Sensitivity of the Quarantine Pest Rough Sweetpotato Weevil, *Blosyrus asellus* to Postharvest Irradiation Treatment

P.A. Follett[1], G. T. McQuate, C. D. Sylva, and **A. Swedman**
USDA-ARS, Daniel K. Inouye U.S. Pacific Basin Agricultural Research Center, Hilo, Hawaii 96720. [1]Correspondence: peter.follett@ars.usda.gov

Abstract. Rough sweetpotato weevil, *Blosyrus asellus* (Olivier), is a new quarantine pest of Hawaii sweetpotatoes. Currently, sweetpotatoes can be exported from Hawaii to the U.S. mainland using a postharvest irradiation treatment of 150 Gy to control three other regulated insect pests. Studies were conducted to determine whether this current radiation dose will also control any rough sweetpotato weevils in export shipments. Adult weevils were treated at various levels between 25 to 125 Gy and egg laying and egg hatch were measured. Rough sweetpotato weevil was found to be highly susceptible to irradiation, with no egg hatch at any radiation dose, even 25 Gy, the lowest dose tested. Results suggest that the 150 Gy irradiation treatment should be sufficient for control of rough sweetpotato weevil in Hawaii sweetpotatoes.

Key words: Irradiation, quarantine pest, Curculionidae, sweetpotato, phytosanitary treatment

Sweetpotato, *Ipomoea batatas* (L.) Lamarck (Convulvulaceae), is grown for its starchy roots and immature leaves which are used for human consumption and animal feed. It is grown in tropical and subtropical climates, as well as in mild temperate regions, between the latitudes 42°N to 32°S (Mukhopadhyay et al. 2011). Sweetpotato is one of the top ten staple crops produced worldwide, trailing only corn, rice, wheat, potatoes, cassava, and barley (FAO 2015). The many insect species that attack sweetpotato can be divided into three groups based on whether the damage is caused to leaves, stems or roots. Root damage can be either internal or external. Internal damage occurs when the larvae of some pest species tunnel in the storage roots, depositing frass in the process. That, in combination with production of terpenes by the roots in response to the damage, can render the roots inedible. External damage reduces root quality and marketable yield (Ames et al. 1997).

Postharvest irradiation at 150 Gy is an USDA-APHIS-approved quarantine treatment for sweetpotatoes exported from Hawaii to the U.S. mainland to control three regulated pests: West Indian sweetpotato weevil, *Euscepes postfasciatus* (Fairmaire) [Coleoptera: Curculionidae], sweetpotato vine borer, *Omphisa anastomosalis* (Guenee) [Lepidoptera: Pyralidae], and sweetpotato weevil, *Cylas formicarius elegantulus* (Summers) [Coleoptera: Curculionidae] (Follett 2006). A new sweetpotato pest, the rough sweetpotato weevil, *Blosyrus asellus* (Olivier) (Coleoptera: Curculionidae), was first detected on the island of Oahu in 2008 and has subsequently been detected on the islands of Kauai (2011) and Hawaii (2014) (Heu et al. 2014, McQuate et al. 2016). This weevil is not present on the U.S. mainland (McQuate et al. 2016) and therefore is considered an actionable pest. Adult *B. asellus* feed on the leaves of sweetpotatoes, but larvae feed on the roots, creating grooves or channels along the surface (Heu et al. 2014). Adults or larvae may occur with exported sweetpotatoes. We conducted research to determine whether the currently used postharvest irradiation treatment of 150 Gy will also control any rough sweetpotato weevils in export shipments.

Materials and Methods

Insects. All adult rough sweetpotato weevils used in experiments were collected from an abandoned sweetpotato farm located in Papaikou on the island of Hawaii (Universal Transverse Mercator [UTM] grid [USGS 2001]: Easting 279713, Northing 2191391 m, [Zone 05 Q]; 145 m elevation). On the mornings of 25 January and 26 February 2016, rough sweetpotato weevil adults were hand collected from the decaying leaf litter underneath old sweetpotato vines and brought back to the laboratory for further processing. Weevils were separated by gender using external characteristics, and male and female weevils were placed in separate 1.0 liter screen-topped HI-PLAS buckets [Highland Plastics, Inc., Mira Loma, CA]. Each bucket contained several fresh undamaged sweetpotato leaves as a food source. The leaves were inserted in a 207-ml cup (HI-PLAS bucket; Highland Plastics, Inc., Mira Loma, CA) holding water.

