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STATUS OF HOT WATER IMMERSION QUARANTINE TREATMENT FOR TEPHRITIDAE IMMATURES IN MANGOS

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Abstract. Research performed by the Agricultural Research Service (ARS) led to approval by the Animal and Plant Health Inspection Service (APHIS) of hot water immersion quarantine treatments for mangos infested with fruit fly immatures. Thus, 'Francis' mangos from Haiti and mangos from Mexico that have been treated with hot water currently are imported into the United States. Similar treatments that disinfest fruit fly immatures in mangos are expected to be recommended by ARS and approved by APHIS for Puerto Rico and Peru. Hot water immersion quarantine treatment research is also underway in Brazil, Jamaica, Venezuela, and Guatemala.

Mangos, *Mangifera indica* L., imported into the United States from countries having fruit fly pests are subjected to federal quarantine regulations. Hot water immersion was approved by the Animal and Plant Health Inspection Service (APHIS) to disinfest Haitian 'Francis' mangos having immatures of the West Indian fruit fly, *Anastrepha obliqua* (Macquart) and Caribbean fruit fly, *A. suspensa* (Loew) (1) and for Mexican mangos infested with Mexican fruit fly, *A. ludens* (Loew) and *A. obliqua* (2). Hot water quarantine treatments were recommended by the Agricultural Research Service (ARS) for Florida mangos infested with *A. suspensa* and mangos from the state of Chiapas, Mexico infested with the Mediterranean fruit fly, *Ceratitidis capitata* (Wiedemann) and the so-called dark fruit fly, *A. serpentina* (Wiedemann) (16, 18). Approved hot water treatments have stimulated work in Caribbean and Central and South American countries which grow mangos and have fruit

flies, as growers desire to export mangos to the United States.

Herein is presented summary data obtained from research performed since 1984 that provides the status of hot water immersion as a quarantine treatment against Tephritidae immatures in mangos.

Materials and Methods

All tests were done with 'Francis,' 'Oro,' and 'Ataulfo' mangos (individual weights of 350-590 g, 0.8-1.3 lb.), and 'Tommy Atkins,' 'Keitt,' and 'Haden' mangos (individual weights of 450-700 g, 1-1.54 lb.). Procedures required to perform all tests were reported by Sharp et al. (15, 16, 17, 18). Briefly, procedures are discussed as follows. Wild strains of fruit flies were collected as larvae from host fruits, and laboratory strains were reared from artificial diets (4, 7, 8, 10, 12). Female flies oviposited in mangos in cages. Infested mangos were cleaned, randomized to insure similar infestation levels per mango, and held for several days so larvae could develop to late second and third instars, the stages at which they move the greatest distance into the pulp. Infested mangos were divided into groups of equal numbers, put into mesh sacks, and immersed in circulating water at $46.1 \pm 0.5^\circ\text{C}$ in metal containers (13) for 10-80 min to obtain laboratory data to estimate Probit 9 security (99.9968% mortality) (3). Equal numbers of infested mangos not immersed were the controls used to estimate the number of treated larvae. Treated and nontreated mangos were held in separate racks over sand (5). The sand below each rack was sifted 2-3 times each week for several weeks to recover all larvae and pupae. Also, mangos held over strainers were sprayed with water to recover all immatures that remained in the fruits (17). The number of normally formed pupae was used in the data analysis. Data were analyzed to the 99% mortality level with the probit and GLM procedures of the Statistical Analysis Systems (SAS) (9). The fiducial limits for immersion times corresponding to 99.9968% mortality were obtained by using the formula in Finney (6). Pupae were held for eclosion, and the number of flies that emerged was recorded.

Results of research only are reported and mention of a trade name does not constitute a recommendation by the U.S. Dept. of Agriculture.

Table 1. Estimated probit analysis values for fruit fly larvae in hot-water treated mangos from Haiti, Mexico, Florida, and Texas.

