Ionizing Irradiation of Adults of Angoumois Grain Moth (Lepidoptera: Gelechiidae) and Indianmeal Moth (Lepidoptera: Pyralidae) to Prevent Reproduction, and Implications for a Generic Irradiation Treatment for Insects

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ABSTRACT Ionizing irradiation is used as a phytosanitary treatment against quarantine pests. A generic treatment of 400 Gy has been approved for commodities entering the United States against all insects except pupae and adults of Lepidoptera because some literature citations indicate that a few insects, namely, the Angoumois grain moth, *Sitotroga cerealella* (Olivier) (Lepidoptera: Gelechiidae), and the Indianmeal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), are not completely controlled at that dose. Radiotolerance in insects increases as the insects develop, so the minimum absorbed dose to prevent F_1 egg hatch for these two species when irradiated as adults was examined. Also, because hypoxia is known to increase radiotolerance in insects, Angoumois grain moth radiotolerance was tested in a hypoxic atmosphere. A dose range of 336–388 Gy prevented F_1 egg hatch from a total of 22,083 adult Indianmeal moths. Dose ranges of 443–505 and 590–674 Gy, respectively, prevented F_1 egg hatch from a total of 15,264 and 13,677 adult Angoumois grain moths irradiated in ambient and hypoxic atmospheres. A generic dose of 600 Gy for all insects in ambient atmospheres might be efficacious, although many fresh commodities may not tolerate it when applied on a commercial scale.

KEY WORDS *Sitotroga cerealella*, *Plodia interpunctella*, quarantine, commodity treatment, radiation

Ionizing irradiation has many uses in entomology, including sterilizing insects for population management and eradication programs, as a phytosanitary treatment, and for general disinfestation of commodities (Bakri et al. 2005). The International Consultative Group on Food Irradiation (ICGFI) suggested that 0.3 kGy be used as a generic, default phytosanitary dose of ionizing radiation for all insects and mites (ICGFI 1991). In 1994, this issue was taken up at a workshop, and a review of the literature showed that 0.3 kGy would probably be insufficient, as some species and life stages of insects and phytophagous mites are reported to reproduce to some extent after an exposure to that dose (Hallman 1994). Questions about methodology and radiation dosimetry in some of the research lead to a call for closer examination of the literature, research methodology, and doses required to achieve quarantine security for a variety of quarantine pests.

A generic, default dose of 0.4 kGy was recently approved as a phytosanitary treatment for all insects other than pupae and adults of Lepidoptera for commodities entering the United States (APHIS 2006). Pupae and adults of Lepidoptera were excluded from that recommendation because available information suggested that doses to prevent reproduction of some species when irradiated at these stages might be >0.4 kGy.

Data for the most radiotolerant insects set the minimum limit for a generic dose for the entire class. A review of the literature revealed that the Angoumois grain moth, *Sitotroga cerealella* (Olivier) (Lepidoptera: Gelechiidae), and the Indianmeal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), seemed to be the most radiotolerant arthropods known, with doses to prevent reproduction estimated at >1 kGy (Hallman 1998). A few female adult Indianmeal moths reportedly reproduced after irradiation with 1 kGy (Cogburn et al. 1966). The only reference to dosimetry reported by Cogburn et al. (1966) was "all dosages [except one noted to be ± 10%] were accurate within a very small tolerance." Brower (1975) found that reproduction in Indianmeal moth was stopped with 450 Gy but not 400 Gy when a total of 20 irradiated 1-d-old adult female were paired with nonirradiated males. Although absorbed doses were not given, doses were "confirmed" with a lithium fluoride dosimetry system.

A dose of 250 Gy did not completely prevent pairs of irradiated Angoumois grain moths from reproduc-

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ing (Qureshi et al. 1968). When Angoumois grain moth adult pairs were irradiated at 220 Gy, but not 200 Gy, hatching of eggs laid posttreatment was stopped (Ismail et al. 1987). Both studies did not report dosimetry.

When only the female Angoumois grain moth was irradiated at 240 Gy there was a 1.75% eclosion rate versus 94% for the nonirradiated control; no higher dose was tried by Ismail et al. (1987). Johnson and Vail (1987) found that ≈300 Gy prevented F_1 egg hatch when female Indianmeal moths were irradiated as pupae (day 9 after infestation); dosimetry is reported.