No artificial diet exists for *B. asellus* and attempts to rear the weevil on sweetpotato roots were unsuccessful. Hence, all experiments used field collected adult weevils and weevils were held after treatment on fresh undamaged sweetpotato leaves. Leaves were brought back to the laboratory, rinsed with water and held in 3.8 liter size buckets with 1.0 liter of water until leaves were used in the experiments. A CI-203 portable laser area meter [CID Bio-Science Inc., Vancouver, WA] was used, with a CI-203A conveyor attachment, to estimate the area of leaves used. Dimensions of leaves used ranged from 8.0 to 11.0 cm long and 6.7 to 9.5 cm wide, with overall leaf area ranging from 24.7 to 51.0 cm^2.

Experiment 1. On 27 January 2016, a total of six hundred *Blosyrus asellus* adults were transferred into 60 individual 29.6 ml polystyrene portion cups (Solo Cup Company, Highland Park, IL) with screened tops such that each cup contained ten males or ten females. Cups were divided into five irradiation treatments: 0, 50, 75, 100, and 125 Gy, with six cups per treatment. Cups with screened tops containing weevils were transported to a nearby commercial x-ray irradiation facility (CW Hawaii Pride, Keaau, Hawaii, USA), and treated with x-rays using an electron linear accelerator (5 MeV, model TB-5/15, Titan Corp., San Diego, California, USA). To minimize the dose uniformity ratio (the ratio of the maximum/minimum dose), wooden racks holding a single row of the cups holding weevils were placed perpendicular to the x-ray beam and elevated by placement on two 1.9 liter plastic containers (polypropylene El-610-64, Highland Plastics, Inc., Mira Loma, CA) and positioned in the center of the carrier. ROW dosimeters (Opti-chromic detectors, FWT-70-83M, Far West Technology, Goleta, California, USA) were placed in empty 1 oz cups in three locations (at both ends and in the middle) on each wooden rack to estimate dose delivery and variation during treatment. Dosimeters were read with an FWT-200 reader (Far West Technology, Goleta, California, USA) at 600-nm absorbance.

At this commercial facility, product moves in front of the x-ray beam on a conveyor belt, so individual vials of weevils pass in front of the beam sequentially and each vial can be considered a replicate. Measured doses for each radiation treatment were 39.4–52.9 Gy (50 Gy), 66.4–85.5 Gy (75 Gy), 88.9–103.5 Gy (100 Gy), and 112.6–126.1Gy (125 Gy).

After irradiation, all adult weevils were brought back to the laboratory at the USDA-ARS Daniel K. Inouye U.S. Pacific Basin Agricultural Research Center in Hilo, HI and individual males and females from the same dosage were paired and transferred to buckets (7.0 cm [diam.] x 7.5 cm screened-top HI-PLAS cups [Highland Plastics, Inc., Mira Loma, CA]). Each bucket contained one sweetpotato leaf inserted in a 4 mm diam. hole on the lid of a 96 ml portion cup [Fabri - Kal, Kalamazoo, MI]. Buckets with weevil pairs and leaves were held in an insect holding room at 21.4 ± 0.03°C, 63.1 ± 0.09% relative humidity (RH) and a 12:12 (L:D) h photoperiod. Leaves were inspected for eggs and changed every 24 h for the first 48 h, then inspected for eggs and changed every 72 h for an additional three weeks (at which time all egg laying had stopped in irradiated weevils). Eggs were typically laid on the tips of the leaves, with a leaf edge folded over the egg cluster to form a protective covering. All sweetpotato leaves having eggs present in the fold covering were opened and eggs counted. Leaves with eggs attached were transferred to 100 mm x 15 mm Petri dishes [Fisher Scientific, Pittsburgh, PA] containing 7.0 cm diam. black filter paper [Ahlstrom Filtration, Mt. Holly Springs, PA] moistened with distilled water and held for hatch at 22.75 ± 2.25 ˚C, 56.5 ± 10% RH and a 8:16 (L:D) h photoperiod. The filter paper was re-moistened twice per week to prevent egg dehydration. Eggs in Petri dishes were inspected for hatch daily from day 9 after treatment until day 23. Untreated control eggs hatched at 10-15 days. Dose response data on eggs laid and egg hatch were subjected to regression analysis.

