Ionizing radiation as a phytosanitary treatment against *Phenacoccus solenopsis* (Hemiptera: Pseudococcidae)

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Abstract

Phytosanitary irradiation (PI) has great potential to disinfest agricultural commodities of quarantine pests. Efficacy of gamma irradiation (5–500 Gy) was studied on various ontogenetic stages of the solenopsis mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae), which is an economic tropical postharvest pest. As *P. solenopsis* showed a parthenogenetic mode of reproduction, studies were focused on female adults to estimate the minimum dose required for sterilization by gamma radiation. The radiosensitivity of the insect decreased as its age and developmental stage increased. The most radiotolerant stage, the 11-12 d-old gravid females required 500 Gy to completely prevent F_1 egg hatch, whereas 200 Gy prevented molting of all F_1 first instar nymphs to the second instar. The next lowest dose attempted was 100 Gy, whereupon 43 F_1 first instar nymphs— hatched from 500 eggs laid by irradiated gravid females,— molted to the F_1 second instar. Therefore, a dose of ~200 Gy might suffice as a phytosanitary treatment against *P. solenopsis.* Large-scale confirmatory testing would be required to confirm a dose that could be used against this insect. Radiotolerance of potato (*Solanum tuberosum* L.; Solanales: Solanaceae)—a major host of *P. solenopsis*—was studied at 200 and 400 Gy. Color, weight, total soluble solids, titratable acidity, pH, vitamin C and ß-carotene content were not affected compared with non-irradiated controls after 7 d storage at 5 °C.

Key Words: cotton mealybug, solenopsis mealybug, molting prevention, phytosanitary irradiation, phytosanitation, potato, nutritional quality analysis

Resumen

La irradiación fitosanitaria (PI) tiene un gran potencial para desinfectar productos agrícolas de plagas cuarentenarias. Se estudió la eficacia de la irradiación gamma (5-500 Gy) en estadios diversos ontogenéticos de la cochinilla solenopsis, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae), que es una plaga económica tropical de poscosecha. Como *P. solenopsis* mostró un modo de reproducción partenogenética, los estudios enfocaron en las hembras adultas para estimar la dosis mínima requerida para la esterilización por radiación gamma. La radiosensibilidad del insecto disminuyó a medida que su edad y estadio de desarrollo se incrementó. El estadio más radiotolerante fueron las hembras grávidas de 11-12 dias de edad que requieren 500 Gy para prevenir completamente la eclosión de los huevos de la F_1, mientras que 200 Gy impidió la muda de todas las ninfas F_1 del primer estadio al segundo estadio. El siguiente intento de dosis más baja fue de 100 Gy, con lo cual F_1 43 ninfas de primer instar , nacido de 500 huevos puestos por las hembras grávidas irradiados, mudadas al segundo instar F_1. Por lo tanto, una dosis de ~ 200 Gy puede ser suficiente como tratamiento fitosanitario contra *P. solenopsis.* Pruebas a realizar que confirmen a gran escala el tratamiento serían necesarias para confirmar una dosis que podría ser utilizada contra este insecto. Se estudió la radiotolerancia - a 200 y 400 Gy de la papa (*Solanum tuberosum* L .; Solanales: Solanaceae) – un hospedero importante de *P. solenopsis*. El color, peso, sólidos solubles totales, acidez titulable, pH, contenido de vitamina C y beta-caroteno no se vieron afectados en comparación con los controles no irradiados después de un almacenamiento de 7 dias a 5 °C.

Palabras Clave: algodón cochinilla, cochinilla solenopsis, prevención de la muda, irradiación fitosanitaria, fitosanidad, papa, análisis de calidad nutricional

Mealybugs (Hemiptera: Pseudococcidae) are important plant pests worldwide (McKenzie 1967; Williams 1985; Williams and Granara de Willink 1992; Miller et al. 2002, 2005). Mealybugs are also reported as important postharvest pests, ultimately posing quarantine barriers to agricultural trade. Hallman (2011) observed that mealybugs were the third most important quarantine pest group requiring phytosanitary treatments, after tephritid fruit flies and larvae of Lepidoptera. The minute and highly mobile crawler stage as well as the high rate of reproduction of *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) makes it an especially threatening pest that they may occur on host material exported from infested areas. The pest has been added to the European and Mediterranean Plant Protection Organization list (EPPO 2011) and the Chinese quarantine pest list (MOA 2009).

