Irradiation as a Phytosanitary Treatment for White Peach Scale (Homoptera: Diaspididae)

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ABSTRACT Irradiation was examined as a potential phytosanitary treatment to control white peach scale, *Pseudaulacaspis pentagona* (Targioni-Tozzetti) (Homoptera: Diaspididae), a serious quarantine pest of papaya, *Carica papaya* L., in Hawaii. Dose–response tests were conducted with second-stage nymphs, adult females without eggs, and adult females with eggs at a series of irradiation doses between 60 and 150 Gy to determine the most tolerant stage. The adult female with eggs was the most tolerant stage. In large-scale validation tests 35,424 adult female scales with and without eggs irradiated at a dose of 150 Gy produced no F_1 generation adults with eggs. Irradiation treatment with a minimum absorbed dose of 150 Gy should provide quarantine security for white peach scale on exported papaya and other commodities.

KEY WORDS phytosanitary treatment, quarantine pest, x-ray, *Pseudaulacaspis pentagona*

White peach scale, *Pseudaulacaspis pentagona* (Targioni-Tozzetti) (Homoptera: Diaspididae) is a polyphagous, cosmopolitan pest. White peach scale was first discovered in Hawaii in 1997 on papaya, *Carica papaya* L., and has become a widespread and serious economic pest on this crop (Follett and Gabbard 1999). Papayas are exported from Hawaii to the U.S. mainland after receiving a postharvest treatment to control quarantine pests. The main pests of quarantine concern are tephritid fruit flies (USDA–APHIS–PPQ 1996). In addition to fruit flies, white peach scale is a quarantined pest for California (Follett and Gabbard 1999), and a high prevalence of white peach scale on the surface of fruit can cause delays at ports-of-entry or rejection.

Irradiation is one of the quarantine treatment technologies used to control pests on papayas exported from Hawaii to the U.S. mainland (Follett 2004). USDA–APHIS recently published a final rule approving a generic irradiation quarantine treatment of 150 Gy to control tephritid fruit flies and 400 Gy for control of all insect pests except lepidopteran pupae and adults (which may require higher doses) (Federal Register 2006). Hawaii papayas are routinely irradiated at a minimum dose of 400 Gy to control white peach scale in addition to fruit flies to prevent interruption of shipments. Lowering the required irradiation dose for white peach scale would lower treatment costs and increase capacity of the irradiation facility. Until now, there was no information on the radiotolerance of white peach scale (IDIDAS 2005). Another diaspidid scale, coconut scale, *Aspidiotus destructor* Signoret, is controlled with an irradiation dose of 150 Gy (Follett 2006).

Irradiation studies were conducted to determine the most tolerant life stage and an effective irradiation dose to control white peach scale. Unlike other disinfestation techniques, irradiation does not need to kill the pest immediately to provide quarantine security, and therefore live (but nonviable or sterile) insects may occur with the exported commodity. The objective of an irradiation quarantine treatment is to stop the insect's ability to reproduce and thereby prevent its introduction and establishment into new areas.

Materials and Methods

A colony of white peach scale was started from newly hatched crawler-stage scales collected from old papaya tree trunks from a papaya field in Keaau, HI, in 2005. Crawlers were transferred using a long-haired goat's wool brush to butternut squash, *Cucurbita moschata* Duch., or Irish potato, *Solanum tuberosum* L., tubers in the laboratory (Ashley and Miller 1981). A colony was maintained on its host plants in the laboratory in ventilated 2- or 3-liter round plastic tubs (Sweetheart, Fort Howard Corp., Green Bay, WI, or Berry Plastics Corp., Evansville, IN) at 25.5°C (range 24–27°C), 60% RH (range 50–75%), and a photoperiod of 14:10 (L:D) h. Large Irish potatoes with thin skins, sold as white "salad" potatoes in Hawaii, were the preferred hosts for the irradiation tests owing to their size, color, and smooth skin. Potato tubers remained high-quality hosts in the laboratory for 3–5 mo, which allowed turnover of two to three scale generations. Butternut squash lasts longer (up to 10 mo), but fe-

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male scales brush off easily when bumped during handling, and the squash are often overlarge for viewing scales with a dissecting microscope. Therefore, butternut squash were used to maintain laboratory colonies but were not used for irradiation tests.

