been dead for 6 months (Charudattan et al. unpublished). Coupled with the fact that a susceptible pepper crop could be grown in the same field site following a TMGMV-infected susceptible tobacco crop (as mentioned above), it is clear that the risk of persistence of TMGMV in field soil is minimal or negligible.

In a study intended to determine the infectivity of purified TMGMV stored at room temperature in three types of water, the virus remained infective for up to one year when stored in sterile deionized water or tap water. The infectivity was lost when stored in water from Lake Alice, Gainesville, a natural lake. Thus, TMGMV is not likely to persist in an infective state in natural waters.

TMGMV can be inactivated by ultraviolet light (Siegel and Wildman 1956; Siegel et al. 1956; Note: TMGMV is referred to in these papers by its older name TMV U2) and high temperatures (85-90°C) (ICTVdB Descriptions, 2011, available online: <<http://www.ictvdb.rothamsted.ac.uk/ICTVdB/00.071.0.01.011.htm>>). In addition, tobamoviruses gradually lose their infectivity and recoverability from soil, the rate of degradation in soil being a function of temperature, moisture, and pH (Allen 1984; Broadbent et al., 1965; Cheo 1980). Temperatures above 24°C cause faster degradation than lower temperatures. Degradation is hastened in moist soil compared to drier soil or flooded soil, although soil dehydration can also accelerate virus degradation. Virus activity in soil is greatly reduced when soil pH changes from 7.0 to 4.9. Therefore, it is likely that TMGMV deposited on soil and grass from spray applications would not survive for prolonged periods.

Populations of tobamoviruses in general and TMGMV in particular are genetically stable, as evidenced by the low frequency of emergence of new strains in nature. Mutations that occur naturally are likely to be deleterious to the fitness, virulence, and survival of the mutants in populations. Fraile et al. (1996, 1997), Garcia-Arenal et al. (1999), and Gibbs (1999) have shown that TMGMV has high genetic stability, limited recombination potential, and low mutability. Therefore, the proposed use of TMGMV as a bioherbicide is unlikely to pose an increased risk of emergence of new strains or intraspecific virus recombinants.

Additional safety features:

The mode of herbicidal action of TMGMV is unique. The host-specific lethal hypersensitive response causes the plant to self-destruct even as the virus titer remains relatively low in TSA tissues. TMGMV kills TSA plants rapidly and completely, minimizing the window of opportunity for environmental persistence, buildup, and horizontal movement. If TMGMV is used before fruit-set, as it will be recommended, the plant will be killed without the likelihood of a new crop of seeds being added to the soil. TMGMV is a mechanically transmitted virus that spreads by physical contact; it is neither vector-transmitted nor seed-borne. Given these factors, the probability of TMGMV spreading from treated TSA plants or from an occasional nontarget plant exposed to the virus in the field will be remote. TMGMV occurs widely in the United States; the isolate that is proposed to be registered as bioherbicide is indigenous and was recovered from an infected, clonally propagated hybrid *Columnea* plant in Florida. Thus, the use of TMGMV is not likely to have adverse effects on the environment. The biology of this virus, the specific host-pathogen interaction that is the basis for the herbicidal action, and the intended methods of application (spot spraying, with an herbicide wiper, or a wet-blade mower) provide excellent safeguards against any adverse environmental impacts. The probability of some cultivars of peppers and tobacco being at risk from the use of TMGMV is negligible and manageable because the bioherbicide will not be labeled for use near these plants grown as crops. Moreover, these plants would be at risk only if TMGMV is applied directly to these plants. Such an application would be inconsistent with the proposed label directions. Lastly, TMGMV can be inactivated, and work areas, tools, hands, etc. decontaminated, with hydrated phosphate detergents, 5% bleach, or sustained temperature above 85°C for 10 minutes. A commercial product, R-D-20 containing benzyl ammonium chloride is also an effective disinfectant viral plant pathogens (Southern Agricultural Insecticides, Inc., 2011; <http://www.southernag.com/docs/labels_msds/rd20.pdf>).