Phenacoccus solenopsis Tinsley (Hemiptera: Pseudococcidae), commonly known as the solenopsis or cotton mealybug, has a wide range of variation in morphological characters and biological adaptations (Hodgson et al. 2008), and is found in diverse ecological zones of many countries (Ben-Dov et al. 2009). It damages > 200 plant species from about 24 countries of tropical and subtropical regions of the world, and is a serious threat to Asian cotton fields, where three-fourths of the world's cotton is grown.

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Phytosanitary treatments are used to disinfest host commodities of insect pests before they are exported to areas free of the pests, and which are susceptible to pest invasion and establishment. Phytosanitary irradiation (PI) is a safe and effective phytosanitary treatment that is increasing in use in an increasing number of countries (Hallman et al. 2016). The measure of efficacy of PI is different from all other phytosanitary treatments in that prevention of further development or successful reproduction instead of acute mortality is the most used criterion of efficacy (FAO 2003). This study is 1 of several in this special issue of Florida Entomologist (Doan et al. 2016; Hofmeyr et al. 2016a; Kuswadi et al. 2016; Seth et al. 2016a,b; Zhan et al. 2016) that together with previous studies led to the proposal of a generic PI treatment for all mealybugs (Hofmeyr et al. 2016b).

Radiotolerance varies among arthropod taxa, and therefore, the estimated generic doses for arthropods vary from 100 Gy for Bruchinae (seed weevils) to 350 Gy for phytophagous mites (Hallman 2012). Radiotolerance may also vary among the life stages of an insect, and Hallman et al. (2010) found that it generally increases as an insect develops through successive stages of its life cycle.

The present study was carried out to assess the radiosensitivities of the various ontogenetic stages of *P. solenopsis* as measured by survival, metamorphic inhibition, and sterility, and to develop a PI treatment against the pest. Further, the influence of gamma irradiation was ascertained on the quality characteristics of potato, *Solanum tuberosum* L. (Solanales: Solanaceae), an important host on which this quarantine pest was reared.

Materials and Methods

MEALYBUG REARING

A culture of *P. solenopsis* was procured from the National Centre for Integrated Pest Management, PUSA Campus, New Delhi, India. Rearing conditions of 27 ± 1 °C, 70 ± 5% RH, and 12:12 h L:D photoperiod were maintained in the incubators. Sprouted potatoes used for rearing the mealybugs were placed in glass jars (1 kg capacity) containing a bed of moist, sterilized soil. Adult mealybugs were then placed on the sprouted potatoes. Developmental changes in life stages (Fig. 1) were observed and recorded daily. Females started egg-laying within 11–12 d, and a total of 200–300 eggs were laid during their ovipositional period of 6–8 d. The hatching of crawlers from eggs occurred within 1–2 h after egg deposition. Female immatures had 3 instars with 7–8 d of developmental time each. Emerged crawlers (N_1) moved toward sprouts, and when they molted to N_2 (second nymphal instar) they showed a tendency to disperse. Food was replenished every wk. In case of males, the N_3 (having filamentous covering around its body) lasted for 2–3 d and molted to the N_4 stage enclosed within a cocoon-like covering and with a developmental period of 4–5 d. Longevity was 2–5 d for male adults and 32–37 d for female adults.

IRRADIATION TREATMENT

Irradiation of mealybugs was conducted at the Radiobiological Unit of the Institute of Nuclear Medicine and Allied Sciences, Ministry of Defense, Delhi using a ^{60}Co source (Gamma cell-5000, BRIT, BARC, Trombay, India). The dose rate was ~2 kGy/h. Fricke dosimetry was performed and later reconfirmed by using a thermocouple chip provided through the IAEA to measure the dose values and the dose distribution.

