Development of a generic radiation dose for the postharvest phytosanitary treatment of mealybug species (Hemiptera: Pseudococcidae)

Hendrik Hofmeyr[1], Thi The Doan[2], Murni Indarwatmi[3], Ranjana Seth[4], and Guoping Zhan[5]

Abstract

Studies were conducted to develop a generic dose of ionizing radiation for the postharvest phytosanitary control of mealybugs (Hemiptera: Pseudococcidae). Details of studies done with 11 species are fully discussed in individual papers elsewhere in this special issue; also additional information from the literature is examined. It was established that tolerance to ionizing radiation increased progressively, but not without exception, as the insects developed through various successive stages from eggs to ovipositing females. The various species were found to require doses ranging from 100 to 200 Gy to either prevent oviposition when either pre-ovipositional or ovipositing females were irradiated or to prevent the production of viable 1st instars in the F_1 generation. Large scale confirmatory tests were conducted with 4 species (3 at the Probit-8.7 level of efficacy) to validate doses of 150 and 200 Gy. In all studies on phytosanitary irradiation of mealybug species a dose of 200 Gy or less has proven to be sufficient to kill the pre-imaginal stages and either to sterilize the imaginal females or to render the F_1 generation 1st instars non-viable. Therefore a generic dose of 250 Gy can be recommended as an effective treatment for the postharvest phytosanitary control of the mealybug species complex.

Key Words: ionizing radiation; generic dose; phytosanitary irradiation; phytosanitation, radiotolerance of developmental stages

Resumen

Se realizaron estudios para desarrollar una dosis genérica de radiación ionizante para el control fitosanitario poscosecha de cochinillas (Hemiptera: Pseudococcidae). Se discuten en detalle los estudios realizados con 11 especies en los documentos individuales en otras partes de este número especial; también se examinó la información adicional de la literatura. Se estableció que la tolerancia a la radiación ionizante aumenta progresivamente, pero no sin excepción, ya que los insectos se desarrollan a través de los diversos estadios sucesivos desde los huevos hasta la oviposición de las hembras. Se encontró que las diversas especies requieren una dosis de 100 a 200 Gy para prevenir la oviposición cuando se irradiaron hembras antes o durante de la pre-oviposición para prevenir la producción de la 1ª estadios viables en la generación F_1. Se realizaron pruebas confirmatorias en gran escala con 4 especies (3 a nivel Probit-8.7 de eficacia) para validar las dosis de 150 y 200 Gy. En todos los estudios sobre la irradiación fitosanitaria de las especies de cochinillas una dosis de 200 Gy o menos ha demostrado ser suficiente para matar los estadios pre-imaginales y para esterilizar las hembras imaginales o hacer que los instares de la primera generación F_1 instares no sea viable. Por lo tanto, una dosis genérica de 250 Gy puede ser recomendada como un tratamiento efectivo para el control fitosanitario poscosecha del complejo de especies de cochinillas.

Palabras Clave: radiación ionizante; dosis genérica; irradiación fitosanitaria; fitosanidad; radiotolerancia de estadios desarrollandose

The mealybug group of insects (Hemiptera: Pseudococcidae) is widely distributed (Wakgari & Giliomee 2003). It is a general pest of ornamental plants and is often of economic concern in agricultural produce traded internationally including avocado (*Persea americana* Mill.; Laurales: Lauraceae), citrus (*Citrus* spp.; Sapindales: Rutaceae), cucurbits (*Cucurbita* spp., *Lagenaria* spp., *Citrullus* spp., *Cucumis* spp., and *Luffa*

[1]Citrus Research International, PO Box 212, Citrusdal 7340, South Africa; jhh9345@gmail.com
[2]Research and Development Center for Radiation Technology, Vietnam Atomic Energy Institute (Vinatom) 202A, Street 11, Linh Xuan Ward, Thu Duc District, Ho Chi Minh City, Vietnam; doanthithe@yahoo.com
[3]Centre for the Application of Isotopes and Radiation Technology, Center for Isotopes and Radiation Application (CIRA), National Nuclear Energy Agency (BATAN), Jl. Lebak Bulus Raya No. 49, Pasar Jumat, Jakarta 12440, Indonesia; murninda@gmail.com
[4]Zoology Department – Desh Bandhu College, University of Delhi – Kalkaji, New Delhi 110019, India; seth.ranjana.27@gmail.com
[5]Institute of Equipment Technology, Chinese Academy of Inspection and Quarantine (CAIQ), Bld. No. 241, Huixinli, Huixin Xijie, Chaoyang District, Beijing 100029, China; zhgp136@126.com
*Corresponding author; E-mail: jhh9345@gmail.com