SolviNix LC, a highly effective bioherbicide for TSA

An aqueous liquid concentrate of TMGMV named SolviNix LC (Solvi from *Solanum viarum* and Nix meaning to put a stop to) has been extensively field-tested in Florida since 2000, including under an Experimental Use Permit granted by the EPA and approved by the State of Florida. To date, 70 field trials have been done at different locations in 19 counties throughout Florida. The trials were done during different times of the year under different climatic conditions and by using various types of application tools. In all of these trials, the virus has performed consistently, and there has been no evidence of resistance among the TSA populations or a change in the ability of the virus to kill TSA. Depending on the method of application, 85% to 100% control can be guaranteed (Charudattan et al. unpublished).

SolviNix LC is easy to apply and control TSA in the field. It can be applied with a backpack sprayer delivering the virus at 80 to 200 psi, certain types of commercially available herbicide wipers, and a wet-blade mover. Unlike conventional chemical

herbicides, it is unnecessary to attempt full coverage of TSA foliage; hitting a few leaves with short bursts of high-pressure spray, each lasting 1 to 3 seconds is all it takes to inoculate the plant. This feature, namely the ability to infect the plant and kill it with a few hits per plant allows the virus to be used in difficult-to-access sites such as wooded sites, cypress domes, etc.

The advantages of SolviNix LC as a control option for TSA include the following: It is an additional tool to manage TSA. An environment-friendly, natural, “green” herbicide, SolviNix is expected to be approved for use in organic beef and milk production. It does not harm forage legumes intercropped with grass and does not remove beneficial broadleaved plants that are essential to maintain biodiversity. It would be cheaper than but as effective as chemical herbicides. It is easy to apply and poses no risks to applicators. There would be no need to move cattle out of treated pastures nor are there re-entry requirements. EPA registration of SolviNix LC is pending at this time.

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VII. G. Management Techniques: Herbicide Strategies for Tropical Soda Apple

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Introduction

Tropical soda apple (TSA) is a perennial weed first identified in the late 1980s in south Florida. Since its introduction, it has spread rapidly throughout Florida as well as most of the states in the Southeastern U.S. Currently, TSA is commonly found in pastures, rights-of-way, citrus groves, and natural areas. During the early invasion process, it was quite common to find dense TSA stands covering hundreds of acres. More recently, however, dense stands of TSA are less common, but the total land area infested likely remains the same.

Many factors have likely led to the decrease in TSA densities throughout Florida, including natural enemies, introduced biocontrol agents, and the use of herbicides. Every control tactic that can be employed to control TSA will continue to decrease the amount of TSA observed in Florida, and ultimately the Southeastern USA. This article will examine much of the work that has been done with herbicides since TSA was first identified in south Florida.

Past Research

Some of the earliest research on TSA control employed mowing and various herbicide applications. Mislevey et al. (1999) reported that mowing TSA plants in June resulted in only 47 and 60% control by 90 and 120 days after treatment (Table 1), respectively, but control was less than 50% when the trial was repeated the following year. Mowing TSA infested pastures two or three times at 60 day intervals increased control to greater than 95%. Currently, mowing pastures costs approximately \$15 to \$20/A, making this an uneconomical choice. Applying triclopyr at 0.5 and 1.0 lb/A resulted in 95 to 100% TSA control, regardless of the number of mowing operations. Therefore, triclopyr became the herbicide standard for TSA control throughout Florida. The disadvantage of using triclopyr is that TSA seedling emergence is common within 120 days after application, resulting in multiple applications during the year. Although the cost of triclopyr has recently decreased due to loss of patent protection, TSA control with this herbicide can range between \$12 to \$30/A depending on the number of applications necessary. Later unpublished studies revealed that TSA was most susceptible to triclopyr when TSA plants were blooming. Since TSA seed germinate continually from October to May (Mislevey et al. 1997), it is common to observe seedling, blooming, and mature plants (with ripe fruit) all growing close in proximity. Therefore, to assure excellent control with triclopyr, it was recommended that TSA infested pastures be mowed (to synchronize blooming) followed by a triclopyr application at 1.0 lb/A 50 to 60

days later. This treatment strategy would cost approximately \$32 to \$50/A at today's prices.