EXPERIMENTAL DESIGN

Various ontogenetic stages were broadly divided into 2 major groups, developing pre-imaginal stages and developed imaginal stages, to investigate the effect of irradiation on *P. solenopsis*.

Pre-imaginal Stages

The bio-efficacy of gamma radiation was evaluated on the development and reproductive behavior of *P. solenopsis,* irradiated as crawlers (N_1), second instars (N_2), third instar females (N_3-♀), and third and fourth instar males (N_3-♂ and N_4-♂) (Fig. 1) with doses ranging from 5 to 500 Gy plus non-irradiated controls. A group of ~50 individuals for N_1 and a group of 15–20 individuals for subsequent ontogenetic stages per replicate were evaluated for their biological characteristics related to metamorphosis and adult formation in response to irradiation. The pre-imaginal stages (N_1-crawlers, N_2 second instar nymphs and N_3♀-third instar female nymphs) were treated with 5–200 Gy to assess the sterilizing potential of gamma radiation. The reproductive behavior (egg laying and hatching) of the resultant adults was monitored daily; 7 individual females constituted each replicate. Effective doses for 50, 90, and 99.9% response (ED_{50}, ED_{90} and $ED_{99.9}$) with respect to inhibition in metamorphic development and adult formation were determined via linear regression, and induced sterility in resultant adults from treated pre-imaginal stages was analyzed via probit analysis (PoloPlus, Petaluma, California).

Imaginal Stages

To investigate the bio-efficacy of gamma radiation on reproductive behavior of imaginal stages of *P. solenopsis* (which exhibited parthenogenesis), only female mealybugs in 3 age groups—(i) freshly formed females (0–1 d-old), (ii) pre-gravid females (5–6 d-old), (iii) gravid females (11–12 d-old) (Fig 1.)—were exposed to gamma radiation (5–500 Gy), and their progeny were compared to those of non-irradiated controls. Radiation efficacy was further evaluated on the development and level of inherited sterility of F_1 progeny derived from the P (parent) female imaginal stage irradiated with 5–200 Gy. Thus the effective doses (ED_{50}, ED_{90} and $ED_{99.9}$) inducing 50, 90 and 99.9% sterility in the parent generation and their carry-over impacts—metamorphic inhibition and inviability in F_1 progeny due to sub-sterilized parent mealybugs—were determined.

Physico-chemical and Nutritional Quality Analysis of Irradiated Potatoes

Several physico-chemical (weight loss, color change, total soluble solids, acidity, and pH) and nutritional (vitamin C and β-carotene) characteristics of potatoes irradiated with 200 and 400 Gy and stored for 7 d at 5 °C were examined as per the following methods. The percentage weight loss of the irradiated potatoes after 1 wk of storage at 5 °C was calculated. The color properties of the potatoes were measured by using an X-Rite CA22 spectrophotometer (X-Rite Inc., Grand Rapids, Michigan, USA). Color was recorded using the CIE-Lab, where the L* value indicates lightness, the a* value indicates chromacity on a green (−) to red (+) axis, and the b* value indicates chromacity on a blue (−) to yellow (+) axis (Ranganna 1986). Total soluble solids were determined using an Abbemat refractometer (Anton Paar Instruments, Seelze-Letter, Germany) at 20 °C and expressed as a percentage by mass of total soluble solids of the total contents (DGHS 2005). Acidity was determined by titrating samples (diluted with distilled water) with 0.1 N NaOH solution using a phenolphthalein indicator. The percentage of titratable acidity was calculated (AOAC 2012). Determination of pH of

