

REVIEW ARTICLE

Expanding radiation quarantine treatments beyond fruit flies

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- Abstract**
- 1 The potential of ionizing radiation as a disinfestation treatment for insects other than tephritid fruit flies is discussed. Radiation quarantine treatments are unique in that insects are not killed immediately but rendered sterile or incapable of completing development.
 - 2 The most tolerant insect stage to radiation is that which is most developed. Female insects, but not always mites, are sterilized with equal or lower doses than males.
 - 3 Insects irradiated with sterilizing doses usually have shorter longevities than non-irradiated ones. Low oxygen conditions often increase tolerance to radiation.
 - 4 Insects in diapause are not more tolerant of radiation than non-diapausing ones.
 - 5 Some pests of several groups, such as aphids, whiteflies, weevils, scarab beetles, and fruit flies, may be controlled with doses ≤ 100 Gy. Some lepidopterous pests and most mites require about 300 Gy. Stored product moths may require as much as 1 kGy to sterilize, and nematodes could need >4 kGy.
 - 6 Even though application of irradiation to pallet-loads of produce could mean that up to three times the minimum required dose is applied to the perimeter of the pallet, many fresh commodities tolerate doses required for quarantine security against many quarantined pests. Irradiation is arguably the most widely applicable quarantine treatment from the standpoint of commodity quality.

Keywords Disinfestation, gamma radiation, ionizing radiation, irradiation, postharvest, quarantine treatment, radiation.

Introduction

Ionizing irradiation was suggested as a possible quarantine treatment 70 years ago, and considerable research to that effect has been done during the past 45 years (Burditt, 1994). However, the commercial application of irradiation as a quarantine treatment has only occurred on a continuing, albeit very limited, basis since 1995; over 200 tonnes of various tropical fruits have been shipped from Hawaii to the northern United States for irradiation and distribution to retail markets (Hallman, 1999). The quarantine treatment world may be poised on the verge of broader acceptance of irradiation as a disinfestation method. The Hawaiian experience has been positive, and there are plans to build an irradiator on the islands in order to expedite shipments.

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Recent surveys have shown that up to 80% of U.S. consumers would buy food labelled as irradiated (Shee, 1998). The U.S. is considering setting minimum treatment doses for imported fruits that could be infested with 10 fruit fly species (APHIS, 1996). On 14 April 1999 the first shipment of guavas (five tonnes) irradiated for disinfestation of Caribbean fruit fly, *Anastrepha suspensa* (Loew), was shipped from Florida to California, and shipments of mangoes are planned for 2000. Florida has petitioned California to accept sweetpotatoes, *Ipomea batatas*, irradiated as a quarantine treatment against the sweetpotato weevil, *Cylas formicarius elegantulus* (Summers).

The use of methyl bromide fumigation as a disinfestation treatment has been debated for several years, as it is considered a significant stratospheric ozone-depleting substance, and alternatives are desired (Ross, 1999). Radiation is a good alternative to methyl bromide, as it is probably the most widely applicable quarantine treatment from the standpoint of fruit quality. Many

key fruits that cross quarantine boundaries, such as apples, cherries, papayas and blueberries, tolerate >700 Gy, which is more than enough to disinfest fruits of most quarantine insect pests. Other fruits, such as citrus and mangoes, may only tolerate lower doses (250–500 Gy), but they are still sufficient to disinfest these fruits of fruit flies and other insects.

Insufficient research has been done to support the use of irradiation as a quarantine treatment against pests, with the exception of tephritid fruit flies. Although much research has been done that is applicable to insects besides fruit flies, none of this research has been done using enough insects to confirm that the high levels of efficacy demanded of a quarantine treatment will be met. Acceptance of a quarantine treatment requires that very high numbers of insects are treated at the recommended dose with no survivors. For example, the U.S. generally demands that quarantine treatments against fruit flies satisfy the probit 9 (99.9968%) level of efficacy (Shannon, 1994), which means that, at the 95% confidence level, a minimum of 93 600 insects must be treated with no survivors (Couey & Chew, 1986). Other countries require that 30 000 insects be treated with no survivors. Regulatory agencies might consider lower numbers of insects treated for pests of relatively lower risk that are difficult to rear. In any case, irradiation efficacy tests have rarely been done with anywhere near thousands of insects for pests other than Tephritidae.

With all other quarantine treatments (heat, cold, fumigation), efficacy is measured by acute mortality. Radiation doses required to provide near 100% acute mortality of insects would be well over 1 kGy, and would damage almost all fresh agricultural commodities. The goal of a quarantine treatment is to prevent the establishment of exotic pests. This goal could be accomplished by less extreme measures than acute mortality, such as prevention of adult development or sterility. Although irradiation does not kill insects quickly at doses tolerated by fresh commodities, insect development can be halted and sterility induced by doses that are tolerated by many fruits and vegetables. Irradiation is unique among quarantine treatments in that sublethal doses are effective in providing quarantine security. The effects of irradiation are experienced most severely by actively growing cells and the very large molecules of DNA (von Sonntag, 1987). That is why growth and sterility are achieved by much lower doses than those necessary to achieve acute mortality and the effects of moderate doses of radiation may not become manifest until the next moult.