White peach scale has four life stages: egg, first-stage nymph (the crawler phase from hatch to settling is ≈12 h), second-stage nymph, and adult (Bennett and Brown 1958, Ball 1980, Gullan and Kosztarab 1997). Approximate development times for females at 24–27°C are 8, 13, and 14 d for the egg, first-stage nymph, and second-stage nymph, respectively (P.A.F., unpublished data). Adult female scales live under the scale cover formed from secreted wax filaments cemented by anal excretions and embedded with the exuviae of the two previous nymphal stages (Gullan and Kosztarab 1997). Adult females begin laying eggs after 14–16 d and live for an average of 50 d. The crawler stage and adult male are mobile and the other stages are sessile. Male and female crawlers can be distinguished by color: females are yellowish pink and males are white. Male nymphs begin producing silk thread after settling at the end of the first-stage nymph and early second-stage nymph and eventually form elongated white cocoons. The tiny winged males hatch from these cocoons after 30 d and live 12–24 h. Adult females require fertilization by males to lay eggs. For purposes of the tests, the adult stage was divided into mature adult females without eggs and adults with eggs. Whether the adult female has eggs cannot always be determined without flipping the scale over, which kills the scale. Therefore the reproductive status of the adult females at the time of treatment was determined partly by age, i.e., at 30–40 d after egg hatch adult females did not have eggs, and at >41 d after hatch adult females had eggs. Survivorship and the reproductive status of adult females were determined at the end of the study by flipping scales and examining for evidence of eggs. Live scales typically have a pinkish color underneath, whereas dead scales are brown.

Approximately 100 crawlers were introduced to each potato to establish cohorts of even age. Potatoes carrying scales were irradiated when most individuals had matured to the desired stage. After irradiation treatment, all irradiated scales and control colonies with emerging adult males were held in a room without rearing container lids for fertilization of eggs of irradiated females. Dose–response tests were conducted with second–stage nymphs, adult females without eggs, and adult females with eggs at a series of irradiation doses between 60 and 150 Gy to determine the most tolerant stage. Individual potatoes with scales served as replicates, and treatments included three to 16 replicates depending on the number of scales on the potatoes. After irradiation treatment, scales on potatoes were examined weekly to determine developmental stage. Untreated controls for each stage were held under identical conditions and examined similarly. In some cases, second and third generation scales on untreated control potatoes numbered in the hundreds. When large numbers of unirradiated scales were present, the surface of the potato was divided with a marking pen into equal sections and counts were made on two or three randomly selected sections then multiplied by the appropriate number to estimate the total. Tests were usually terminated after 2 mo, at which time most irradiated scales had died or potatoes had rotted, and final counts were made of all live and dead females at each stage of development as well as the number that laid eggs. Validation testing was done with large numbers of mature adult females with and without eggs at 150 Gy to confirm the efficacy of this dose as a potential quarantine treatment. Methods for the validation tests were identical to those described above for the dose–response tests. All stages of white peach scale can occur on exported commodities, and gravid adult females are often present. Therefore, the desired response for an effective irradiation treatment was to prevent development of females to the adult stage or reproduction by adult females of the subsequent (F_1) generation.

Irradiation treatment was conducted at a nearby commercial x-ray facility (CW Hawaii Pride LLC, Keaau, HI) by using an electron linear accelerator (5 MeV, model TB-5/15, L-3 Communications Titan Corp., San Diego, CA) at ambient temperature. Dosimeters (Opti-chromic detectors, FWT-70–83M, Far West Technology, Goleta, CA) were placed on the sides and upper surface of potatoes at each dose in each replicate to measure dose variation. The dosimeters were read with a FWT-200 reader (Far West Technology) at 600-nm absorbance to verify the minimum absorbed dose and dose variation in each replicate. To minimize the dose uniformity ratio (the ratio of the maximum/minimum dose), infested potatoes were placed in plastic tubs in a single row perpendicular to the x-ray beam. Dose mapping demonstrated that doses were sometimes lower near the sides and floor of the metal carrier, so the tubs with potatoes were elevated by placement on a cardboard box and positioned in the exact center of the carrier. Each carrier passed in front of the beam in a forward then reverse orientation. The dose uniformity ratio during the white peach scale research was consistently <1.2. After irradiation treatment, infested potatoes were held in ventilated plastic tubs for scale development. The maximum dose received during large-scale validation testing becomes the minimum dose for a quarantine treatment. During validation testing all measured irradiation doses were below 150 Gy.

Data on survivorship were subjected to analysis of variance (ANOVA) after testing for equal variances and normality. Percentage of survivorship data were arcsine transformed to improve normality. Welch's ANOVA was used to evaluate data when a Levene's test suggested variances were unequal (SAS Institute 2002). Means separations were done using a Tukey's test. Scale count data in the F_1 generation were divided by the numbers of individuals in the parent generation, transformed using log $(x + 1)$, and subjected to ANOVA and means separations to evaluate reproductive success.