spp.; Cucurbitales: Cucurbitaceae), dates (*Phoenix dactylifera* L.; Arecales: Arecaceae), numerous deciduous fruits, mangosteen (*Garcinia mangostana* L.; Malpighiales: Clusiaceae), rambutan (*Nephelium lappaceum* L.; Sapindales: Sapindaceae), pitaya (*Stenocereus* spp.; Caryophyllales: Cactaceae) and dragon fruit (*Hylocerus* spp.; Caryophyllales: Cactaceae).

Small numbers of mealybugs are often cryptic pests in the field as they tend to congregate, multiply, and survive best in secluded spots on host plants such as cracks and crevices on the trunk, leaf axils, and fruit calyxes (Gimpel & Miller 1996). In the absence of natural or artificial controls they can develop large, easily noticeable populations feeding on vegetative and reproductive tissues. The damage caused by mealybugs is diverse and comprises direct and indirect effects that are common to many species. Foliar damage is apparent in stunted and bunched growth, leaf malformation and in more severe cases, leaf drop. Honeydew secreted by these insects causes sooty mold growth that can inhibit photosynthesis and promote the incidence of fungal decay such as *Alternaria* (Hattingh et al. 1995). Feeding activities can also result in the drop of fruitlets and malformation of remaining fruit. Infestations under the calyces of fruits, in bunches of berries, or on fruits can be problematic in exportable produce (Williams 2004; Jiao et al. 2011) resulting in rejections from export (Jahn 1993; Hattingh et al. 1998). At best, delays can occur as the insects are being identified to avoid confusion with mealybug species that may be of phytosanitary concern (Wakgari & Giliomee 2003).

Phytosanitary irradiation has been approved and used to disinfest pests on agricultural commodities to overcome quarantine barriers for safe trade between countries or areas (Hallman 2011). Hallman (2012) found that after tephritid fruit flies, mealybugs comprised the next most important group of quarantine pests for which phytosanitary treatments were required, and Hallman (2011) suggested that a phytosanitary dose of 250 Gy for the group might be feasible. The objective of this project was to establish a generic radiation dose for mealybugs suitable for commercial application in cases where dose rates for particular species were unspecified.

Some of the research on individual species mentioned in this paper is fully reported elsewhere in this issue of Florida Entomologist. For a detailed discussion on a particular species (inclusive of correspondence) please consult the authors as follows:

Hendrik Hofmeyr: *Planococcus citri*, *Pl. ficus*; E-mail: jhh9345@gmail.com

Thi The Doan: *Dysminococcus neobrevipes*, *Pl. lilacinus*, *Pl. minor*; E-mail: doanthithe@yahoo.com

Murni Indarwatmi: *Exallomochlus hispidus*, *Pseudococcus cryptus*; E-mail: murninda@gmail.com

Ranjana Seth: *Phenacoccus solenopsis*, *Maconellicoccus hirsutus*, *Paracoccus marginatus*; E-mail: seth.ranjana.27@gmail.com

Zhan Guoping: *Ps. jackbeardsleyi*; E-mail: zhgp136@126.com

Materials and Methods

Different development stages of 11 species of mealybugs were individually assessed to determine the stage most tolerant to ionizing radiation. The stages assayed were eggs (4 species), 1st to 3rd instar (10 species), as well as females in pre-ovipositional and ovipositional phases (10 and 6 species, respectively).

REARING SUBSTRATE

In all cases the relatively short shelf life (as short as 4–5 d) of the host produce—e.g., dragon fruit and mangosteen—precludes their use for experimental purposes. The various mealybug species were therefore reared, treated, and evaluated on either butternut squash (*Cucurbita moschata* Duchesne ex Poir.; Cucurbitales: Cucurbitaceae), Kabocha squash (*C. maxima* Duchesne) or potato (*Solanum tuberosum* L.; Solanales: Solanaceae). These hosts extended the exploitable shelf life to more than 45 d, enabling the production of a complete generation without the need to transfer insects to fresh stock. Depending on the species, rearing temperatures and relative air humidity, respectively, varied from 24–29 °C and 60–75%.