The goal of radiation quarantine treatments against fruit flies is prevention of adult emergence from irradiated eggs and larvae; pupae and adults are not considered to be present in packed fruits. Although a lower dose might allow for emergence of sterile adults, prevention of adult emergence precludes the possibility of exotic flies being found in monitoring traps and triggering expensive delimiting and eradication efforts. For many other quarantine pests (Coleoptera, Homoptera, Thysanoptera, Acarina, Mollusca), adults may be present in the packed commodity. For many Lepidoptera, pupae as well as larvae may be present. Very high doses are required to prevent adult emergence from lepidopteran pupae late in their development; therefore, adult sterility will also be the objective for lepidopterous quarantine pests present as pupae in the commodity.

The irradiation process

Absorbed radiation dose is measured in grey (Gy), which is the amount of ionizing radiation energy absorbed by the material being irradiated in joules per kilogram of material. (The discontinued unit is the rad, which equals 0.01 Gy.) There are four sources of ionizing radiation for use on food: gamma irradiation with the isotope of (1) cobalt⁶⁰ or (2) caesium¹³⁷, (3) electron beam (beta particles) or (4) X-ray (bremsstrahlung) radiation produced when an electron beam strikes a high density converter. The energy efficiency of the latter process is low; only about 5% of the energy in the beta particles is converted to bremsstrahlung. Bremsstrahlung radiation is very similar to gamma ray from isotopes; both can penetrate pallet-loads of produce. Electron beam radiation penetrates only a few centimetres and, thus, is limited to small products, such as shallow boxes of berries, passing the irradiation source on a conveyor line. All other things being equal, such as dose rate, dose uniformity and product density, there is no difference in effect among the four ionizing radiation sources used on food items.

Two different procedures have been developed for commercial irradiation. (1) A chamber is loaded with the materials to be irradiated and the radioactive source is raised into the chamber for the appropriate amount of time to achieve the required absorbed dose. (2) A conveyor system carries the materials to be irradiated past the exposed source at a speed that will give the required absorbed dose. The unidirectional nature of electron beam and bremsstrahlung radiation require that they be applied in a conveyor system.

Concerns about irradiation quarantine treatment research

This review uses data from a multitude of publications to suggest possible quarantine treatment doses for a variety of pests.

Dosimetry

Making accurate dose measurements is fundamental to any dose-response work. Most radiation research has been done with the radioactive isotopes cobalt⁶⁰ and caesium¹³⁷. These isotopes emit gamma rays at a highly predictable rate, which can be calculated years into the future. The half lives of ⁶⁰Co and ¹³⁷Cs are 5.3 and 30 years, respectively. Many researchers have obviously counted on the predictability of radiation dose when doing research with irradiators which were calibrated when they were installed. However, the predicted dose given in decay charts is usually the dose found at the centre of the irradiation chamber when a specific material was irradiated during calibration. Most of the irradiation chamber will be subjected to a higher dose. Denser materials will attenuate the dose reaching the centre slightly, while less dense materials will, of course, have the opposite effect. Geometry, homogeneity and arrangement of the product to be irradiated can also affect dose. It is therefore imperative that routine dosimetry traceable to accredited standards be performed throughout the research. One must take into account factors, such as temperature, age and light, that may also affect the dosimeters. Farrar (1999) and the

American Society for Testing & Materials (1998) present information that should be recorded when documenting dosimetry. Farrar (1999) considers that many reports on quarantine irradiation research are of little value because of insufficient information on dosimetry.

Dose rate

The rate at which any process is done can alter the efficacy of that process; this also holds true for radiation quarantine treatments. Usually, the faster the rate, the greater the effect. Dose rate is often omitted from reports of radiation quarantine treatment studies. In this paper the dose rate is given when I felt it was of relevance.

Brown & Davis (1972) applied 30 Gy to grain mite, *Acarus siro* L., eggs at different dose rates and observed a consistent but modest increase (not statistically tested) in mortality for those eggs treated at 25.8 Gy/min vs. those treated at 3.6 Gy/min. A dose of 20 Gy applied to female Mexican fruit fly, *Anastrepha ludens* (Loew), pharate adults resulted in egg hatches from 0 to 44% depending on the dose rate (Rhode *et al.*, 1961). However, dose rates were all much slower (0.1–0.9 Gy/min) than would be applied commercially. A marked difference in the effect of dose rate was noted by Lester & Wolfenbarger (1990). When 10 Gy were applied to third-instar Mexican fruit flies at the rate of 40 Gy/min, 92% of the adults failed to emerge, whereas only about 17% failed to emerge when the dose was 1 or 0.1 Gy/min. Although results of Burditt *et al.* (1989) are in general agreement with increasing dose rate being related to increased efficacy, there is one curious exception: 67.5 Gy applied at the rate of 201.5 Gy/min yielded lower efficacies ($P < 0.05$) than 52 Gy/min.