Table 1. Maturation and reproduction of white peach scale after irradiation of second-stage nymphs

Target dose (Gy)	No replicates	No. second-stage nymphs	Adult females		Adult females with eggs		F_1 generation adult females with eggs
			No	%	No.	%	
Control	7	158	116	72.7a	103	59.0a	196
60	6	172	77	64.9a	62	60.2a	237
90	6	257	147	67.4a	41	11.5b	0
120	6	294	147	62.0a	0	0.0b	0
150	6	359	156	73.3a	0	0.0b	0

Means within a column followed by different letters were significantly different by a Tukey's test ($P < 0.05$).

Results and Discussion

Irradiation of second-stage nymphs resulted in reduced survivorship to the adult stage with eggs ($F = 11.5$; df = 4, 30; $P < 0.0001$) (Table 1). At an irradiation dose of 90 Gy, 11.5% of treated second-stage nymphs matured to the adult stage and laid eggs, whereas 59.0% of untreated scales matured to the adult stage and laid eggs. Although 62.0 and 73.3% of the second stage nymphs treated with 120 or 150 Gy, respectively, developed to the adult stage, none laid eggs (Table 1).

Irradiation of adult females with no eggs resulted in a reduced number of adult females with eggs compared with controls ($F = 11.9$; df = 4, 29; $P < 0.001$) (Table 2). Reproduction in adults in the 60- and 90-Gy irradiation treatments was not significantly different from untreated control adults. In the 120 and 150 Gy treatments, 0.6 and 0.0% of adults produced eggs, respectively, which was significantly lower than the other treatments (Tukey's test; $P < 0.05$). None of the progeny of adult female scales irradiated at any dose became F_1 adults with eggs (Table 2).

Irradiation of adult females with eggs resulted in a reduction of F_1 adult females compared with the untreated control treatment ($F = 23.2$; df = 4, 42; $P < 0.001$) (Table 3). Production of F_1 females with eggs was significantly lower in the 60- and 90-Gy treatment compared with the untreated controls (Tukey's test; $P < 0.05$). F_1 females with eggs produced F_2 second stage nymphs as did the controls. No F_1 adult females with eggs were produced in the 120- and 150-Gy irradiation treatments (Table 3).

The reproductive adult was the most tolerant stage tested. At an irradiation dose of 90 Gy, second-stage nymphs (Table 1) and adults without eggs (Table 2) produced no F_1 generation adults with eggs, whereas adults with eggs did successfully produce F_1 adults with eggs (Table 3).

Irradiation of reproductive females with irradiation doses of 120 or 150 Gy completely prevented the production of F_1 females with eggs. An irradiation dose of 150 Gy was selected as a potential quarantine treatment because it conforms to the current generic dose approved for tephritid fruit flies. Also, the use of conservative doses will help ensure irradiation treatments never fail. In large-scale validation tests, irradiation of 35,424 adult female scales with and without eggs at an irradiation dose of 150 Gy (measured doses were 128–149 Gy) resulted in no successful development of F_1 adults with eggs (Table 4). During validation testing, ≈75% of the adult females had eggs at the time of treatment. The highest dose measured during validation testing becomes the lowest dose for a quarantine treatment. Therefore, irradiation doses ≥150 Gy prevent reproduction and generation turnover in white peach scale. Lowering the required irradiation dose will shorten treatment time, which will lower treatment costs and increase capacity of the irradiation facility.

In the past, the U.S. Department of Agriculture (USDA) has used 99.9968% efficacy (probit 9) as the basis for approving quarantine treatments, particularly for tephritid fruit flies (Follett and Neven 2006). To achieve probit 9 mortality at the 95% confidence level, a minimum of 93,613 insects must be tested with no survivors (Couey and Chew 1986). In recent years, the USDA has been flexible in approving quarantine treatments with less than probit 9 numbers of insects, particularly if the potential economic and environmental impact of the pest, should it be introduced, is low. Other countries (Japan, Australia, and New Zea-

Table 2. Maturation and reproduction of white peach scale after irradiation of adult females without eggs

Target dose (Gy)	No. replicates	No. adult females	Adult females with eggs		F_1 generation adult females with eggs[a]	F_2 generation second-stage nymphs
			No.	%		
Control	4	93	43	50.7ab	868a	Yes
60	10	358	133	38.9bc	0b	No
90	8	536	421	87.0a	0b	No
120	5	418	2	0.6c	0b	No
150	3	170	0	0.0c	0b	No

Means within a column followed by different letters were significantly different by a Tukey's test ($P < 0.05$).
[a] ANOVA performed on log(F_1 generation females with eggs/parental females with eggs +1) was significant ($P < 0.05$).