Fruit fly identification	Strain, origin	Slope $\pm$ SEM	Intercept $\pm$ SEM	Estimated 99.9968% mortality in minutes (an lower and upper fiducial limits at the 95% confidence level)		
<i>A. obliqua</i> ^z	lab, Haiti	0.890 $\pm$ 0.0124	3.8411 $\pm$ 0.3161	58.0	(47.4	84.9)
<i>A. obliqua</i> ^y	lab, Haiti	0.0803 $\pm$ 0.0063	3.6391 $\pm$ 0.1784	66.8	(59.5	78.1)
<i>A. obliqua</i> ^y	wild, Mexico	0.0621 $\pm$ 0.0048	3.8116 $\pm$ 0.1584	83.6	(74.0	98.3)
<i>A. suspensa</i> ^z	lab, Florida	0.1450 $\pm$ 0.0146	2.5837 $\pm$ 0.3233	44.3	(39.1	53.3)
<i>A. suspensa</i> ^y	lab, Florida	0.0840 $\pm$ 0.0015	3.9326 $\pm$ 0.0403	60.3	(59.1	61.7)
<i>A. ludens</i> ^y	wild, Mexico	0.0618 $\pm$ 0.0020	4.5860 $\pm$ 0.0564	71.4	(68.7	74.5)
<i>A. ludens</i> ^y	lab, Texas	0.0639 $\pm$ 0.0061	4.838 $\pm$ 0.1367	65.1	(57.2	77.3)
<i>A. ludens</i> ^z	lab, Texas	0.0819 $\pm$ 0.0140	4.4132 $\pm$ 0.2660	56.0	(43.3	91.0)
<i>A. serpentina</i> ^y	wild, Mexico	0.0810 $\pm$ 0.0059	3.7813 $\pm$ 0.1654	64.5	(58.4	73.3)
<i>C. capitata</i> ^y	lab, Mexico	0.0538 $\pm$ 0.0049	5.3693 $\pm$ 0.1495	67.5	(60.4	78.5)

^zTests performed with 'Francis' mangos, average weight 525 grams.

^yTests performed with 'Tommy Atkins,' 'Keitt,' 'Haden,' and 'Kent,' average weight 625 grams.

Results and Discussion

Probit analyses of data obtained from Haiti, Florida, and Mexico are presented in Table 1. Probit 9 values ranged from 44 to 84 min and indicated that a wild strain of *A. obliqua* from Mexico was the least susceptible to hot water immersion compared with values for other species. Strain and species differences appeared to be major factors affecting larval mortality. The stage of mango ripeness, percent moisture, and pulp thickness measured from the surface to the seed apparently were the more critical factors that affected both the transfer of heat and larval mortality than the different cultivars used in the tests (Sharp, unpublished data).

Summary data for confirmatory tests are presented in Table 2. Mangos were immersed for 75 min (for 'Francis') and 90 min (for other cultivars) in water at 46.1°C $\pm$ 0.5°C for confirmatory tests to provide quarantine security. No flies emerged from pupae treated as larvae, and all tests indeed resulted in quarantine security. Also, no approved treatment produced damage to the mangos (11, 14, 15, 16, 17, 18).

Data provided by researchers in Chiapas, Mexico and Puerto Rico are being reviewed by ARS, and work is near completion in Peru. Research is underway in Brazil, Guatemala, Venezuela, and Jamaica to develop hot water treatments for mangos infested with fruit flies primarily of the genera *Anastrepha* and *Ceratitidis*. Several large scale commercial hot water facilities have been built in Mexico and Haiti and will be built in other countries. Many hot water

quarantine treatments are expected to be approved by APHIS after all data gathered from researchers in other countries are analyzed and reviewed by ARS.

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Table 2. Summary data of confirmatory tests for fruit fly larvae.

Fruit fly identification	Strain, origin	Estimated No. treated larvae
<i>A. obliqua</i> ^z	lab, Haiti	147,993
<i>A. obliqua</i>	lab, Haiti/Texas ^y	101,049
<i>A. obliqua</i>	wild, Mexico	116,869
<i>A. suspensa</i> ^z	lab, Florida	102,509
<i>A. suspensa</i>	lab, Florida	116,031
<i>A. ludens</i>	wild, Mexico	226,054
<i>A. ludens</i>	lab, Texas	187,114
<i>A. serpentina</i>	wild, Mexico	111,031
<i>C. capitata</i>	lab, Mexico	138,443

^zTests performed with 'Francis,' all others done with 'Oro,' 'Ataulfo,' 'Tommy Atkins,' 'Keitt,' and 'Haden.'

^yStrain originated in laboratory colony in Haiti, but was transported to and colonized in the laboratory in Texas.

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EVALUATION OF PRESTORAGE TREATMENT EFFECTS ON VASE LIFE OF LEATHERLEAF FERN

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Index words. *Rumohra adiantiformis*, frond curl syndrome, frond wilt, desiccation, 8-hydroxyquinoline citrate, antitranspirants.