However, in the same study cited above (Lester & Wolfenbarger, 1990), percentage reduction of adult Mexican fruit fly from irradiated third instars subjected to 20 Gy was 99–100%, regardless of whether the dose rate was 80 or 0.2 Gy/min. This indicates that at the high levels of control where quarantine security lies (>99.9%), the effect of dose rate on insect response may not be significant. Dose rate may be more critical to commodity quality, especially when tolerance of a given commodity is not much higher than the doses that may be applied on a commercial scale to provide quarantine security. In any case, because of the possible effect of dose rate on efficacy and commodity quality, the dose rate used in research should be similar to the dose rate that will be applied commercially. A faster rate applied commercially than that used in the research will probably not be detrimental to efficacy, although it could negatively affect commodity quality for produce that is not very tolerant of radiation.

Dose uniformity ratio

Because radiation from radioactive isotopes applied on a commercial scale originates from a source and spreads as distance from the source increases, the ratio of maximum to minimum dose absorbed by the product (dose uniformity ratio) will be greater with irradiation than with virtually any other quarantine treatment. Radiation attenuates as it passes through objects, thus reducing absorbed dose even further. The dose uniformity ratio in a commercial isotopic irradiator may be as

much as 3:1. This means that irradiated produce must be able to tolerate up to three times the minimum dose needed to achieve efficacy against the pest. Fortunately, many fruits tolerate these doses well, although some do not. Irradiation appears to be the most broadly applicable quarantine treatment from the standpoint of fruit quality. In any case, because of the large dose uniformity ratio as well as concerns with time, overall cost and quality, researchers should always strive to achieve the lowest possible effective dose.

Sterility vs. longevity

Radiation at minimum absorbed doses to prevent reproduction usually shortens the longevity of arthropods, although there are some occasions where no relationship is noted and fewer still where arthropods irradiated with sterilizing doses have lived longer than unirradiated ones (Table 1). Stored product arthropods are primarily represented in this table because they have been studied extensively for the effect of radiation on reproduction and longevity.

Although insects may live for considerable periods of time after irradiation, they often feed little, thus reducing the risk of additional damage occurring to the commodity. For example, although plum curculio, *Conotrachelus nenuphar* (Herbst), adults live for up to 3 weeks after receiving the minimum absorbed dose for sterility (80 Gy), they do not make feeding or ovipositional slits at this dose (G.J. Hallman, unpublished data).

Hypoxia

Low oxygen tension is one factor known to appreciably reduce the efficacy of irradiation. Doses of 62 and 110 Gy were required in air and carbon dioxide, respectively, to sterilize female pharate adult screwworms, *Cochliomyia homnivorax* (Coquerel) (Baumhover, 1963). Female pharate adults of the house fly, *Musca domestica* L., were completely sterilized when irradiated with 30 Gy in air, but not in carbon dioxide (Smittle, 1967). An increase in radiation dose of 28–60% was required to achieve the same level of sterility to late male pharate adult Mediterranean fruit flies, *Ceratitis capitata* (Wiedemann), when irradiated in a nitrogen, carbon dioxide or helium atmosphere or under a partial vacuum compared with air (Hooper, 1971; Ohinata *et al.*, 1977). Male boll weevils, *Anthonomus grandis* Boheman, were completely sterilized with 80 Gy applied in air, but 1–3% remained fertile when irradiated in nitrogen (Earle *et al.*, 1979). Additionally, after 21 days all of the weevils irradiated in air had died, whereas 20% of those irradiated in nitrogen were still alive. All of the above studies were performed with insects held under hypoxic conditions for relatively short periods of time (5–90 min), starting before and continuing through the irradiation process. When confused flour beetles, *Tribolium confusum* Jacquelin du Val, were held in 100% carbon dioxide for 30 min ending 1 h prior to irradiation, no reduction in mortality over the control was found (Tilton *et al.*, 1965).

Temperature

Temperature is a significant factor to consider in radiation disinfestation because most perishable commodities will be at

Table 1 Longevity (near 100%) of adult arthropods irradiated with the estimated minimum absorbed dose to prevent reproduction