TEST INSECTS

Relative Radiotolerances of Different Stages

Eggs. Eggs were prepared for study by transferring 10–15 ovisacs from the rearing stock to each of 3–5 replicates of fresh pumpkins or potatoes (hereafter ‘host substrate’). On average each ovisac contained 50–350 eggs and totals of 500–17,500 eggs were used per species.

Pre-imaginal and imaginal stages. For 9 species pre-ovipositional females were removed from the rearing stock by hand and transferred to fresh host substrate for reproduction or treatment. Each replicate, consisting of a single pumpkin or potato, was infested with 30–100 individuals. For all species 3–5 replicates were used per instar. For 2 species 1st instars were allowed to migrate naturally to fresh butternut squash fruits placed on top of the rearing substrate, resulting in 350–3,700 test insects per replicate per instar. When the progeny had developed to the required stages they were treated on the host substrate with 40–400 Gy at 25–100 Gy increments. The treated mealybugs were incubated and monitored to determine their ability to produce a viable F_1 generation.

Mass Assessments

Females in pre-ovipositional or ovipositional phases of *D. neobrevipes*, *Pl. citri*, and *Ps. jackbeardsleyi* were used to confirm the lowest dose to prevent the development of viable F_1 progeny. Approximately 30,000–118,000 insects were used per species, conforming to or exceeding, a Probit-8.7 level of efficacy (no survivors in ~30,000 insects treated). A fourth species, *Pl. ficus*, was evaluated using 10,000 individuals. Host substrate with large numbers of mealybugs was prepared for treatment by naturally infesting fresh pumpkins or potatoes with 1st instars from infested rearing stock.

DOSIMETRY

Three gamma cell/chamber type irradiators, an electron beam accelerator, a Cobalt-60 source located 5 m underground and raised to floor level for operational purposes, as well as a panoramic point source irradiator with primary and secondary turntables for treatment targets, were employed. Dosimetry studies were conducted to calibrate the equipment and to record dose uniformities and uncertainties. Specific information on the radiation equipment and dosimetry used in the 7 contributing studies on phytosanitary irradiation of various mealybug species is provided in this issue of Florida Entomologist (Doan et al. 2016; Hofmeyr et al. 2016a; Kuswadi et al. 2016; Seth et al. 2016a, 2016b; 2016c; Zahn et al. 2016).

Results

Results for all 11 species of mealybug studied are summarized in Table 1. Due to space constraints the data for ionizing radiation doses

Table 1. The effects of a range of doses of ionizing radiation on pre—imaginal and imaginal stages of 11 species of mealybug. The progressive increase in treatment dose between imagoes of the assessed species is demonstrated by shaded cells.