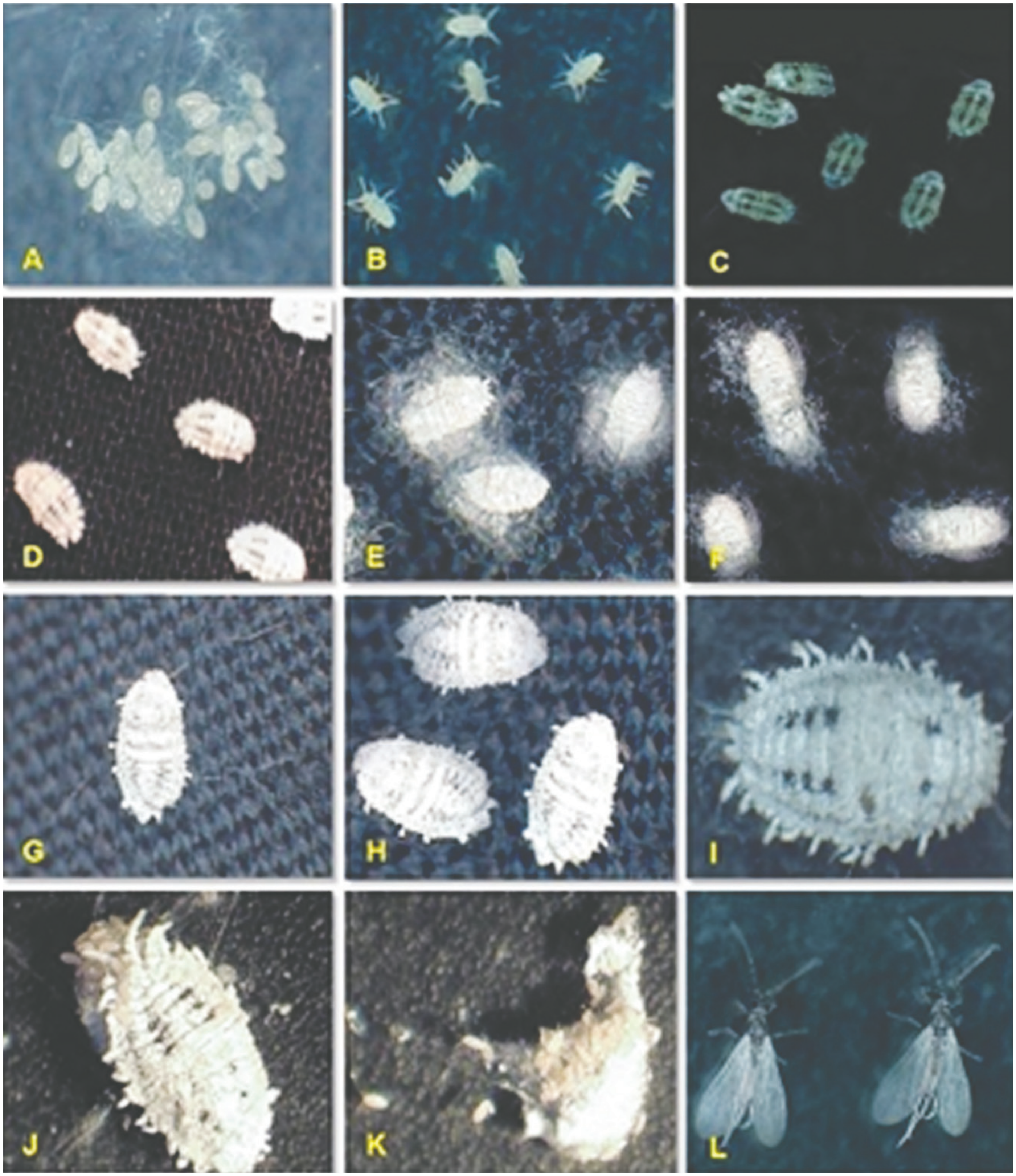

Fig. 1. Life cycle of *Phenacoccus solenopsis*: A) eggs, B) first instars (crawlers; N_1), C) second instars (N_2), D) third instar females (N_3-♀), E) third instar males (N_3-♂), F) fourth instar males (N_4-♂), G) 0-1 d-old female adult, H) 5-6 d-old (pre-gravid) female adult, I) 11-12 d-old (gravid) female adult, J) gravid female with ovisac, K) crawlers emerging from ovisac, and L) male adults.

homogenized potato was done by a pH meter (Orion-420, ThermoScientific, Waltham, Massachusetts, USA) (DGHS 2005). Vitamin C was extracted and titration performed using 2,6-dichlorophenol indophenol dye (AOAC 2012). β-Carotene was extracted and the quantification was carried out by high performance liquid chromatography (Shimadzu Scientific Instruments, Kyoto, Japan). The β-carotene was detected by absorbance at 436 nm. The concentration of carotene in the samples was determined by comparing the sample area of the peak with the standard β-carotene (AOAC 2012).