Table 3. Maturation and reproduction of white peach scale after irradiation of adult females with eggs

Target dose (Gy)	No. replicates	No. adult females with eggs	F_1 generation adult females with eggs[a]	F_2 generation second-stage nymphs
Control	6	94	677a	Yes
60	12	512	311b	Yes
90	11	593	294b	Yes
120	16	925	0b	No
150	3	152	0b	No

Means within a column followed by different letters were significantly different by a Tukey's test ($P < 0.05$).
[a] ANOVA performed on log(F_1 generation females with eggs/parental females with eggs +1) was significant ($P < 0.05$).

land) accept quarantine treatment efficacy at 99.99% (at the 95% confidence level), which is obtained by treating a minimum of 29,956 insects (nominally 30,000 insects) with no survivors (Follett and Neven 2006). During validation testing, 35,424 white peach scale adults were irradiated at 150 Gy with no reproduction in the F_1 generation. Therefore, irradiation of white peach scale with a minimum dose of 150 Gy exceeds the 99.99% quarantine treatment efficacy requirement.

A generic treatment is a single treatment to control a broad, taxonomically related group of pests without affecting the quality of a wide range of commodities (Follett and Armstrong 2004, Follett and Neven 2006). The rationale behind generic irradiation doses is that information on radiotolerance for a limited number of species could be extrapolated to related species to arrive at a single effective dose for the group. Irradiation is the ideal technology for developing generic treatments because radiation—from an isotope source such as cobalt-60, or x-rays—penetrates fruit easily and is effective against insects at doses that generally do not injure the fruit. Generic irradiation treatments will accelerate the approval of irradiation quarantine treatments for specific crops and expedite new trade in agricultural products because research will no longer be needed for each quarantine pest and commodity.

Scale insects are currently covered in the United States under new regulations that specify a generic (or default) dose of 400 Gy for all insects except lepidopteran pupae and adults (Federal Register 2006). Because coccid and diaspidid scales are quarantine pests on many traded fruits and vegetables, establishment of a specific generic dose lower than 400 Gy that is predicted to be effective against all scale insects would be beneficial. However, the number of irradiation studies on scale insects is small (IDIDAS 2005). Species studied to date include San Jose scale, *Quadraspidiotus perniciosus* (Comstock) (Angerilli and Fitzgibbon 1990); green scale, *Coccus viridis* (Green) (Hara et al. 2002); and coconut scale (Follett 2006). Angerilli and Fitzgibbon (1990) showed that irradiation doses from 300 to 600 Gy increased mortality of San Jose scale on apples in cold storage. Hara et al. (2002) showed that an irradiation dose of 250 Gy (the lowest dose tested) stopped reproduction in green scale. Neither of these studies conducted the large-scale validation tests that are required to recommend an irradiation quarantine treatment and demonstrate quarantine security. Follett (2006) demonstrated that 150 Gy controlled coconut scale. The current study, and Follett (2006), use no reproduction in the F_1 generation as the desired response and present data from large-scale validation tests to confirm the efficacy of a potential irradiation dose. If radiotolerance in white peach scale and coconut scale are representative of the group, a generic dose of 150 Gy could be proposed; however, research on additional species of scale insects is needed to support these findings before a case can be made for a generic irradiation dose.

The dose uniformity ratio for commercially irradiated papaya in Hawaii is ≈1.5, meaning some fruit receive a dose 1.5 times the target dose. The current requirement for a minimum absorbed dose of 400 Gy (to control white peach scale in addition to fruit flies) results in some papayas receiving a dose of 600–650 Gy. Research indicates that papayas are relatively tolerant of irradiation although storage temperature, fruit maturity at harvest, and temperature during the growing season all may influence postirradiation ripening behavior (Paull 1996). Irradiation may actually improve the quality of exported papayas. Papayas irradiated at 500–750 Gy showed delayed ripening, more uniform softening, and longer shelf life compared with unirradiated papayas (Akamine and Goo 1977a,b; Moy et al. 1973; Paull 1996). The predominant papaya variety now exported from Hawaii to the U.S.