Table 1. Influence of mowing prior to Remedy (triclopyr) application on mature tropical soda apple. Means within columns comparing control vs. chemical treatments followed by the same letters are not significantly different. Data adapted from Mislevy et al. 1999.

| # of mowing | Remedy rate | 90 DAT | 120 DAT |
|-------------|-------------|---------------------|---------|
| | pt/A | -----% control----- | |
| 1 | 0.0 | 47 b | 60 b |
| 1 | 1.0 | 98 a | 98 a |
| 1 | 2.0 | 96 a | 95 a |
| 2 | 0.0 | 95 b | 95 b |
| 2 | 1.0 | 100 a | 100 a |
| 2 | 2.0 | 100 a | 100 a |
| 3 | 0.0 | 98 a | 96 a |
| 3 | 1.0 | 100 a | 100 a |
| 3 | 2.0 | 100 a | 100 a |

In the early 2000s, a new active ingredient called aminopyralid (Milestone) was tested for its activity on TSA. Research by Ferrell et al. (2006) demonstrated that 4 to 7 oz/A provided good to excellent control, and was comparable to triclopyr (Remedy) at the higher application rate (Figure 1) for 335 days. After refining the rates, it was determined that Milestone reduced seedling emergence by nearly 100% compared to Remedy at 75 days after treatment (Figure 2). Initially, Milestone costs ranged from \$15 to \$18/A, which was cheaper than using a combination of mowing and Remedy application. Since this time, GrazonNext (Milestone + 2,4-D) has been registered, and is more economical for broadcast applications. Our research has shown that GrazonNext provides similar levels of activity on TSA compared to Milestone alone. Current prices for GrazonNext range from \$8 to \$12/A.

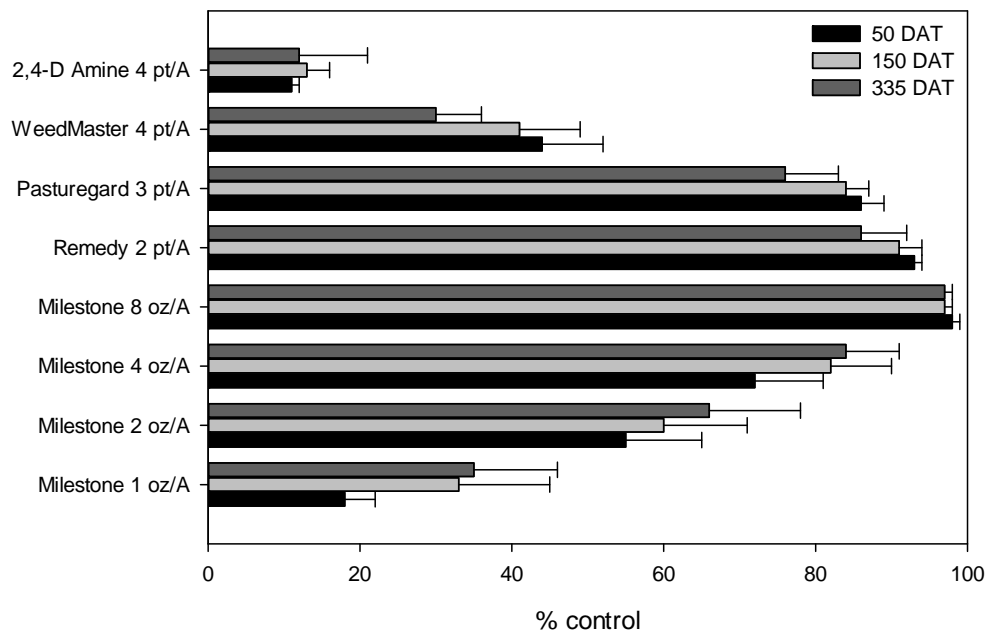


Figure 1. Control of tropical soda apple with various herbicides at 50, 150 and 335 days after treatment (DAT). Error bars represent the standard error of the mean. Data adapted from Ferrell et al. 2006.