Abstract. Various postharvest treatments, applied prior to storage at 4°C, were evaluated for extending the vase life of leatherleaf fern fronds. Prestorage immersion (dipping) of fronds in calcium chloride (1, 10, 100 mmolar) or magnesium oxide (0.01 and 0.1 mmolar) solutions had no effect on vase life while triadimefon (150 and 300 ppm) dips reduced vase life. Delaying postharvest immersion treatments by 3.5 hr reduced vase life by 26% (7.3 versus 5.4 days). Prestorage dipping treatments using antitranspirants (Envoy, Folicote, Pro-Tec, VaporGard or WiltPruf) did not affect vase life compared to immersion in deionized water when frond curl syndrome did not occur (mean vase life of 21.9 days). Stipe and whole frond treatment with 8-hydroxyquinoline citrate (1,000 ppm) prior to storage was not effective in extending vase life of leatherleaf fern fronds when frond curl syndrome occurred.

Reductions in vase life of leatherleaf fern [*Rumohra adiantiformis* (Forst.) Ching], the dominant cut foliage crop produced in Florida, can be a serious problem (5, 9). Rapid poststorage desiccation (frond curl syndrome, wilt) and premature yellowing are the conditions that reduce frond vase life, with the former condition causing the greater reduction in longevity (7, 10, 13). Prestorage treatments that could help maintain poststorage water balance and thereby increase the vase life of this crop would be very helpful in maintaining the market for this cut foliage crop.

In the field, numerous chemicals have been used to induce stomatal closure in attempts to reduce water stress to intact plants. Examples include calcium chloride, magnesium oxide and triadimefon (1). However, preharvest field spraying of leatherleaf fern beds may not be practical as previous research has shown it to be ineffective (8), and because only mature fronds are harvestable it would be wasteful to treat all fronds prior to harvest.

Commercially, leatherleaf fern fronds are harvested and left in the field for several hours before they are taken to packing sheds where they are dipped in water prior to packaging and storage. The practice of immersing fronds lends itself to prestorage treatment of fronds. This process has been tried with antitranspirants and appears to

have potential for increasing vase life (4, 8). However, previous experiments with antitranspirant dips did not include storage, a necessary component in commercial practice.

Pre- and poststorage treatments of leatherleaf fern fronds with floral preservatives have generally been of no benefit in prolonging vase life; however, 10-15 minute pulse treatments with 800-1,000 ppm solutions of 8-hydroxyquinoline citrate (8-HQC) have increased frond vase life from 19-37% at some harvests (12, 13). Marousky reported that 8-HQC holding solutions closed stomata of cut gladiolus leaves (3).

These experiments were designed to evaluate the effects of prestorage treatments on the vase life of leatherleaf fern fronds.

Materials and Methods

The leatherleaf fern used in these experiments was grown in ground beds of Millhopper fine sand at the Central Florida Research and Education Center—Apopka under 73% shade obtained using polypropylene shade fabric. Fertilization consisted of bimonthly applications of 17-2.6-10 controlled release fertilizer containing minor elements (Sierra Chemical, Milpitas, CA) applied at a 560 kg/ha/yr rate. Temperatures were maintained above 4°C and irrigation was by overhead sprinklers. Mature dark green fronds were harvested using clippers and stipes were recut to 17 cm below the lowest pinna prior to storage. The immersion/dipping treatments were applied for 15 minutes after which fronds were sealed according to treatment in nonvented polyethylene bags containing 10 ml of water. Bags were placed in waxed, corrugated fiberboard boxes and boxes were stored at 4°C.

After storage, stipes were recut 15 cm below the lowest pinna using razor blades and placed in deionized water. Vase life and water uptake of individual fronds were determined daily as described previously (13). Fronds were held at $23 \pm 1.5^\circ\text{C}$, $65 \pm 5\%$ RH, and $12 \mu\text{mol}\cdot\text{s}^{-1}\cdot\text{m}^{-2}$ 12 hr/day for vase life evaluations. Poststorage diffusive conductance of the abaxial surface of fronds was measured using a steady state porometer (LI-COR LI-1600).

Expt. 1. (Dipping solutions experiment). Fronds were harvested 29 Sept. and immersed in aqueous solutions of 1) deionized water, 2) 1 mmolar CaCl_2 , 3) 10 mmolar CaCl_2 , 4) 100 mmolar CaCl_2 , 5) 0.01 mmolar MgO , 6) 0.1 mmolar MgO , 7) 150 ppm triadimefon, or 8) 300 ppm triadimefon solutions either immediately after harvest or after being held in the field for 3.5 hrs (delayed treatment/commercial practice). Fronds were stored for 5 days. Individual fronds were the experimental unit. Vase life measurements were replicated 20 times, and water uptake and