ORDER: Family, species and sex	Sterilization dose (Gy) ¹	Time to 100% mortality	Mortality of unirradiated control during same time (%)	Reference
COLEOPTERA:				
Anobiidae				
Cigarette beetle, <i>Lasioderma serricorne</i> (F.)	♂ 175	7 weeks	88	Tilton <i>et al.</i> (1966b)
	♀ 250	6 weeks	88	Tilton <i>et al.</i> (1966b)
Bostrichidae				
Lesser grain borer, <i>Rhyzopertha dominica</i> (F.)	50	> 30 weeks	100	Watters & MacQueen (1967)
Bruchidae				
Cowpea weevil, <i>Callosobruchus maculatus</i> (F.)	100	7 d	90	Dongre <i>et al.</i> (1997)
Cucujidae				
Sawtoothed grain beetle, <i>Oryzaephilus surinamensis</i> (L.)	125	4 weeks	1	Tunçbilek (1997)
Curculionidae				
Boll weevil, <i>Anthonomus grandis</i> Boheman	70	13 d	low	Davich & Lindquist (1962)
Plum curculio, <i>Conotrachelus nenuphar</i> (Herbst)	80	3 weeks	low	G.J. Hallman (unpublished data)
Sweetpotato weevil, <i>Cylas formicarius-elegantulus</i> (Summers)	150	< 10 d	4	Dawes <i>et al.</i> (1987)
	150	33 d	35	G.J. Hallman (unpublished data)
Granary weevil, <i>Sitophilus granarius</i> (L.)	100	3 weeks	40	Watters & MacQueen (1967)
	100	3 weeks	6	Brown <i>et al.</i> (1972)
Maize weevil, <i>S. zeamais</i> Motschulsky	100	5 weeks	20	Brown <i>et al.</i> (1972)
Dermestidae				
Black carpet beetle, <i>Attagenus unicolor</i> (Brahm)	130	4 weeks	100	Tilton <i>et al.</i> (1966a)
<i>Trogoderma glabrum</i> (Herbst)	175	7 weeks	100 (6 weeks) ²	Tilton <i>et al.</i> (1966b)
<i>T. inclusum</i> LeConte	200	5 weeks	100	Brower & Tilton (1972)
Warehouse beetle, <i>T. variabile</i> Ballion	♀ 100	4 weeks	100	Brower & Tilton (1972)
	♂ 250	4 weeks	100	Brower & Tilton (1972)
Tenebrionidae				
Depressed flour beetle, <i>Palorus subdrepessus</i> (Wollaston)	♀ 400	3 weeks	0	Brower (1973a)
	♂ 400	3 weeks	0	Brower (1973a)
Yellow mealworm, <i>Tenebrio molitor</i> L.	♀ 50	7 weeks	98	Brower (1973b)
	♂ 150	3 weeks	47	Brower (1973b)
Dark mealworm, <i>T. obscurus</i> F.	♀ 100	2 weeks	37	Brower (1973b)
	♂ 100	2 weeks	14	Brower (1973b)
DIPTERA:				
Culicidae				
Malaria mosquito, <i>Anopheles quadrimaculatus</i> Say	♀ 130	3 weeks	100	Davis <i>et al.</i> (1959)
	♂ 130	3 weeks	100	Davis <i>et al.</i> (1959)
LEPIDOPTERA:				
Pyralidae				
Almond moth, <i>Cadra cautella</i> (Walker)	♀ 300	12 d	72	Cogburn <i>et al.</i> (1973)
	♂ 300	14 d	89	Cogburn <i>et al.</i> (1973)
Tortricidae				
Codling moth, <i>Cydia pomonella</i> (L.)	♀ 400	18 d	100 (14 d) ²	Proverbs & Newton (1962)
ACARI:				
Acaridae				
Grain mite, <i>Acarus siro</i> L.	300	25 weeks	100	Burkholder <i>et al.</i> (1966)
Mold mite, <i>Tyrophagus putrescentiae</i> (Schränk)	♀ 300	80 d	40	Ignatowicz (1997)
	♂ 300	80 d	80	Ignatowicz (1997)
Tetranychidae				
Carmine spider mite, <i>Tetranychus cinnabarinus</i> (Boisduval)	♀ 300	15 d	22	Ignatowicz & Banasik-Solgala (1999)
Two-spotted spider mite, <i>T. urticae</i> Koch	♀ 300	18 d	18	Ignatowicz & Banasik-Solgala (1999)
<i>T. urticae</i> (diapausing)	300	40 d	100 (34 d) ²	Lester & Petry (1995)

¹Minimum absorbed dose that might completely prevent reproduction; inferred from references cited and reviews by Brower & Tilton (1985) and Hallman (1998a).

²100% values followed by time periods for 100% mortality of controls in parentheses indicate instances where irradiated arthropods lived longer than controls.

cool temperatures after irradiation and often before and during irradiation. The little data that address the effect of temperature on irradiation of insects suggest cool temperatures might lessen

the effects of radiation (Nelson & Stafford, 1972). Storage at moderately low temperatures after any disinfestation treatment will frequently delay mortality, but because mortality is not the

objective of irradiation quarantine treatments, this delay would not be important. The only study I could locate that examined the effect of temperature on sterility found no difference when 0–1-day-old adult granary weevils, *Sitophilus granarius* (L.), were held at 15 or 30 °C before, during or after irradiation with 31–250 Gy (Pendlebury, 1966).

When efficacy is measured by prevention of adult emergence, lower temperatures apparently lessened the effect of irradiation of Queensland fruit fly, *Bactrocera tryoni* (Froggatt), at doses of 18–30 Gy (Macfarlane, 1966). It is not possible from the data to determine if the difference is manifested at minimum absorbed doses needed for quarantine security (60–75 Gy). Seo *et al.* (1973) found no effect of storage at 7.2 or 10 °C vs. ambient temperatures on adult emergence from larvae of oriental fruit fly, *Bactrocera dorsalis* (Hendel), or Mediterranean fruit fly irradiated with 102 or 225 Gy (oriental) or 246 Gy (Mediterranean). Their studies indicated that the minimum absorbed doses required for quarantine security of oriental and Mediterranean fruit flies were about 250 and 225 Gy, respectively. There is some evidence that these doses might be excessive (Hallman, 1999).