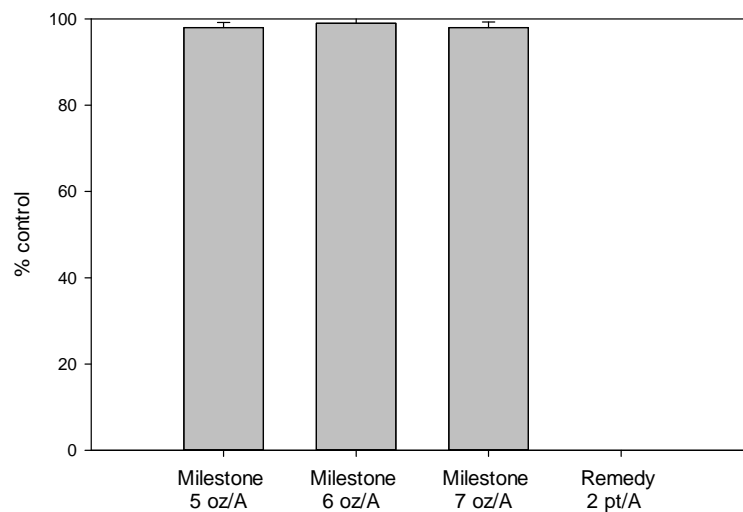


Figure 2. Influence of herbicide application on establishment of tropical soda apple from seed under field conditions. Error bars represent the standard error of the mean. Data adapted from Ferrell et al. 2006.

Current Herbicide Strategies

Dense infestations

Milestone and GrazonNext herbicides are the most effective for controlling dense stands of TSA. These herbicides possess postemergence control of existing plants and preemergence control of germinating seeds. Our research has shown that Milestone and Forefront will control germinating seedlings for over 6 months after application.

The application rate for Milestone is 5-7 oz/A while GrazonNext is 2-2.6 pt/A. Although the lower application rates are highly effective on existing plants, the higher rates will provide more soil activity and are suggested if large amounts of TSA seed are present in the soil. Although mowing prior to herbicide application is not required, it is important to add a non-ionic surfactant (0.25% v/v) and apply in at least 20 gallons of water per acre.

Another option is to use Remedy herbicide. When using Remedy, mow plants to a 3-inch stubble height as soon as possible to keep plants from producing fruit and seed. Repeat mowing when plants reach the flowering stage (50-60 days) through April. Fifty to 60 days after the April mowing, when plant regrowth is at the first flower stage (late May-June), spray Remedy at 1 qt/A + 0.25% v/v non-ionic surfactant in 40 gal/A of water. Remedy does not possess soil residual activity and follow up applications to control escaped or new seedlings will be necessary.

Regardless which herbicide is used, regular scouting after treatment is necessary. TSA can produce fruit at almost any time during the growing season and give rise to hundreds of additional plants. It is important to monitor the fields to ensure that no plants are allowed to reestablish and produce fruit.

Sparse infestations

Areas with low TSA infestation should be targeted and each plant sprayed individually. Recommended herbicides for 95 to 100% control are as follows:

- 1 Milestone at 0.5-0.8 oz per 2.5 gal (15 to 20 ml per 2.5 gal) + 0.25% v/v non-ionic surfactant + color marker. (Use a color marker with the herbicide solution to avoid spraying the same plant twice, or not spraying a plant at all).
- 2 GrazonNext or Remedy at 0.5% solution (50 ml per 2.5 gal) + 0.25% non-ionic surfactant + color marker.

When spot-spraying cover the entire plant with spray solution to ensure herbicide uptake and maximum control.

Allow herbicides to dry on plants 3-4 hours before rainfall. Monitor treated areas monthly and treat new TSA seedlings. Do not allow plants to produce fruit. Be sure to follow the guidelines for spraying volatile herbicides such as Remedy (see EDIS publication SS-AGR-12 Florida's Organo-Auxin Herbicide Rule [<http://edis.ifas.ufl.edu/WG051>] for more information).