STATISTICAL ANALYSIS

The data obtained in the above experiments were usually replicated 10 times; any variation in replicate number is mentioned in the text. Effective doses ED_{50}, ED_{90}, and $ED_{99.9}$ with respect to metamorphic inhibition and adult formation were analyzed with linear regression. Prevention of reproduction was analyzed with probit analysis (PoloPlus, Petaluma, California) of the dose-response data. The data were subjected to analysis of variance (ANOVA). A probability of $P < 0.05$ was considered significant. Percentage data were transformed using arcsine √x value before ANOVA. Means were separated at the 5% significance level by least significant difference (LSD) test among the different treatments (SPSS 22.0, Armonk, New York, USA).

Results

EFFECT OF GAMMA RADIATION ON DEVELOPMENT AND SURVIVAL OF THE PRE-IMAGINAL STAGES

Effect of Irradiation of Different Pre-imaginal Stages on Metamorphic Inhibition

Figures 2–6 show the extent of reduction in developmental success of the different pre-imaginal stages of *P. solenopsis* as the radiation dose was increased from 5 to 200 Gy. Males were generally more radiosensitive than females for this characteristic. Radiosusceptibility was inversely proportional to developmental stage.

Effective doses (ED_{50}, ED_{90}, and $ED_{99.9}$) for metamorphic inhibition of the irradiated N_1 to the N_2 were statistically calculated as 36 Gy, 146 Gy and 174 Gy, respectively. Among the various pre-imaginal stages, the third instar female nymph (N_3-♀) was found to be the most radio-resistant as ED values for 50, 90 and 99.9% inhibition of metamorphosis of the irradiated N_3-♀ to the female adult were determined as 140 Gy, 301 Gy and 340 Gy, respectively. These doses were much higher than the ED values observed for metamorphic inhibition in other ontogenetic stages ($F = 65.9$ among ED_{50}; $F = 18.9$ among ED_{90}; $F = 16.4$ among $ED_{99.9}$; df = 5, 54; $P < 0.05$) (Table 1).

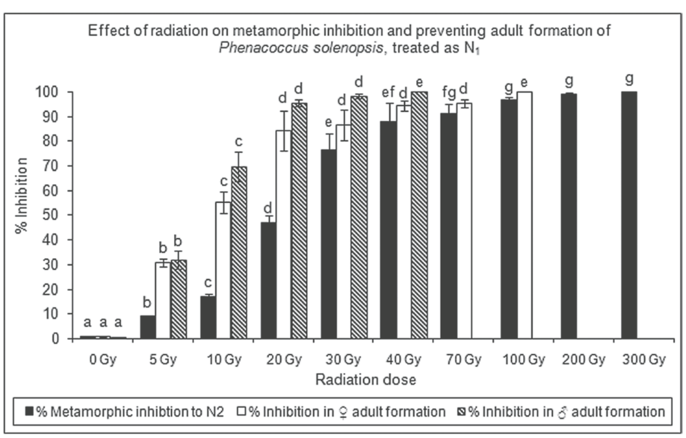

Fig. 2. Effect of various doses of gamma radiation applied to *Phenacoccus solenopsis* N_1 nymphs (crawlers) on the prevention of metamorphosis into either N_2 and subsequently into either adult males or adult females. Means followed by the same letter within each of the 3 molts are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Data were arcsine transformed before ANOVA, but data in the graphs are back transformed. Number of replicates = 10. Fifty crawlers within each treatment regimen constituted 1 replicate.