Diapause

Because diapause is a state of arrested development, it seems logical to suspect that insects in diapause would be more tolerant of radiation than non-diapausing ones because rapidly developing tissue is most susceptible to radiation damage. However, the literature does not show diapausing arthropods to be significantly more tolerant of radiation than those not in this state. In fact, Burditt (1986) found that 34.7% of non-diapausing and 2.9% of diapausing cocooned, fifth-instar codling moths in walnuts pupated following irradiation with 177 Gy. Although Lester & Petry (1995) stated that diapausing two-spotted spider mites, *Tetranychus urticae* Koch, appeared to be more tolerant to radiation than mites not in diapause, the difference is slight and probably insignificant. Raun *et al.* (1967) found diapausing European corn borer, *Ostrinia nubilalis* (Hübner), fifth instars suffered less somatic damage after irradiation than non-diapausing larvae; however, the latter were probably less developed than the diapausing larvae. Diapausing blueberry maggots, *Rhagoletis mendax* Curran, were less tolerant of radiation-induced prevention of pupation than non-diapausing larvae of the very closely related *Rhagoletis pomonella* (Walsh) (Hallman & Thomas, 1999). Ignatowicz (1996) found khapra beetle, *Trogoderma granarium* Everts, larvae equally susceptible to radiation-induced mortality regardless of whether or not they were in diapause. Diapausing khapra beetle larvae are more resistant to insecticides and fumigants than non-diapausing larvae. Hallman

(unpublished data) found that diapausing plum curculio were more susceptible to radiation than non-diapausing ones.

Diapausing insects must survive additional hurdles not faced by non-diapausing insects in order to survive and reproduce; i.e. they must be subject to the proper conditions to break diapause for a lengthy period of time before they can seek out a mate and reproduce. Because of this, the possibility that pests in diapause transported to a new area would establish an infestation is considerably less than with non-diapausing insects.

Post-treatment re-infestation

Quarantine treatment research involves treating large numbers of insects in situations where one survivor can require that the treatment be done anew at a higher dose. A radiation treatment is further complicated by the fact that insects treated at the doses used for quarantine purposes do not die quickly and must be held until they do die to determine if the treatment has been successful as measured by failure to develop or reproduce. Under this scenario, opportunities for misplaced insects causing erroneous results are great, and this has apparently happened in published research. Balock *et al.* (1966) felt that two adult Mediterranean fruit flies emerging from larvae up to 5 days-old in papayas irradiated with 100 Gy were the product of post-treatment reinfestation. Burditt & Hungate (1988) suspected that one western cherry fruit fly, *Rhagoletis indifferens* Curran, found in cherries irradiated with 127 Gy entered after the cherries were irradiated. Six surviving oriental fruit flies after a papaya heat treatment were dismissed as 'accidental reinfestation' (Couey & Hayes, 1986). One wonders how many flaws like this in the literature went unsuspected.

Doses for different quarantine pest groups

Objectives for measuring radiation efficacy depending on the most advanced stage present are suggested in Table 2. Prevention of the adult stage or sterility is almost invariably achieved with increasingly higher doses as the insect develops so that the most advanced stage present is the most tolerant to radiation. The most notable (and perhaps questionable) exception I have found is the body louse, *Pediculus humanus humanus* L.; adults were sterilized with ≤ 100 Gy, whereas nymphs required > 500 Gy (Cole *et al.*, 1959). If an insect is ready to change to the next stage this transformation will not be stopped with doses of a few hundred grey. Lower doses, however, will prevent it from reaching the next moult after that. Consequently it is difficult to prevent the adult from emerging once it is completely developed in the pupal case.

Table 2 Suggested objectives of radiation quarantine treatments based on most advanced growth stage found on commodity.

Most advanced stage	Objective of treatment
Egg	Prevent development beyond first instar
Early instars (simple metamorphosis)	Prevent late instar or adult
Early instar (complete metamorphosis)	Prevent late instar, pupariation, or pupation
Late instar	Prevent pupation or adult emergence
Pupa	Adult sterility
Adult	Sterility

Most groups of quarantine pests will have actively reproducing adults present, so prevention of reproduction will be the objective of an irradiation treatment against them. Key exceptions are the tephritid fruit flies and boring Lepidoptera. The objective of quarantine treatments against fruit flies is prevention of adult emergence to eliminate the possibility of an adult, although sterile, from being caught in a monitoring trap and triggering an expensive eradication response. Prevention of adult emergence could also be the goal against Noctuidae and Tortricidae that do not pupate in the infested commodity. For those Pyralidae and Tortricidae that pupate in the commodity, but for which there is no risk of the adult being present, the goal must still be prevention of reproduction, as adults will readily emerge from individuals irradiated late in the pupal stage.

Hallman (1998a) observed that female insects (but not always mites) are of equal or less tolerance to radiosterility than males. The three exceptions found in the literature are debatable. First, Sharp's (1995) contention that female sweet potato weevils are more tolerant of radiation than males contradicts a previous study (Dawes *et al.*, 1987). Second, although fertility in male alfalfa weevils, *Hypera postica* (Gyllenhal), declined at a faster rate than females as dose increased, both sexes still required about 45 Gy for complete sterility (Burgess & Bennett, 1966; Ferrer *et al.*, 1975). Third, in a review, Brower & Tilton (1985) list doses of 80 and 50 Gy as necessary to sterilize females and males, respectively, of the dermestid *Anthrenus flavipes* LeConte. No reference is given, and I have not been able to locate the actual study. The fact that female insects are generally no more tolerant than males to radiosterility is fortuitous because only the female needs to be sterilized in a quarantine treatment. Considerable research has been done on doses required to sterilize many male insects for sterile insect release programmes. It is almost assured that the dose required to sterilize the adult male insect will also sterilize the female.