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VII. H. Management Techniques: Can Nematodes Control Tropical Soda Apple?

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Introduction

Tropical soda apple (TSA), *Solanum viarum* Dunal, is one of three non-native species of the genus *Solanum* that are considered invasive weeds of agricultural and natural areas in Florida (Langeland et al. 2008). TSA is more widely recognized than either wetland nightshade (WNS), *S. tampicense* Dunal, or turkey berry (TBY), *S. torvum* Swartz, because it spreads rapidly throughout the southeastern United States after establishing in Florida (Westbrooks 1998). TSA and WNS were discovered in Florida in the early 1980s, and therefore are relatively new introductions (Wunderlin et al. 1993). However, TBY was introduced into Florida over a century ago but its invasive potential was not recognized until recently (Langeland et al. 2008).

All three *Solanum* spp. are included on the Federal and Florida Noxious Weed Lists (USDA/APHIS/PPQ 1999; FDACS 1999), and are listed as Category I or II invasive species by the Florida Exotic Pest Plant Council (FLEPPC 2009). Category I plants are “. . . invasive exotics that are altering native plant communities by displacing native species, changing community structures or ecological functions, or hybridizing with natives. . .” (FLEPPC 2009). Likewise, Category II plants are “. . . Invasive exotics that have increased in abundance or frequency but have not yet altered Florida plant communities to the extent shown by Category I species. [However], These species may become ranked Category I, if ecological damage is demonstrated. . .” (FLEPPC 2009). Although it is unclear why these non-native solanaceous plants have become invasive weeds, the lack of host-specific natural enemies in the southeastern United States where they have been introduced may have afforded these plants a competitive advantage over native species (Williams 1954).

Silverleaf nightshade (SLN), *S. elaeagnifolium* Cav., is a native congener that also occurs in Florida (Boyd et al. 1983, Wunderlin 1982). Furthermore, the native SLN is attacked by a complex of natural enemies that also may attack the three invasive *Solanum* spp. One of these is the foliar and stem-galling nematode *Ditylenchus phyllobius* (Thorne) Filipjev (Parker 1991). This nematode severely damages SLN by attacking the plant's leaves, petioles and axillary buds. Infected plants often are stunted and exhibit reduced flowering as well as premature leaf and shoot abscission. Seedlings and young vegetatively-produced shoots are often killed by the nematode.

From 1995-1997, we conducted a quarantine laboratory study to test the biological control potential of *D. phyllobius* against TSA and WNS (see Cuda et al. 1998). The rationale for this study was the ‘new association’ principle for biological control (Hokkanen & Pimental 1984). The two invasive *Solanum* were hypothesized to

be highly susceptible to infection by *D. phyllobius* because they evolved in different geographical regions without the nematode, and therefore lacked the defense mechanisms to resist attack. However, we discovered that both TSA and WNS exhibited an immune response following exposure to *D. phyllobius*, even at the highest treatment rate of ~320,000 infective juvenile nematodes per plant. Because we found that TSA and WNS were unsuitable host plants for *D. phyllobius*, we concluded that this foliar nematode had no value as a biological control agent for these two invasive *Solanum* species. However, TBV, which was unavailable when TSA and WNS were tested, should be tested. Additional surveys in the native ranges of all three *Solanum* spp. also may yield other foliar nematode species comparable to *D. phyllobius* that may have biological control potential.

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VII. I. Management Techniques: Can Native *Leptinotarsa* Beetles Control Tropical Soda Apple?

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Introduction

Tropical soda apple, *Solanum viarum* Dunal, wetland nightshade, *S. tampicense* Dunal, and turkey berry, *S. torvum* Swartz (Solanaceae), are currently recognized as three of Florida's most invasive nonnative plant species (FLDACS 1999, FLEPPC 2009, Langeland et al. 2008). Although it is unclear why these exotic solanaceous plants have become weeds, the lack of host-specific natural enemies in Florida (the introduced range) may have afforded these plants a competitive advantage over native species (Williams 1954). Tropical soda apple and wetland nightshade are native to South America (and possibly the West Indies), and Mexico, respectively (Wunderlin et al. 1993), whereas turkey berry is thought to have originated in West Africa (Ivens et al. 1978), Central or South America and the Caribbean region (Morton 1981, Waterhouse & Norris 1987), or Asia (Medal et al. 1999).