Table 4. Large-scale validation tests irradiating white peach scale adult females with and without eggs

Target dose (Gy)	No. replicates	Measured doses	No. adult scales treated[a]	No. F_1 second-stage nymphs	No. F_1 adults with eggs
Control	33	—[b]	316	—[c]	5,480
150	519	128–149	35,424	2165	0

[a] Approximately 75% of white peach scale adults had eggs, and 25% had no eggs at the time of treatment.
[b] Untreated.
[c] Stage was not counted

mainland using irradiation treatment is the papaya ring spot-resistant cultivar 'Rainbow'. The affects of irradiation on Rainbow papayas have not been adequately tested. Lowering the commercial irradiation dose for papaya to 150 Gy for quarantine control of fruit flies and white peach scale (resulting in commercial doses up to 250–300 Gy) might alter papaya ripening behavior and shelf life.

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References Cited

Akamine, E. K., and T. Goo. 1977a. Respiration, ethylene production and shelf life extension in irradiated papaya fruit after storage and simulated shipping condition. Hawaii Agric. Ext. Stn. Tech. Bull. 93: 12–18.

Akamine, E. K., and T. Goo. 1977b. Effects of gamma irradiation on shelf life extension of fresh papaya (*Carica papaya* L. var. Solo). Hawaii Agric. Ext. Stn. Res. Bull. 165: 23–38.

Angerilli, N.P.D., and F. Fitzgibbon. 1990. Effects of cobalt gamma radiation on San Jose scale (Homoptera: Diaspididae) survival on apples in cold and controlled-atmosphere storage. J. Econ. Entomol. 83: 892–895.

Ashley, T. R., and D. Miller. 1981. Propagation of the white peach scale, *Pseudaulacaspis pentagona,* and a technique for producing virgin females. J. Ga. Entomol. Soc. 16: 501–504.

Ball, J. C. 1980. Development and fecundity of the white peach scale at two constant temperatures. Fla. Entomol. 63: 188–194.

Bennett, F. D., and S. W. Brown. 1958. Life history and sex determination in the diaspine scale, *Pseudaulacaspis pentagona* (Targ.) (Coccoidea). Can. Entomol. 9: 317–324.

Couey, H. M., and V. Chew. 1986. Confidence limits and sample size in quarantine research. J. Econ. Entomol. 79: 887–890.

Federal Register. 2006. Treatments for fruits and vegetables. rules and regulations 71: 4451–4464.

Follett, P. A. 2004. Irradiation to control insects in fruits and vegetables for export from Hawaii. Radiat. Phys. Chem. 71: 163–166.

Follett, P. A. 2006. Irradiation as a phytosanitary treatment for *Aspidiotus destructor* (Homoptera: Diaspididae). J. Econ. Entomol. 99: 1138–1142.

Follett, P. A., and J. W. Armstrong. 2004. Revised irradiation doses to control melon fly, Mediterranean fruit fly and oriental fruit fly (Diptera: Tephritidae) and a generic dose for tephritids. J. Econ. Entomol. 97: 1263–1268.

Follett, P. A., and Z. Gabbard. 1999. Efficacy of the papaya vapor heat quarantine treatment against white peach scale in Hawaii. HortTechnology 9: 506.

Follett, P. A., and L. G. Neven. 2006. Current trends in quarantine entomology. Annu. Rev. Entomol. 59: 359–385.

Gullan, P. J., and M. Kosztarab. 1997. Adaptations in scale insects. Annu. Rev. Entomol. 42: 23–50.

Hara, A. H., J. A. Yalemar, E. B. Jang, and J. H. Moy. 2002. Irradiation as a possible quarantine treatment for green scale *Coccus viridis* (Green) (Homoptera: Coccidae). Postharvest. Biol. Technol. 25: 349–358.

[IDIDAS] International Database on Insect Disinfestation and Sterilization. 2005. International Database on Insect Disinfestation and Sterilization. International Atomic Energy Agency, Vienna, Austria. (http://www-ididas.ieae.org/ididas/).

Moy, J. H., E. K. Akamine, N. Wenkam, A. M. Dollar, M. Hanaoka, H. Y. Kao, W. L. Liu, and L. M. Revetti. 1973. Tolerance, quality, and shelf life of gamma-irradiated papaya grown in Hawaii, Taiwan and Venezuela. In: Radiation preservation of food. International Atomic Energy Agency, Vienna, Austria.

Paull, R. E. 1996. Ripening behavior of papaya (*Carica papaya* L.) exposed to gamma irradiation. Postharvest Biol. Technol. 7: 359–370.

[USDA–APHIS–PPQ] U.S. Department of Agriculture–Animal and Plant Health Inspection Service–Plant Protection and Quarantine. 1996. Movement of papaya fruit *Carica papaya* from Hawaii into other regions of the United States: qualitative, pathway-initiated pest risk assessment. USDA–APHIS–PPQ, Riverdale, MD.

SAS Institute. 2002. JMP user's guide. SAS Institute, Cary, NC.

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