Experiment 2. A second experiment was conducted at lower radiation doses in an attempt to identify the threshold for sterility. On 3 March 2016, 480 weevils were transferred into 48 individual 29.6 ml portion cups with screened tops each containing ten males or ten females. Cups were divided into four irradiation dosages: 0, 25, 50, and 75 Gy, six cups per treatment. Cups were irradiated and held for egg laying and hatch as described above. In the first experiment mold formed on the leaves carrying eggs when transferred to the moist filter paper, which may have negatively affected hatch. In the second experiment eggs were removed from the leaves and transferred directly to the filter paper to improve hatch. Dose response data on eggs laid and egg hatch were subjected to regression analysis.

Results and Discussion

The most advanced life stage that is associated with the commodity is invariably the most tolerant of irradiation when the goal of the treatment is to stop successful reproduction (e.g. Follett 2006). Adult rough sweetpotato weevils may be found with export shipments of sweetpotatoes hence we irradiated only the adult stage of rough sweetpotato weevil and examined the effects of the treatment on egg laying and egg hatch.

In the first experiment, 32–52% of the irradiated weevils laid eggs across all treatment levels (50-125 Gy). The percentage of weevils laying eggs did not appear to be correlated with irradiation dose as 28.3% (17 out of 60) of the untreated control weevils laid eggs and 51.7% (31 out of 60) of the weevils in the 100 Gy treatment laid eggs (Table 1). Regression analysis indicated that the number of eggs laid was unrelated to dose as the slope of the line describing the relationship (log [number of eggs laid] = 3.3 + 0.0015 [log dose], R^2 = 0.003, P = 0.56) was not significantly different from zero. None of the eggs laid by adults in any of the irradiation treatments hatched, whereas egg hatch in the untreated control treatment was 48.5 % (Table 1).

In the second experiment, 25–32% of the irradiated weevils laid eggs across all treatment levels (25-75 Gy). The percentage of weevils laying eggs was not negatively affected by irradiation dose as 21.7% (13 out of 60) of the untreated control weevils laid eggs and 31.7% (19 out of 60) weevils in the 50 and 75 Gy treatments laid eggs (Table 1). Regression analysis indicated that the number of eggs laid was unrelated to dose as the slope of the line describing the relationship (log [number of eggs laid] = 3.0 – 0.00128 [log dose], R^2 = 0.005, P = 0.57) was not significantly different from zero. None of the eggs laid by adults in the irradiation treatments hatched, whereas egg hatch in the untreated control treatment was 73.6% (Table 1).

Rough sweetpotato weevil was highly susceptible to irradiation, with no egg hatch even at the lowest dose tested, 25 Gy. The other weevils found in Hawaii sweetpotatoes, sweetpotato weevil and West Indian sweetpotato weevil, are considerably more radiation tolerant and require doses of 140–145 Gy for sterilization (Follett 2006), but several other species of curculionids were reported to show levels of susceptibility similar to rough sweetpotato weevil. For example, diapausing northern strain plum curculio, *Conotrachelus nenuphar* (Herbst) (Hallman 2004) and coffee berry borer, *Hypothenemus hampei* (Ferrari) (Follett unpublished) were sterilized (no egg hatch) at 40 Gy, and the diaprepes root weevil, *Diaprepes abbreviatus (L.)*, was sterilized at 50 Gy (Gould and Hallman 2004). To provide a margin of safety, a radiation dose of 50 Gy could be proposed to prevent reproduction in rough sweetpotato weevil and thereby provide quarantine security. This proposed dose is based on information from 17 adult females irradiated at 25 Gy and 38 adult females irradiated at 50 Gy which all together laid a total of 1,353 eggs that did not hatch. These numbers are far below the numbers normally required to confirm the efficacy of a quarantine treatment (30,000 treated individuals with no survivors or reproduction) (Follett and Neven 2006). Hawaii sweetpotatoes are normally irradiated at 150 Gy to control the three primary regulated pests (sweetpotato weevil, West Indian sweetpotato weevil, and sweetpotato vine borer) before export (Follett 2006). Our results show that rough sweetpotato weevil is significantly less radiation tolerant than the other regulated sweetpotato pests and therefore the 150 Gy treatment should safely control any rough sweetpotato weevils present in export shipments. USDA-APHIS has approved generic radiation treatments of 150 Gy for tephritid fruit flies and 400 Gy for all other insects except lepidopteran pupae and adults (Follett 2009). Currently, if rough sweetpotato weevil is found during pre-shipment inspection of Hawaii sweetpotatoes, all cartons from the specific grower (i.e., in the shipment) must be treated at 400 Gy instead of the usual 150 Gy. For large shipments, this more than doubles the treatment time and thereby increases the cost of treatment significantly. Our results show that the currently approved 150 Gy treatment should be sufficient for control of rough sweetpotato weevil and therefore increasing the dose to 400 Gy should not be required.