Species	Treated stage (P generation)	Dose (Gy) 0	100	125	150	200	300—400
Pseudococcus cryptus	Pre-ovipositional ♀	Fertile ♀♀	Sterile ♀♀	—	—	Sterile ♀♀	
Exallomochlus hispidus	1st instar	Fertile ♀♀	Sterile ♀♀	—	—	Sterile ♀♀	—
	2nd instar	Fertile ♀♀	Sterile ♀♀	—	—	Sterile ♀♀	—
	3rd instar	Fertile ♀♀	Sterile ♀♀	—	—	Sterile ♀♀	—
	Pre-ovipositional ♀	Fertile ♀♀	Fertile ♀♀	Sterile ♀♀	—	Sterile ♀♀	—
Pseudococcus jackbeardsleyi	1st instar	Fertile ♀♀	Sterile ♀♀	Treated 1st instars killed	—	—	—
	2nd instar	Fertile ♀♀	Sterile ♀♀		—	—	—
	3rd instar	Fertile ♀♀	Sterile ♀♀		—	—	—
	Pre-ovipositional ♀	Fertile ♀♀	Sterile ♀♀			—	—
	Ovipositing ♀	Fertile ♀♀	Fertile ♀♀	F_1 1st instars die			Sterile ♀♀ (>200 Gy)
Planococcus citri	Egg	Fertile ♀♀	1st instars die	—	—	—	—
	1st instar	Fertile ♀♀	2nd instars die	—	Treated 1st instars killed		
	2nd-3rd instar	Fertile ♀♀	Sterile ♀♀	—	Sterile ♀♀	Treated 2nd—3rd instars killed	
	Pre-ovipositional ♀	Fertile ♀♀	Fertile ♀♀	—	Sterile ♀♀		
	Ovipositing ♀	Fertile ♀♀	—	—	Sterile ♀♀	—	—
Planococcus ficus	Ovipositing ♀	Fertile ♀♀	—	—	Sterile ♀♀	—	—
Planococcus lilacinus	1st instar	Fertile ♀♀	Treated 1st instars killed	—	Treated 1st instars killed		
	2nd instar	Fertile ♀♀	Treated 2nd instars killed	—	Treated 2nd instars killed		
	3rd instar	Fertile ♀♀	Sterile ♀♀	—	Sterile ♀♀		Treated 3rd instars killed
	Pre-ovipositional ♀	Fertile ♀♀	Sterile ♀♀	—	Sterile ♀♀		
Planococcus minor	Egg	Fertile ♀♀	Fertile ♀♀	—	Sterile ♀♀		
	1st instar	Fertile ♀♀	Treated 1st instars killed	—	Treated 1st instars killed		
	2nd instar	Fertile ♀♀	Treated 2nd instars killed	—	Treated 2nd instars killed		
	3rd instar	Fertile ♀♀	Fertile ♀♀	—	Sterile ♀♀		Treated 3rd instars killed
	Pre-ovipositional ♀	Fertile ♀♀	Fertile ♀♀	—	Sterile ♀♀		
Paracoccus marginatus	Egg	Fertile ♀♀	Treated eggs die	—	—	—	—
	1st instar	Fertile ♀♀	3rd instars die	—	—	Treated 1st instars killed	—
	2nd instar	Fertile ♀♀	Sterile ♀♀	—	—	Treated 2nd instars killed	—
	3rd instar	Fertile ♀♀	Sterile ♀♀	—	—	Sterile ♀♀	Treated 3rd instars killed
	3-4 d pre-ovipositional ♀	Fertile ♀♀	F_1 1st instars die	—	—	Sterile ♀♀	—
	6-7 d pre-ovipositional ♀	Fertile ♀♀	F_1 1st instars die	—	—	Sterile ♀♀ (165 Gy)	—
Dysminococcus neobrevipes	1st instar	Fertile ♀♀	Fertile ♀♀	—	Fertile ♀♀	Sterile ♀♀	Treated 1st instars killed
	2nd instar	Fertile ♀♀	Fertile ♀♀	—	Fertile ♀♀	Sterile ♀♀	
	3rd instar	Fertile ♀♀	Fertile ♀♀	—	Fertile ♀♀	Sterile ♀♀	
	Pre-ovipositional ♀	Fertile ♀♀	Fertile ♀♀	—	Fertile ♀♀	Sterile ♀♀	
Maconellicoccus hirsutus	Egg	Fertile ♀♀	Treated eggs die	—	—	—	—
	1st instar	Fertile ♀♀	Sterile ♀♀	—	—	3rd instars die	Treated 1st instars killed
	2nd instar	Fertile ♀♀	Sterile ♀♀	—	—	3rd instars die	Treated 2nd instars killed
	3rd instar	Fertile ♀♀	Sterile ♀♀	—	—	Sterile ♀♀	Sterile ♀♀
	3-4 d pre-ovipositional ♀	Fertile ♀♀	Fertile ♀♀	—	—	F_1 1st instars die	—
	6-7 d pre-ovipositional ♀	Fertile ♀♀	Fertile ♀♀	—	—	F_1 1st instars die	Sterile ♀♀ (235 Gy)

Table 1. (Continued) The effects of a range of doses of ionizing radiation on pre—imaginal and imaginal stages of 11 species of mealybug. The progressive increase in treatment dose between imagoes of the assessed species is demonstrated by shaded cells.