Effect of Irradiation of the Different Pre-imaginal Stages on Prevention of Adult Formation

Development of irradiated crawlers to the adult stage decreased with increasing dose of radiation (Fig. 2). Doses of 40 Gy and 100 Gy completely prevented male and female adult formation, respectively, when treated as crawlers. A dose of 100 Gy applied to the N_2 completely inhibited male and female adult formation (Fig. 3). Both development and survival of irradiated N_3♀ to the adult were prevented by 300 Gy (Fig. 4). A dose of 200 Gy applied to the N_3-♂ completely inhibited male adult formation (Fig. 5), while 400 Gy inhibited N_4-♂. A dose of 300 Gy applied to the N_4-♂ completely inhibited male adult formation (Fig. 6).

Therefore, a decrease in radiation impact was evident with an increase in ontogenetic stage. Effective doses for 99.9 % ($ED_{99.9}$) in male adult formation from irradiated N_1, N_2, N_3-♂ and N_4-♂ were computed as 32 Gy, 85 Gy, 161 Gy and 270 Gy, respectively. Whereas, the effective doses for 99.9% check ($ED_{99.9}$) in female adult formation from irradiated N_1, N_2, N_3-♀ were 74 Gy, 149 Gy and 340 Gy, respectively, they were relatively higher doses than the ED values needed to prevent male adult formation (Table 2).

Effect of Irradiation in Pre-imaginal Stages on Reproductive Behavior

Oviposition rate and egg viability exhibited an inverse relationship with increasing doses and with the progression of ontogenetic development. Egg fertility in adults developed from irradiated crawlers (N_1) was drastically affected with no egg hatch at 30 Gy. The radiosensitivity of first instar nymph (N_1) was not markedly different from that of second instar nymph (N_2) in terms of reproductive inhibition. For instance, a dose of 20 Gy resulted in 8.5% and 10.9% fertility in adults that developed from N_1 and N_2 respectively. Further, in case of third instar female nymphs (N_3-♀), the dose of irradiation was negatively correlated with ovipositional performance, and with the fertility of adults derived from N_3-♀ ($F = 278.7$; df = 4, 45; $P < 0.05$). Thus, for example, the 20 Gy dose resulted in 18.3% fertility in the resultant adult, which revealed that the older developmental stages were the more radio-resistant (Table 3). A dose of 40 Gy applied to third instar female nymphs resulted in 63.3% adult formation, all of which were completely sterile. The effective doses applied to the N_1, N_2 and N_3-♀ stages for inducing almost complete sterility ($ED_{99.9}$) were 25.7 Gy, 27.3 Gy and 35.5 Gy, respectively (Fig. 7).

EFFECT OF GAMMA RADIATION ON REPRODUCTIVE BEHAVIOR OF IMAGINAL STAGES AND THEIR F_1 PROGENY

Oviposition and Fertility of Adults Irradiated in Different Age Groups

The oviposition rate of irradiated female adults in different age groups decreased with increasing dose. For example, in the case of irradiated 5–6 d female adults, the total eggs laid were observed to be 248 at 5 Gy, which was almost the same as the total eggs laid in the control (256); whereas, an 18–67% reduction in total laid eggs was observed within the dose range of 10–500 Gy when compared to the control ($F = 734.8$; df = 11, 108; $P < 0.05$). Further, in case of 11–12