A brief report in a proceedings found that 1% of eggs laid by Indianmeal moth adults irradiated with a target absorbed dose of 400 Gy hatched (Hallman 2004). Subsequent observations questioned the validity of that preliminary conclusion, suggesting that untreated control insects were contaminating petri dishes holding irradiated eggs. After controls and irradiated insects were kept in separate rooms and petri dishes taped shut, no more first instars were found after 6,238 additional adults were irradiated with a target dose of 400 Gy (absorbed dose range was 395–448 Gy; G.J.H., unpublished data).

In the same proceedings, Zolfagharieh (2004) briefly reported that 350 Gy sterilized 200 adult Indianmeal moths. Sterilization was not defined, nor was dosimetry measurement or absorbed dose range given. Also in the same proceedings, Ignatowicz (2004) found that a target dose of 600 Gy applied to a mixture of an estimated 57,013 Angoumois grain moth of all stages, including adults prevented development of F_1 adults. At a target dose of 500 Gy applied to 1–2-d-old adults, 7.7% of F_1 eggs hatched. A target dose of 400 Gy applied to a mixture of an estimated 57,013 Angoumois grain moth of all stages, including adults, resulted in 0.77% reaching the F_1 adult stage. The dosimetry system used and dose range measurements were not given.

A phytosanitary treatment must be effective against the most tolerant life stage of quarantine pests that could normally be present in the shipped commodity, regardless of whether that stage is less commonly found than others. Insects increase in radiotolerance as they develop, when using a common measure of efficacy (Hallman 2000). Therefore, actively reproducing adults would require the highest dose to prevent successful reproduction.

Prevention of insect reproduction can be defined in various ways that could be subdivided by life stage and age within life stage as long as each subdivision could be defined. Development of phytosanitary treatments should include a precise description of the response that achieves efficacy. It is insufficient to state that a phytosanitary treatment prevents reproduction; prevention of reproduction should be defined as precisely as practical. Regulatory agencies must know when the threshold has been crossed and the treatment may be considered unsuccessful.

An insect can be quickly killed with radiation, although acute mortality requires doses so high (>3 kGy) that they are not tolerated by most fresh commodities. Doses to achieve acute mortality are much higher than needed to prevent the establishment of exotic invasive species (Hallman 1998). Prevention of the development of reproductive organs would be an early step to preventing reproduction when acute mortality is not the goal. This would only suffice for those species not present as actively reproducing adults; most quarantined insect species could be present as adults. Prevention of the production of gametes is a further step that could define treatment efficacy. That step could be further refined by restricting it to the prevention of egg production, which is usually accomplished with a lower dose than sperm production in insects (Hallman 1998). Further steps could be prevention of oviposition, then oviposition but no emergence of first instar, or prevention of successive F_1 life stages. Some studies base efficacy on prevention of the F_2 generation. For example, Follett (2006) based efficacy of an irradiation treatment of coconut scale, *Aspidiotus destructor* Signoret, on prevention of F_1 adults with F_2 eggs when parent generation adults with eggs and newly hatched crawlers were irradiated. Prevention of the F_2 generation might be especially relevant to Lepidoptera, which may suffer greater sterility in subsequent generations than in the one irradiated (Bloem et al. 2003, Wee et al. 2005).

When the same life stage is irradiated, prevention of each successive stage is usually accomplished by a lower irradiation dose than needed to prevent the previous life stage because more total developmental change must be achieved. However, each successive step in lowering the dose by accepting more development is probably accompanied by increased risk of treatment failure, because as development progresses the number of developmental stages until reproduction is successful logically decreases.

A major disadvantage with phytosanitary irradiation treatments compared with other quarantine treatments is that because irradiation is the only commercially applied treatment that does not result in significant acute mortality, phytosanitary inspectors have no independent verification of treatment efficacy. Because of this, any deficiency in the research or application of irradiation may lead to a greater risk of establishment of invasive species compared with other treatments for which discovery of live insects posttreatment denotes treatment failure. Research and application of phytosanitary irradiation treatments should be held to higher standards than other treatments. It may be prudent, therefore, to leave a few developmental steps between the phytosanitary treatment effect and successful reproduction so that unexpected variations in efficacy will have a lower risk of leading to successful reproduction and establishment of invasive species.

The objective of this research was to determine minimum absorbed doses required to prevent F_1 egg hatch when the two moths are irradiated. Because the Angoumois grain moth seems to be more radiotolerant than the Indianmeal moth, egg hatch of the former was tested under hypoxic conditions also. Hypoxia is used to prolong shelf life of stored commodities and may