Species	Treated stage (P generation)	Dose (Gy) 0	100	125	150	200	300—400
Phenacoccus solenopsis	1st instar	Fertile ♀♀	3rd instars die	—	—	2nd instars die	Treated 1st instars killed
	2nd instar	Fertile ♀♀	3rd instars die	—	—	3rd instars die	Treated 2nd instars killed
	3rd instar	Fertile ♀♀	Sterile ♀♀	—	—		Sterile ♀♀
	5-6 d pre-ovipositional ♀	Fertile ♀♀	Fertile ♀♀	—	—		Sterile ♀♀
	11-12 d pre-ovipositional ♀	Fertile ♀♀	Fertile ♀♀	—	—	F_1 1st instars die	Sterile ♀♀ (374 Gy)

less than 100 Gy have been omitted. For more details readers are referred to individual authors' papers.

In all 11 species treated stages were killed outright, died in a subsequent stage, or developed into either fertile or sterile females. In this paper "sterile" is defined as the absence of egg production or the production of infertile eggs by the female. In 10 species the effective dose was considered to be the lowest dose to prevent egg production by irradiated females. In the 11th species female sterility as such was not confirmed, but the effective dose caused mortality of all F_1 1st instars.

RELATIVE RADIOTOLERANCE OF DIFFERENT STAGES

It is generally accepted that radiotolerance increases commensurately with development of the insect, not only within successive stages, but also within a particular stage (Dohino & Masaki 1995; Christopher et al. 2003; Ravuiwasa et al. 2009; Hallman et al. 2010; Doan et al. 2012; Shao et al. 2013; Hofmeyr et al. 2016b). However, in a number of cases an inverse relationship in the treatment tolerance of different stages of mealybug was recorded:

The eggs of 4 species were assessed. In 2 of these species the eggs were killed at doses lower than those killing 1st and 2nd instars (*Pa. marginatus* and *M. hirsutus*) (Table 1). Treated eggs of the 3rd species (*Pl. citri*) hatched into 1st instars that died, and the treated 1st and 2nd instars were killed at a higher dose. These 3 species displayed the expected effect of a less developed stage that is less radiotolerant than a more developed successive stage. Eggs of the 4th species (*Pl. minor*) developed into sterile females, while the 1st and 2nd instars were killed outright at a lower dose, showing an inverse relationship between lethal doses and succeeding life stages.

Mortality of treated 1st and 2nd instars was compared between 6 species; in 5 of these species both instars died when treated with a similar dose (*Pl. lilacinus*. *Pl. minor*, *Pa. marginatus*, *M. hirsutus,* and *Ph. solenopsis*) (Table 1). In the 6th species (*Pl. citri*) the 2nd instar was more tolerant than the 1st instar.

In 6 of 9 species the 3rd instar was sterilized at the same dose as females in the pre-ovipositional phase (*Ps. jackbeardsleyi, Pl. lilacinus, Pl. minor, Pa. marginatus, D. neobrevipes* and *E. hispidus)* (Table 1). In the remaining 3 species (*Pl. citri, M. hirsutus,* and *Ph. solenopsis*) sterility of the females in the pre-ovipositional phase was achieved at higher doses than those necessary for 3rd instar.

Females in pre-ovipositional and ovipositional phases were compared in 2 species, *Pl. citri* and *Ps. jackbeardsleyi*. In *Pl. citri* both categories of females were equally susceptible to sterilization following treatment at the same dose (Table 1). In *Ps. jackbeardsleyi* a higher dose was necessary to prevent the continued development of F_1 1st instars produced by treated pre-ovipositional than ovipositing females.

Although the relative radiotolerance of different stages for acute mortality varied, the general trend was for mealybug susceptibility to ionizing radiation to decrease as they developed in reproductive capability from eggs to females in the ovipositional phase. Hallman et al. (2010) pointed out that what matters is that the trend of radiotolerance increasing with development is for measurements of efficacy that are used in phytosanitary treatments. Although acceptable as a measure of efficacy, acute mortality is so far not used. Hallman et al. (2010) further discussed apparent exceptions to this trend that may be due to differences in treatment techniques and evaluations used for different stages.

MOST RADIOTOLERANT STAGE FOR TREATMENT VALIDATION

Females in pre-ovipositional and ovipositional phases in the 11 assessed species were more tolerant than the earlier stages. There were substantial differences in radiotolerance between the species, with doses varying from 100 Gy to a maximum of nearly 400 Gy to sterilize the females. Consequently, the highest doses had to be selected for the various species that would either completely sterilize the females or result in the production of non-viable F_1 1st instars (Table 2). These doses were used in large-scale validation experiments with 4 species to validate a single generic dose for mealybug.