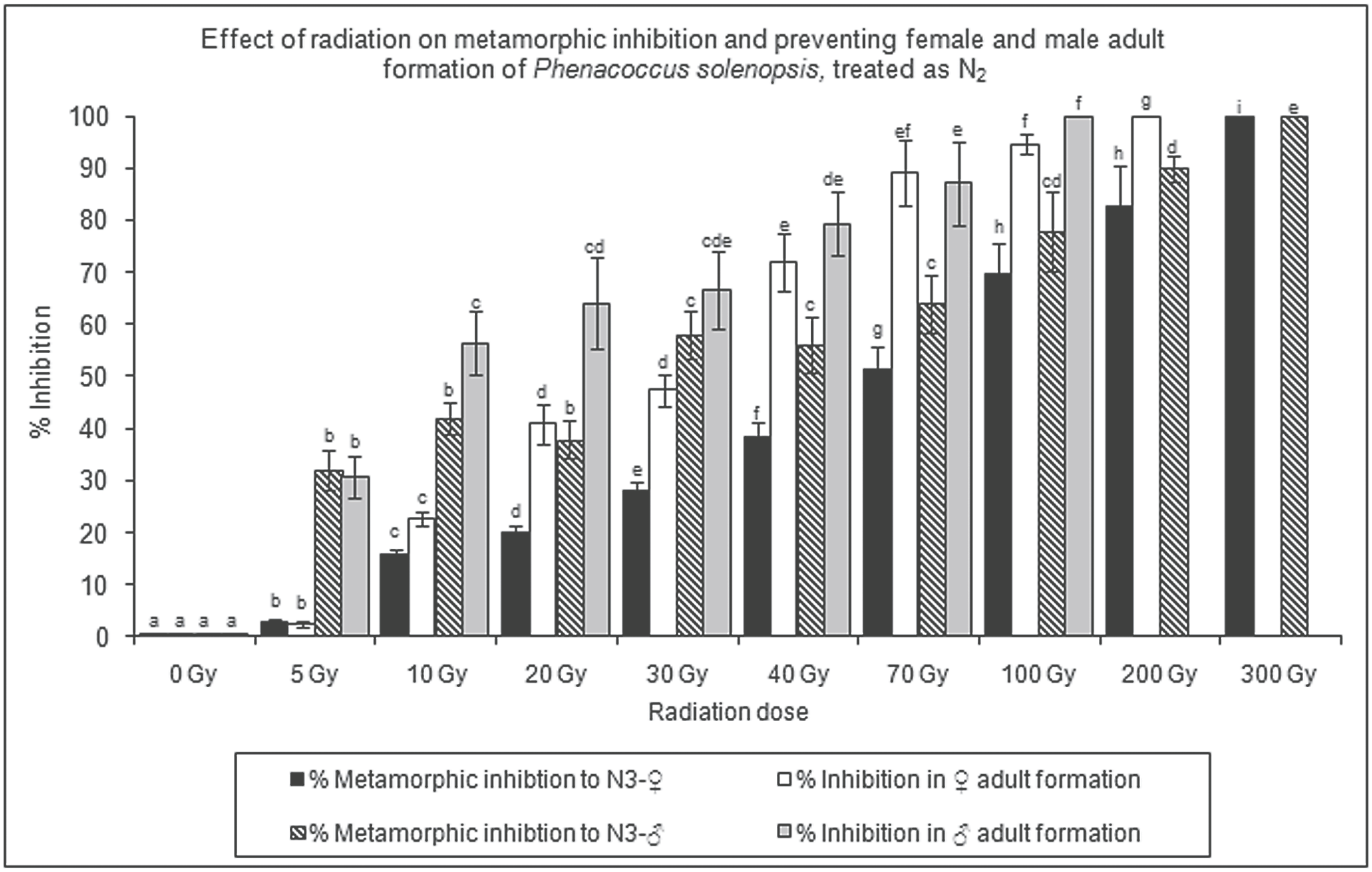

Fig. 3. Effect of various doses of gamma radiation applied to *Phenacoccus solenopsis* N_2 nymphs on the prevention of metamorphosis into either N_3-♂s and N_3-♀s or subsequently into adult males and females. Means followed by same letter within each of the 4 molts are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Percentage data were arcsine transformed before ANOVA, but data in graph are back transformed. Number of replicates = 10. Fifteen N_2 within each treatment regimen constituted 1 replicate.

d-old gravid females—where 500 Gy caused only a 42.1% reduction in number of eggs laid compared with the control—the relative impact of irradiation was relatively less than that observed in case of irradiated younger females in which the reduction in number of eggs laid reached a low of 72% for 0-1 d-old females ($F = 112.8$; df = 11, 108; $P < 0.05$). Irradiation drastically affected the fertility of 0–1 d-old female adults ($F = 512.3$; df = 6, 63; $P < 0.05$). For instance, 70 Gy or more absolutely inhibited hatching of eggs laid by irradiated 0-1d-old females. Irradia-