Silverleaf nightshade, *Solanum elaeagnifolium* Cav., a close relative of the three aforementioned invasive Solanums, is native to the southern United States, Mexico and possibly Argentina (Goeden 1971, Boyd et al. 1983), and belongs to the same subgenus *Leptostemonum* (D'Arcy 1972, Nee 1991). Silverleaf nightshade is attacked by many insect herbivores in the southwestern United States and Mexico (Goeden 1971). Two of the most damaging insect herbivores are the defoliating beetles *Leptinotarsa defecta* (Stål) and *L. texana* (Schaeffer) (Coleoptera: Chrysomelidae) (Jacques 1988). Both *L. defecta* and *L. texana* were released in South Africa for biological control of silverleaf nightshade (Olckers et al. 1999), and their biologies were summarized by Olckers et al. (1995).

Silverleaf nightshade is considered the natural host plant of *L. defecta* and *L. texana* (Goeden 1971, Neck 1983, Jacques 1988). This solanum defines the actual, realized or field host range of the beetles (Cullen 1990, Sheppard et al. 2005). Host range encompasses those plants on which an insect completes normal development in nature (Hanson 1983). However, the study by Olckers et al. (1995) demonstrated that under laboratory conditions these two beetles also developed and reproduced on other *Solanum* species that do not occur in the insects' native ranges. Hsiao (1981) also observed that *L. texana* developed and reproduced to some extent on eggplant, *Solanum melongena* L., as well as three native plant species- *S. dulcamara* L., *S. carolinense* L. and *S. rostratum* Dunal. These solanaceous plants are not typically exploited by the beetles in nature but are capable of supporting some development and reproduction, and comprise what is considered the insects' potential, physiological or fundamental host range (Cullen 1990, Sheppard et al. 2005). Horsenettle (*S. carolinense*) and presumably Florida horsenettle (*S. carolinense* L var. *floridanum* Chapm.) are the only potential host plants of *L. texana* that

are native to Florida (Wunderlin & Hansen 2008). In spite of its native status, Florida horsenettle is listed as a troublesome weed by Hall & Vandiver (1991).

Silverleaf nightshade *sensu stricto* is adventive in Florida, occurring sporadically from the Panhandle to the Keys (Wunderlin 1982, Wunderlin & Hansen 2008). Its natural enemies *L. defecta* and *L. texana* have not spread to Florida (Jacques 1988), presumably because the Gulf of Mexico is an effective barrier to insects like *L. texana* that are incapable of long range aerial dispersal (see Hoffmann et al. 1998). However, a computer model (CLIMEX) that uses various climatic factors to predict whether insects can colonize and persist in new geographic areas (Sutherst & Maywald 1985) indicated that *Leptinotarsa* beetles collected from silverleaf nightshade in the Brownsville area of south Texas could establish and persist in peninsular Florida if tropical soda apple, wetland nightshade or turkey berry were suitable host plants.

In a quarantine laboratory using single plant and paired plant tests, we determined the extent to which the invasive tropical soda apple, wetland nightshade or turkey berry were capable of supporting normal development and continuous reproduction of the North American silverleaf nightshade leaf beetles *L. defecta* and *L. texana* (see Cuda et al. 2002 for details). If these native insects were capable of establishing ‘new associations’ with the exotic *Solanums* (Hokkann & Pimental 1984), they could be introduced into Florida for biological control of these weeds after pre-introduction host specificity tests demonstrated they were safe to release.