MASS ASSESSMENTS FOR TREATMENT VALIDATION

Ovipositing females of *Pl*. *citri* and *Pl*. *ficus* were sterilized at a dose of 150 Gy. Ovipositing females of *Ps*. *jackbeardsleyi* were not sterilized at 200 Gy, but produced fertile eggs that eclosed into non-viable F_1 1st instars. All pre-ovipositional females of *D*. *neobrevipes* were sterilized at a dose of 200 Gy.

Discussion

It was established for certain species of mealybug that sterilization of the F_1 generation could be achieved by treating P females with doses as low as approximately 40 Gy. However, although such low doses are acceptable from a practical point of view, it would necessitate rearing treated mealybugs to F_1 ovipositional adults to confirm sterility. This process could take 4–6 wk; thus precluding any chance that treated agricultural produce would be approved for distribution by inspection authorities. Furthermore, such an operationally unverifiable measure of efficacy may not be acceptable to some plant protection organizations (Hallman et al. 2010).

The acute knock-down effect of ionizing radiation at doses of up to 400 Gy (the maximum dose tested in the various studies) was variable between the 11 species and could not be relied on to kill all stages outright or in a successive stage.

There were substantial differences in radiotolerance between the 11 species of mealybug studied in this project. Nonetheless, in all these studies a dose of 200 Gy killed the pre-imaginal stages, sterilized the imaginal females or rendered the F_1 generation 1st instars non-viable. Comparable results with additional species have been reported in the literature:

Planococcus minor was studied by Ravuiwasa et al. (2009), who reported no fertile eggs from adult females treated with 150 Gy. In a current study on *Pl*. *minor* pre-ovipositional females were sterilized with 150 Gy (Table 1).

The treatment of 'mature, adult female' *Ps. comstocki* with 200 Gy resulted in 2.5% egg hatch in the F_1 generation for only those eggs laid within 4 d after irradiation. No eggs laid subsequently hatched (Dohino & Masaki 1995). The few F_1 adults that developed did not lay any eggs. The next highest dose studied (400 Gy) resulted in no egg hatch in the F_1 generation. A mixed population of 3rd instar *Ps*. *comstocki*, including 77.5% pre-ovipositional adult females, was sterilized with the same dose (Dohino et al. 1997).

In an additional study on *Ph*. *solenopsis*, the production of F_1 adults was prevented when pre-ovipositional females were treated with either 150 Gy or 200 Gy (Huang et al. 2014). No information on the effect of these doses on earlier F_1 stages was presented.

Jacobsen & Hara (2003) reported that adult *M*. *hirsutus* females treated with 100 Gy produced small numbers of fertile eggs, and were completely sterile at 250 Gy. The results from a current study on *M*. *hirsutus* indicate approximately comparable effects in a similar dose range, i.e., imaginal females treated with 200 Gy and 235 Gy yielded, respectively, non-viable F_1 instars and sterile parental females (Table 1).

With the exception of *Ps. comstocki*, individuals of mealybug species reported on in this paper were either killed, sterilized, or produced non-viable F_1 instars at 200 Gy. We are unaware of published evidence of a mealybug species that remained fertile or produced viable F_1 progeny at 250 Gy. If all this is taken into account, we conclude that a 250 Gy dose could be an appropriate generic dose for the post-harvest control of mealybug species, and that would prevent the accidental or unintentional importation of any invasive mealybug species.

Acknowledgments

This work was part of the FAO/IAEA Coordinated Research Project D62008 on Development of Generic Irradiation Doses for Quarantine Treatments. Achmad Nasroh Kuswadi (Center for Application of Isotope and Radiation Technology, Center for Isotopes and Radiation Application (CIRA), National Nuclear Energy Agency (BATAN). Jakarta, Indonesia) conducted most of the research on *E. hispidus* until he retired in 2012. His work is gratefully acknowledged. The significant research and dosimetry contributions rendered by Rakesh Kumar Seth (Applied Entomology and Radiation Biology Laboratory, University of Delhi), and Kobus Slabbert (Radiation Biophysics, iThemba LABS, Somerset-West, South Africa) are particularly appreciated.