Acknowledgments

We thank sweetpotato farmers in the Papaikou area of the island of Hawaii for permission to collect insects and gather leaves. We thank Russell Ijima, Austin Bloch, and Eva Brill (USDA-ARS-DKIPBARC, Hilo, HI) for their field assistance. Roxana Myers and Scott Geib (USDA ARS, Hilo, HI) provided helpful reviews of an early draft of the manuscript. This article reports the results of research only. Mention of a proprietary product does not constitute an endorsement or a recommendation by the USDA for its use. USDA is an equal opportunity provider and employer.

Table 1. Number of eggs laid and eggs hatched after irradiation of rough sweetpotato weevil adult females.

Exp.	Target dose (Gy)	Total no. females	No. gravid females	No. eggs laid (mean ± SE)	No. eggs hatched (mean ± SE)	% eggs hatched (mean ± SE)
1	0	60	17	68.8 (23.2)	35.5 (11.9)	51.6 (7.0)
	50	60	19	28.5 (4.8)	0	--
	75	60	20	43.3 (4.9)	0	--
	100	60	31	55.2 (5.6)	0	--
	125	60	20	33.3 (6.7)	0	--
2	0	60	13	60.5 (22.9)	44.5 (16.9)	73.7 (7.8)
	25	60	15	18.1 (3.5)	0	--
	50	60	19	29.8 (6.6)	0	--
	75	60	19	22.9 (4.2)	0	--

Literature Cited

Ames, T., N.E.J.M. Smit, A.R. Braun, J.N. O'Sullivan, and **L.G. Skoglund.** 1997. Sweetpotato: Major pests, diseases, and nutritional disorders. International Potato Center, Lima, Peru.

Food and Agriculture Organization [FAO]. 2015. FAOSTAT (2013 production data). Available online at: http://faostat3.fao.org/browse/rankings/commodities_by_regions/E (accessed 5 May, 2016).

Follett, P.A. 2006. Irradiation as a methyl bromide alternative for postharvest control of *Omphisa anastomosalis* (Lepidoptera: Pyralidae) and *Euscepes postfasciatus* and *Cylas formicarius elegantulus* (Coleoptera: Curculionidae) in sweetpotatoes. Journal of Economic Entomology 99: 32–37.

Follett, P.A. 2009. Generic radiation quarantine treatments: the next steps. Journal of Economic Entomology 102 (4):1399–1406.

Follett, P.A., and **L.G. Neven.** 2006. Current trends in quarantine entomology. Annual Review of Entomology 51: 359–385.

Gould, W.P., and **G.J. Hallman.** 2004. Irradiation disinfestation of diaprepes root weevil (Coleoptera: Curculionidae) and papaya fruit fly (Diptera: Tephritidae). Florida Entomologist 87 (3): 391–392.

Hallman, G. J. 2004. Ionizing irradiation quarantine treatment against plum curculio (Coleoptera: Curculionidae), Journal of Economic Entomology 96 (5): 1399–1404.

Heu, R.A., D.M. Tsuda, S.K. Fukuda, C.L. Young, and **M.I. Lee.**

2014. A rough sweetpotato weevil, *Blosyrus asellus* (Olivier) (Coleoptera: Curculionidae). New Pest Advisory no. 09-01, updated December 2014. Hawaii Department of Agriculture, Honolulu, HI. Available online at: http://hdoa.hawaii.gov/pi/files/2013/01/NPA-Blosyrus-MASTER.pdf (accessed 5 May, 2016).

McQuate, G.T., C.D. Sylva and **B.R. Kumashiro.** 2016. First field collection of the rough sweetpotato weevil, *Blosyrus asellus* (Olivier) (Coleoptera: Curculionidae), on Hawaii island, with notes on detection methods. Proc. Hawaiian Entomol. Soc. 48: 1–8.

Mukhopadhyay, S.K., A. Chattopadhyay, I. Chakraborty, and **I. Bhattacharya.** 2011. Crops that feed the world 5. Sweetpotato. Sweetpotatoes for income and food security. Food Security 3: 283–305.

United States Geologic Survey [USGS]. 2001. The Universal Transverse Mercator (UTM) Grid. Fact Sheet 077-01 (August 2001). Available at: http://pubs.usgs.gov/fs/2001/0077/report.pdf (accessed 9 Aug., 2016).