Results and Conclusions

Detailed information on the procedures used and results obtained in this study are presented in Cuda et al. (2002). Neonate larvae of *L. defecta* developed to the pupal stage only on their natural host plant silverleaf nightshade. Feeding damage on turkey berry and wetland nightshade was negligible and no feeding occurred on tropical soda apple (Table 1). Likewise, *L. texana* fed minimally on wetland nightshade and not at all on tropical soda apple. However, development and reproduction of *L. texana* on the non-native turkey berry were comparable with silverleaf nightshade. Larvae of *L. texana* consumed twice the amount of leaf tissue on turkey berry compared to silverleaf nightshade, but did not exhibit a preference for their natural host plant when given a choice (Table 1). Although these results suggest that turkey berry may be included in the potential host range of the native silverleaf nightshade beetle *L. texana*, tropical soda apple was not accepted as a host plant by either *Leptinotarsa* spp. Therefore, these two native *Leptinotarsa* beetles have no value as biological control agents of tropical soda apple. However, *L. texana* could be recommended for release in Florida for biological control of turkey berry if host range testing showed it would not attack other native *Solanum* species.

Table 1. Feeding* (cm²) by larvae of *Leptinotarsa defecta* and *Leptinotarsa texana* on *Solanum* spp. in the laboratory.

| Test/Plant Species | <i>L. defecta</i> Mean (+SEM) | <i>L. texana</i> Mean (+SEM) |
|-----------------------|----------------------------------|---------------------------------|
| Single Plant | | |
| Silverleaf nightshade | 64.00 (9.2)a** | 52.30 (7.7)b |
| Turkey berry | 0.02 (0.01)c | 104.50 (26.4)a |
| Tropical soda apple | 0.00 (0.0)c | 0.00 (0.0)d |
| Wetland nightshade | 0.17 (0.04)b | 0.08 (0.4)c |
| Paired Plant | | |
| Silverleaf nightshade | —.— | 76.20 (6.69)b |
| Turkey berry | —.— | 40.00 (12.9)b |

*Amount of feeding per n=3 groups of 10 larvae.

**Means followed by the same letters within columns are not statistically different (p<0.05) according to Tukey's Studentized Range (HSD) test.

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VIII. Before and After Photos

Pictures taken by Julio Medal, Philip Stansly, Rodrigo Díaz (last two-sets of pictures in St. Lucie County, FL).

It is well known that many times photos describe situations better than words. Herein, we present pictures taken at different release sites through Florida.



May 14, 2003. Polk County
Before beetles release inside a cage



August 21, 2003. Polk County
After beetles released inside a cage



May 2003. Polk County. Before release



April 2008. Polk County. After release



August 5, 2004. Hendry County
Before beetles release



October 19, 2004
After beetles released



August 11, 2004. Okeechobee
Before beetles release



October 26, 2004. Okeechobee
After beetles released



August 26, 2004. St. Lucy County
Beetles released



September 22, 2004. St. Lucie County
After beetles released



August 15, 2005. Sumter County
Before beetles release



August 14, 2007. Sumter County
After beetles released



June 2, 2005. Okeechobee County
Before beetles release



July 7, 2005. Okeechobee County
After beetles released



June 2006. Sumter County
Before beetles release



July 2007. Sumter County
After beetles released



June 2006. Sumter County
Before beetles released



June 2007. Sumter County
After beetles released



July 2006. St. Lucie County
Before beetles released



October 2007. St. Lucie County
After beetles released



July 2006. St. Lucie County
Before beetles released



October 2007. St. Lucie County
After beetles released

Tropical soda apple plants treated with the bioherbicide SolviNix containing Tobacco mild green mosaic virus as the active ingredient. Pictures taken by R. Charudattan.



Before treatment with SolviNix



33 days after treatment



Before manual inoculation with SolviNix (left) and 27 days after treatment



Left: Cypress hammock where TSA plants were killed by selectively applied SolviNix. Middle: TSA treated on May 14, 2008. Right: Dead TSA plant 33 days after treatment.



Left: May 22, 2009, site before SolviNix-treatment; row of untreated border row TSA plants marked with red paint. Right: July 23, 2009 all except the untreated plants are dead.