We are thankful for the dosimetry advice and surveys conducted by Andrew Parker (NAFA-IPC, Seibersdorf, Austria), the research assistance provided by Guy Hallman (NAFA-IPC, Seibersdorf, Austria) and the input of Peter Leach (Tropical Phytosanitary Solutions, Cairns, Queensland, Australia) during RCM sessions. We also thank Carl Blackburn and Yves Hénon (IAEA, Vienna, Austria) for their organizational support and general assistance. Ray Cannon (Food and Environment Research Agency, York, United Kingdom) was responsible for excellent reportage at Research Coordination Meetings throughout the course of this project.

Table 2. Minimum treatment dose of ionizing radiation of each of eleven mealybug species resulting either in sterile females or non-viable F_1 generation 1st instars.

Species	Development stage of treated females	Treatment dose (Gy)	Treatment effect
Pseudococcus cryptus	pre-ovipositional	100 Gy	Females sterilized
Planococcus citri	ovipositing	150 Gy	Females sterilized
Exallomochlus hispidus	pre-ovipositional	125 Gy	Females sterilized
Pseudococcus jackbeardsleyi	ovipositing	125 Gy	Non-viable F1 1st instars
Planococcus ficus	ovipositing	150 Gy	Females sterilized
Planococcus lilacinus	pre-ovipositional	150 Gy	Females sterilized
Planococcus minor	pre-ovipositional	150 Gy	Females sterilized
Paracoccus marginatus	pre-ovipositional	165 Gy	Females sterilized
Dysminococcus neobrevipes	pre-ovipositional	200 Gy	Females sterilized
Maconellicoccus hirsutus	pre-ovipositional	200 Gy	Non-viable F1 1st instars
Phenacoccus solenopsis	pre-ovipositional	200 Gy	Non-viable F1 1st instars

References Cited

Doan TT, Nguyen TK, Vo TKL, Cao VC, Tran TTA, Nguyen HHT. 2012. Effects of gamma irradiation on different stages of mealybug *Dysmicoccus neobrevipes* (Hemiptera: Pseudococcidae). Radiation Physics and Chemistry 81: 97-100.

Doan TT, Nguyen TK, Vo TKL, Nguyen TL, Cao VC, Tran TTA, Nguyen HHT. 2016. Phytosanitary irradiation against the mealybugs, *Dysmicoccus neobrevipes*, *Planococcus lilacinus* and *Planococcus minor* (Hemiptera: Pseudococcidae) infesting dragon fruit (Caryophyllales: Cactaceae) in Vietnam. Florida Entomologist 99(special issue 2): 159-165.

Dohino T, Masaki S. 1995. Effects of electron beam irradiation on Comstock mealybug, *Pseudococcus comstocki* (Kuwana) (Homoptera: Pseudococcidae). Research Bulletin of the Plant Protection Service Japan 31: 31-36.

Dohino T, Masaki S, Takano T, Hayashi T. 1997. Effects of electron beam irradiation on sterility of Comstock mealybug, *Pseudococcus comstocki* (Kuwana) (Homoptera: Pseudococcidae). Research Bulletin of the Plant Protection Service Japan 33: 31-34.

Gimpel WF, Miller DR. 1996. Systematic analysis of the mealybugs in the *Pseudococcus maritimus* complex (Homoptera: Pseudococcidae). Contributions on Entomology, International 2: 1-163.

Hallman GJ, Levang-Brilz NM , Zettler JL, Winborne IC. 2010. Factors affecting ionizing radiation phytosanitary treatments, and implications for research and generic treatments. Journal of Economic Entomology 103: 1950-1963.

Hallman GJ. 2011. Phytosanitary applications of irradiation. Comprehensive Reviews in Food Science and Food Safety 10: 143-151.

Hallman GJ. 2012. Generic phytosanitary irradiation treatments. Radiation Physics and Chemistry 81: 861-866.

Hattingh VH, Tate B, Richards G. 1995. The effect of mealybug infestation on the incidence of post-harvest *Alternaria* decay in navel oranges. Citrus Journal 5: 18-19.

Hattingh VH, Cilliers JC, Bedford ECG. 1998. Citrus mealybugs, pp. 112-120 *In* Bedford ECG, van Den Berg MA, De Villiers EA [eds.] Citrus Pests in the Republic of South Africa. 2nd Edition, Dynamic AD, Nelspruit, South Africa.