Stored product pests

Much research has been conducted on the use of radiation to control stored product pests (Brower & Tilton, 1985; Hallman, 1998a; Johnson & Marcotte, 1999). Stored product pests, especially some Lepidoptera such as the Angoumois grain moth, *Sitotroga cerealella* (Olivier), and the Indian meal moth, *Plodia interpunctella* (Hübner), are among the most tolerant arthropods to irradiation, requiring doses up to and exceeding 1 kGy for sterilization. Fortunately, stored products tolerate these high doses. Many key stored product pests were transported world-wide years ago. However, many new ones become of quarantine significance as trade increases. The khapra beetle may be the most notorious stored product pest quarantined by those countries which do not yet have it, which includes most of the Western Hemisphere, Oceania, the eastern half of Asia & Australia. Nair & Rahalkar (1963) found that a dose of 60 Gy applied to pupae was sufficient to prevent hatching of eggs from the emerged adults; a larger dose will most likely be required to sterilize the adult stage. Radiation research has also been conducted on other stored product insects which are not yet cosmopolitan. For example, female rice moths, *Corcyra*

cephalonica (Stainton), could be sterilized with as little as 100 Gy (Huque, 1971), whereas doses of 70–100 Gy have been found to sterilize several species of bruchid seed weevils (Hallman, 1998a).

Weevils

Radiation studies have been done with several nonstored product weevils, with the most studied being the plum curculio, a pest of many fruits in temperate North America. Hallman (1998b) found no successful reproduction after >20 000 adult plum curculios were irradiated with a minimum absorbed dose of 80–92 Gy. The mango seed weevil, *Cryptorhynchus mangiferae* (F.) is a quarantine pest of mangoes in south Asia, Guam, New Caledonia, Hawaii, eastern Australia, southern Africa and the eastern Caribbean islands. When mangoes are harvested, all stages of the insect may be in the fruits. Heather & Corcoran (1992) obtained no weevil emergence from 'large numbers' of mangoes irradiated with 298–339 Gy. However, adults irradiated with 300 Gy may live up to 111 days (Seo *et al.*, 1974) and Heather & Corcoran (1992) believed that a dose of 300 Gy on mangoes would be marginal as far as mango quality. Doses to prevent reproduction of mango seed weevil may be as low as 50 Gy (Seo *et al.*, 1974). As with most other insects where adults are present, lack of reproduction should be the goal of an irradiation quarantine treatment against mango seed weevil.

Actively reproducing boll weevils require a sterilization dose of about 100 Gy (Davich & Lindquist, 1962). When irradiated in a hypoxic atmosphere, the dose must be higher; 100 Gy applied to boll weevils in a nitrogen atmosphere resulted in a mean of 0.13 adult progeny per female (Earle *et al.*, 1979). Sterility of male alfalfa weevils is achieved with about 50 Gy (Burgess & Bennett, 1966; Ferrer & Hower, 1975). Sterility of female pine weevils, *Pissodes strobi* (Peck), was achieved with 100 Gy except for one questionable female irradiated at 200 Gy (Jaynes & Godwin, 1957). Perhaps this is another case of an insect being misplaced.

The sweetpotato weevil, *Cylas formicarius elegantulus* (Summers), appears to be the most tolerant to radiation of the weevils studied. When irradiated males and females were mated with non-irradiated individuals of the opposite sex, males were sterilized with 300 Gy and females required 200 Gy (Dawes *et al.*, 1987). However, when both irradiated sexes were placed together (three replicates of 10 pairs each), few offspring were produced at 100 Gy and none at 150 Gy. Sharp (1995) found no reproduction when both sexes irradiated with 100 Gy were mated (nine replicates of 150 pairs each). An irradiation quarantine treatment need only examine the effect of radiation on both sexes together, and not one irradiated sex paired with the other non-irradiated sex, as it is assumed that all insects arriving with the shipped produce will have been irradiated. Accepting that criterion and taking both studies on sweetpotato weevil together, a dose of >100 Gy and ≤150 Gy might provide quarantine security against this pest. Sweetpotato is not very tolerant of radiation; although 200 Gy was not damaging, 400 Gy caused mottled darkening of cooked medullary tissue (McGuire & Sharp, 1995). This is one insect for which maintaining the dose as low as possible is critical to the treatment's success.

Only the egg stage of the Fuller rose beetle, *Pantomorus cervinus* (Boheman), occurs on citrus (under the calyx) as the insect does not develop on the fruit. Johnson *et al.* (1990) found that doses of 100 and 150 Gy prevented hatch of 99.9 and 100%, respectively, of 10–13-day-old eggs.

Other Coleoptera

Other than weevils and stored product insects, not much work has been done on irradiation of beetles. Mexican bean beetles, *Epilachna varivestis* Mulsant, were sterilized with 80 Gy (Henneberry *et al.*, 1964). Japanese beetle, *Popillia japonica* Newman, was sterilized with 150 Gy (Ladd *et al.*, 1973), whereas European chafer, *Rhizotrogus majalis* (Razoumowsky), required only 50 Gy (Chung *et al.*, 1971).