Tropical soda apple plants treated with the herbicide Milestone 5 oz/A two weeks after treatment and two months after treatment. Pictures taken by Brent Sellers.



IX. Conclusions and Recommendations

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The sustainable management of tropical soda apple (TSA) below levels that cause economical/ecological damage will be based on the implementation of a combination of biological (insects, pathogens), chemical herbicides, mechanical control (mowing), cultural, and regulation of movement of livestock, contaminated hay, sod-grass, seeds (e.g. Bahia grass) to prevent the interstate movement of contaminated produce with the TSA fruits/seeds.

The TSA leaf beetle, *Gratiana boliviana* Spaeth (Chrysomelidae), introduced from Argentina-Paraguay, and released from 2003 to 2011, successfully established in central and south Florida, and post-release monitoring through Florida indicated it is causing significant defoliations, reducing the stand density and decreasing fruit production of TSA. This biological alternative provides the most sustainable and economically long-term suppression of TSA in locations where it established. However, as indicated earlier, the beetle has not been able to establish in north Florida, or southern parts of Georgia and Alabama. Great progress in suppressing TSA in Georgia and Alabama has been obtained based on prevention, mechanical, and chemical practices. To prevent interstate movement of TSA, the southern states have developed voluntary compliance agreements that outline regulations to insure products (hay, sod, seeds, soil) from areas infested with TSA are free of fruits/seeds. All these control efforts should be integrated with other control tactics that could be implemented in the northern regions (north of the frost free zone) including the utilization of more tolerant and better cold-winter adapted strains of the *G. boliviana*, and/or new potential biocontrol agents from cooler areas of South America. Additional host-specificity testing should be continued with an undescribed species of a nocturnal leaf-feeding beetle *Platyphora* sp., Preliminary host feeding-tests conducted on this beetle at the Universidade Federal do Paraná in Curitiba, and at the Universidade do Centro-Oeste in Iratí, Paraná state, Brazil, indicated that it has high specificity for feeding and larvipositing on TSA, and did not feed on economical solanaceous crops including potato, tomato, and eggplant (Medal et al. unpublished data). Another potential biocontrol candidate to control TSA in the USA, is a flea beetle, *Epitrix* sp. (Chrysomelidae), which has been found feeding on the TSA leaf and flower-buds west of Curitiba, Brazil. Preliminary host-specificity feeding/oviposition tests could be initiated with Brazilian collaborators.

An integrated approach including all the management tactics available should be utilized to successfully suppress TSA at a reasonable cost. No control technique should be excluded. Large TSA infestations could be initially treated with an effective herbicide and/or Solvinix, once it is approved and commercially available. Mechanical mowing before TSA fruit setting is essential to prevent the large fruit/seed production per plant

and to reduce new re-infestations. Once the TSA stand is reduced, other biological control methods could be implemented.

In the 1990s, coordinated efforts were initiated to manage TSA infestations with the participation of the University of Florida, the Florida Department of Agriculture and Consumer Services- Division of Plant Industry and Division of Forestry, USDA-APHIS, USDA-ARS, Florida Farm Bureau, Florida Cattlemen's Association, Florida A& M University, and collaborators in Brazil (Universidade Estadual Paulista – Jaboticabal campus; Universidade Federal do Paraná –Curitiba campus; Universidade de Santa Catarina – Blumenau campus; Universidade do Centro-Oeste, Iratí campus, Paraná state; Universidade Federal de Viçosa, Minas Gerais), Argentina (USDA-ARS in Hurlingham, Buenos Aires; INTA-Cerro Azul, Misiones province), Paraguay (Museo de Entomología, Asunción). This Task Force has implemented a management program in the southeastern USA, and a TSA Management Plan has now been developed to provide information on management options. Continued research to develop alternative control tactics, and to obtain more cold-tolerant biocontrol agents for those geographical regions where the *Gratiana boliviana* beetles have not established is needed. Post-release monitoring should continue, and studies on the compatibility of different control strategies (biological, chemical, mechanical, regulation) should be conducted along with studies on the economic benefits of the control tactics.