Hofmeyr H, Hofmeyr M, Slabbert K. 2016a. Postharvest phytosanitary irradiation disinfestation of *Planococcus citri* and *P. ficus* (Hemiptera: Pseudococcidae). Florida Entomologist 99(special issue 2): 166-170.

Hofmeyr H, Hofmeyr M, Slabbert JP. 2016b. Post-harvest phytosanitary disinfestation of false codling moth, *Thaumatotibia leucotreta* (Meyrick) (Lepidoptera: Tortricidae) in export citrus fruit from South Africa: Tolerance of eggs and larvae to ionizing radiation. Florida Entomologist 99(special issue 2): 48-53..

Huang F, Li WD, Li XQ, Bei YW, Lin WC, Lu YB, Wang BK. 2014. Irradiation as a quarantine treatment for the solenopsis mealybug, *Phenacoccus solenopsis*. Radiation Physics and Chemistry 96: 101-106.

Jacobsen CM, Hara AH. 2003. Irradiation of *Maconellicoccus hirsutus* (Homoptera: Pseudococcidae) for phytosanitation of agricultural commodities. Journal of Economic Entomology 96: 1334-1339.

Jahn GC. 1993. Gray pineapple mealybugs, *Dysmicoccus neobrevipes* Beardsley (Homoptera: Pseudococcidea), inside closed pineapple blossom cups. Proceedings of the Hawaiian Entomological Society 32: 147-148.

Jiao Y, YU DJ, Xu L, CHEN ZL, Lou DF, Kang L. 2011. The interception of Jack Beardsley mealybug on wax-apple imported from Thailand. Plant Quarantine 25: 63-65.

Kondo T, Ramos-Portilla AA, Vergara-Navarro EV. 2008. Updated list of mealybugs and putoids from Colombia (Hemiptera: Pseudococcidae and Putoidae). Bulletin of the Museum of Entomology at the University of Valle 9: 29-53.

Kuswadi AN, Indarwatmi M, Nasution IA, Sasmita HI. 2016. Minimum gamma irradiation dose for phytosanitary treatment of the cacao mealybug, *Exallomochlus hispidus* (Hemiptera: Pseudococcidae). Florida Entomologist 99(special issue 2): 69-75.

Ravuiwasa KT, Lu KH, Shen TC, Hwang SY. 2009. Effects of irradiation on *Planococcus minor* (Hemiptera: Pseudococcidae). Journal of Economic Entomology 102: 1774-80.

Seth RK, Zarin M, Khan Z, Seth R. 2016a. Efficacy of ionizing radiation as phytosanitary treatment against the various ontogenic stages of the Solenopsis mealybug, *Phenacoccus solenopsis* (Hemiptera: Pseudococcidae). Florida Entomologist 99(special issue 2): 76-87.

Seth R, Zarin M, Khan Z, Seth RK. 2016b. Effects of gamma radiation on metamorphic disruption and sterility in the pink hibiscus mealybug, *Maconellicoccus hirsutus* (Hemiptera: Pseudococcidae), to establish phytosanitary irradiation against infested agro-commodities. Florida Entomologist 99(special issue 2): 107-113.

Seth R, Zarin M, Khan Z, Seth RK. 2016c. Towards phytosanitary irradiation of *Paracoccus marginatus* (Hemiptera: Pseudococcidae): Ascertaining the radiosensitivities of all life stages. Florida Entomologist 99(special issue 2): 88-101.

Shao Y, Ren LL, Liu YJ, Wang YJ, Jiao Y, Wang QL, Zhan GP. 2013. The primary results of the impact on the development and reproduction of Jack Beardsley mealybug irradiated with Cobalt-60 gamma rays. Plant Quarantine 27(6): 51-55.

Wakgari WM, Giliomee JH. 2003. The biology of three mealybug species (Hemiptera: Pseudococcidae) found on citrus in the Western Cape Province, South Africa. African Entomology 11: 173-182.

Williams DJ. 2004. Mealybugs of southern Asia. Southdene Sdn Bhd, Kuala Lumpur, Malaysia.

Zhan GP, Shao Y, Yu Q, Xu L, Liu B, Wang YJ, Wang QL, 2016. Phytosanitary irradiation of Jack Beardsley mealybug (Hemiptera: Pseudococcidae) females on rambutan (Sapindales: Sapindaceae) fruits. Florida Entomologist 99(special issue 2): 114-120.