Lepidoptera

There is more information on the use of irradiation as a disinfestation treatment for codling moth, *Cydia pomonella* (L.), than any other lepidopteran. The most developed stage that is present in shipped fruits is the larva. All female fifth (last)-instar codling moths died before pupation when irradiated with 232 Gy (Proverbs & Newton, 1962). Twelve percent of the males pupated, but none emerged as adults. Although a dose of 93 Gy applied to fifth instars prevented only 19 and 31% of males and females, respectively, from emerging as adults, all of the resulting females and almost all of the males were sterile. When fifth-instar diapausing codling moth larvae were irradiated with 100 Gy (3 Gy/min), 4.5% developed to abnormal-looking adults (all males), and none were normal looking (Burditt & Moffitt, 1985). The mean estimated dose to prevent 99.9968% emergence of normal-looking adults from larvae in apples was 137 Gy; to prevent emergence of any adults the estimated dose was 207 Gy. Burditt (1986) found that diapausing codling moth larvae were more susceptible to irradiation than non-diapausing larvae. Doses of 156 and 230 Gy were estimated to prevent emergence of only apparently normal adults and all adults, respectively, from last-instar larvae in walnuts. No adults emerged from 4230 fifth instars in apples irradiated with 139–177 Gy (4 Gy/min) (Burditt & Hungate, 1989).

No fifth-instar *Spodoptera litura* (F.) cutworms formed normal-looking, viable pupae after irradiation with 100 Gy (Dohino *et al.*, 1996a). Inhibition of adult emergence from last-instar leafroller *Ctenopseustis obliquana* (Walker) could be achieved with about 150 Gy (Lester & Barrington, 1997). Inhibition of pupation might require twice that dose. At least 250 Gy would be necessary to prevent adult emergence from larvae of the litchi stem borer, *Conopomorpha sinensis* Bradley, in litchi fruits (Mei-ying *et al.*, 1998). Faria *et al.* (1998) irradiated small numbers of the orange fruit borer, *Ecdytolopha aurantianum* (Lima), in oranges and obtained one deformed adult of 19 total insects found at 300 Gy.

Several lepidopterous pests may be present in commodities as pupae. Stopping adult emergence from late pupae with irradiation requires impractically high doses. Therefore, the goal of a radiation quarantine treatment when pupae are present

should be adult sterility. No eggs hatched from ≥ 12 -day-old female gypsy moth, *Lymantria dispar* (L.) pupae irradiated with 100 Gy and mated with unirradiated males (Godwin *et al.*, 1964). A dose of 300 Gy was required to prevent hatch of eggs laid by Mexican rice borers. *Eoreuma loftini* (Dyar), irradiated as late pupae (Darmawi *et al.*, 1998).

Hemiptera

Heather (1992) cites a personal communication where a dose of 100 Gy was reported to sterilize the green peach aphid, *Myzus persicae* (Sulzer). The asparagus aphid, *Brachycorynella asparagi* (Mordvilko), did not reproduce when treated with 100 Gy, and produced all moribund offspring at doses as low as 10 Gy (rate of 17 Gy/min) (Halfhill, 1988). Female greenhouse whiteflies, *Trialeurodes vaporariorum* Westwood, were sterilized with 50 Gy, the lowest dose tried (Calvitti *et al.*, 1997), whereas 20 Gy sterilized cotton stainer, *Dysdercus koenigii* F., females (Srivastava & Deshpande, 1983). Female *Pseudococcus comstocki* (Kuwana) mealybugs irradiated with 200 Gy laid some viable eggs for a few days before becoming completely sterile 4 days post-irradiation (Dohino & Masaki, 1995).

Thysanoptera

Adult *Thrips tabaci* Lindeman seemed slightly more tolerant of radiosterilization than *T. palmi* Karny, and sterilization of both species would require a little over 200 Gy (Dohino *et al.*, 1996b). *Retithrips syriacus* (Mayet) adults (1–2-day-old) were sterilized with 150 Gy (Bhuiya *et al.*, 1999).

Acari

Spider mite adults require 200–320 Gy for sterilization (Hallman, 1998a). In some cases, females were more radiosterilization tolerant than males (Beavers *et al.*, 1971; Wakid *et al.*, 1972; Majumder *et al.*, 1996).

Doses to prevent reproduction of mites of the family Acaridae are around 300 Gy (Ignatowicz, 1997). Female grain mites, *Acarus siro* L., were more tolerant of radiosterility than males (Burkholder *et al.*, 1966), while for adult bulb mites, *Rhizoglyphus echinopus* (Fumouze & Robin), the contrary was found (Ignatowicz, 1992).

Nematoda

Nematodes require much higher doses than arthropods, and irradiation disinfestation of nematodes may be impractical except for highly tolerant commodities such as logs and raw lumber products. Over 4 kGy was required to prevent gall formation and subsequent egg production of second-stage juveniles of the root-knot nematode *Meloidogyne javanica* (Treub) (Chinnasri *et al.*, 1997). Doses up to 0.5 kGy did not affect growth or development of the stem nematode, *Ditylenchus dipsaci* (Kuhn) (Ignatowicz, 1998).

Quality of irradiated commodities

Besides being efficacious against the quarantined pest, a disinfestation treatment must not harm the quality of the

commodity to a point that it is unmarketable. Morris & Jessup (1994) review the types of damage caused by irradiation to some commodities and tolerance limits to radiation. More recent tolerances are presented in Table 3. Irradiation at the doses required for quarantine security is tolerated by many fresh commodities.

Irradiation will be applied to most commodities after they are packed and on pallets. The dose absorbed by commodities on the outside of the pallet could be 2–3 times the dose received in the middle. Therefore, commodities must tolerate 2–3 times the minimum dose required for quarantine security. Fortunately, all of the commodities in Table 3 (and many others) except sweet potato should show no significant detrimental effects from the dose of irradiation applied for quarantine security.

Detection of irradiated insects

Because radiation quarantine treatments do not kill insects outright, inspectors finding live insects have no independent confirmation that the product has been properly treated. Although the certification accompanying the load should be designed to guarantee that the irradiated produce has been adequately processed (validation is depended on for many other irradiated products, such as medical devices and sterile insects), a reliable, easy technique to identify irradiated insects would be desirable. A number of physical, chemical and biological methods have been investigated for determination of irradiated products (Haire *et al.*, 1997; Delincée, 1998). Some suffer from specificity, practicality and dose determination uncertainties; many are not effective at the relatively low radiation doses used for disinfestation

treatments. Chemical methods are generally not very promising because processes besides irradiation give the same results, and low-dose radiation quarantine treatments cause extremely minute chemical changes. Because DNA is such a large target for ionizing radiation, several detection approaches involve detecting DNA fragmentation, both nuclear and mitochondrial.

Some approaches rely on retardation of a specific developmental process, such as reduction of phenoloxidase production (Nation *et al.*, 1995) or size of the supraoesophageal ganlion (Rahman *et al.*, 1990) measured several days post-irradiation. Techniques that depend on developmental anomalies are of little practical benefit because they are not discernible soon after irradiation and may not detect insects that were irradiated when they were late instar. The insects best detected by these methods, early instars, would be killed by the higher doses needed to stop development of late instars or sterilize adults and, thus, not be available for detection anyway.

Electron spin resonance (ESR) spectroscopy, which detects free radicals formed radiolytically, might be suited to radiation dose determination of heavily sclerotized insect parts. ESR responses have proven quite variable, however, when applied to crustacean exoskeletons (Delincée, 1998). Ignatowicz & Banasik-Solgala (1999) could not distinguish irradiated from non-irradiated rice weevil or confused flour beetle when they were analysed dry and whole 2 weeks after irradiation.

Conclusions and recommendations

Irradiation promises to be a versatile disinfestation treatment which can be used against a wide variety of organisms and is

Table 3 Tolerance of various commodities to radiation from recent literature.

Quarantine pests	Plausible quarantine dose (Gy)	Commodities	Maximum tolerance (Gy)	Reference
Lepidopterous borers	300	Sugarcane	> 900	Hallman (unpublished data)
Codling moth	250	Apple, pear	900	Drake <i>et al.</i> (1998)
Plum curculio	80	Blueberry	750	Miller <i>et al.</i> (1994)
Codling moth	250	Apricot, cherry, peach	600	Drake & Neven (1998)
<i>Anastrepha</i> fruit flies	75	Grapefruit	≥ 300	Miller & McDonald (1996)
Mango seed weevil	~100	Mango	≥ 250	Hallman (1999)
Sweetpotato weevil	150	Sweetpotato	400	Hallman (unpublished data)

Pest group	Objective	Dose (Gy)
Aphids and whiteflies	Sterilize actively reproducing adult	50–100
Seed weevils (Bruchidae)	Sterilize actively reproducing adult	70–100
Scarab beetles	Sterilize actively reproducing adult	50–150
Fruit flies (Tephritidae)	Prevent adult emergence from 3rd instar	60–250
Weevils (Curculionidae)	Sterilize actively reproducing adult	80–200
Borers (Lepidoptera)	Prevent adult development from late larva	100–280
Thrips	Sterilize actively reproducing adult	150–250
Borers (Lepidoptera)	Sterilize late pupa	200–300
Mites	Sterilize actively reproducing adult	200–320
Stored product beetles	Sterilize actively reproducing adult	50–400
Stored product moths	Sterilize actively reproducing adult	100–1,000
Nematodes	Sterilize actively reproducing adult	~4,000

Table 4 Absorbed doses which might achieve quarantine security of several pest groups.

tolerated by many agricultural commodities. Possible dose ranges to control many quarantine pest groups are presented in Table 4.

Although the irradiation procedure itself is inherently dangerous, as are many quarantine treatments and food management processes, consumption of irradiated produce poses no risk to the consumer. A joint study by the World Health Organization, Food and Agricultural Organization, and International Atomic Energy Agency concluded in 1997 that there is no safety basis for setting a limit to the amount of irradiation that can be applied to food.

Information for suggesting possible doses is almost totally lacking for certain important pest groups, such as snails, slugs, gall mites, leaf miners and scale insects. Except for fruit flies and the plum curculio, quarantined pests have not been studied in sufficient numbers to validate efficacy.

The modifying affects of factors such as hypoxia and cold storage must be included in radiation disinfestation research as fresh commodities are frequently subjected to these conditions.

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