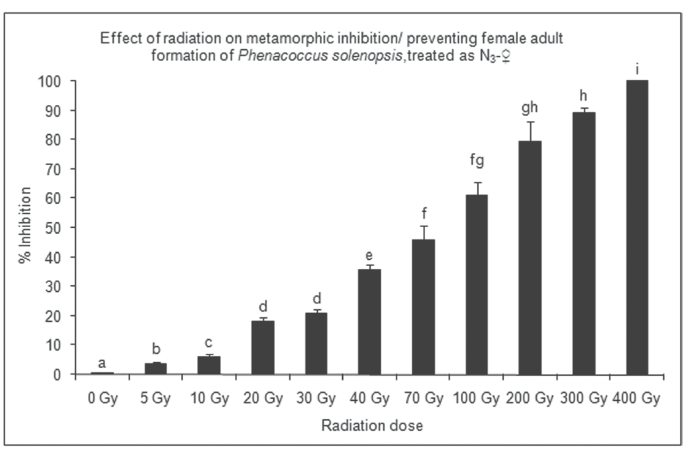

Fig. 4. Effect of various doses of gamma radiation applied to *Phenacoccus solenopsis* N_3-♀ nymphs on the prevention of metamorphosis into female adults. Means followed by same letter within the last molt are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Data were arcsine transformed before ANOVA, but data in the graphs are back transformed. Number of replicates = 10. Fifteen N_3-♀s within each treatment regimen constituted 1 replicate.

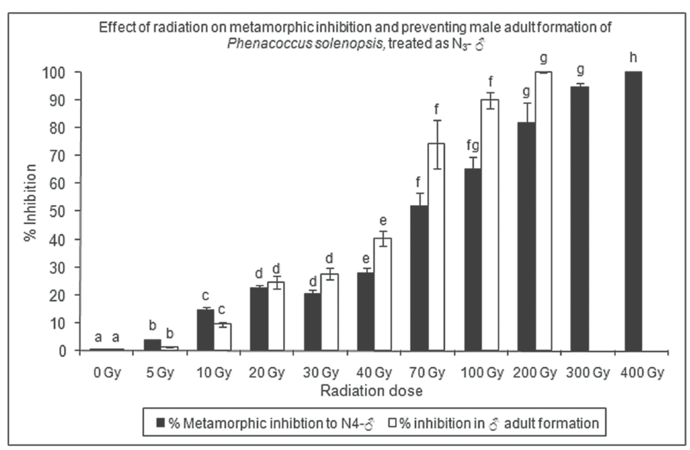

Fig. 5. Effect of various doses of gamma radiation applied to *Phenacoccus solenopsis* N_3-♂ nymphs on the prevention of metamorphosis into either N_4-♂ males or adult males. Means followed by the same letter within each of the 2 molts are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Data were arcsine transformed before ANOVA, but data in the graphs are back transformed. Number of replicates = 10. Fifteen N_3-♂s within each treatment regimen constituted 1 replicate.

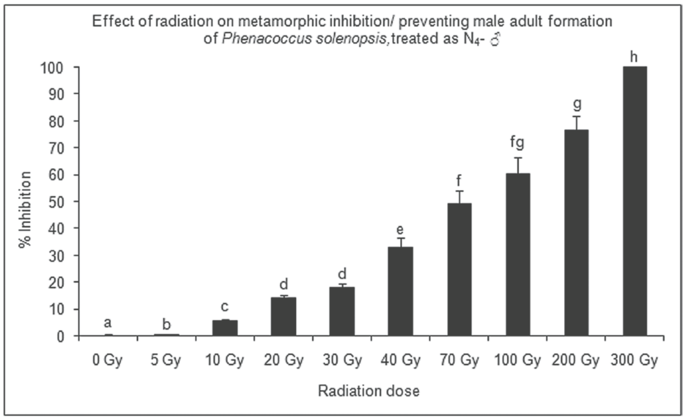

Fig. 6. Percent inhibition of development of *Phenacoccus solenopsis* N_4-♂ nymphs into male adults by various doses of gamma radiation applied to the N_4-♂ nymphs. Means followed by same letter are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Data were arcsine transformed before ANOVA, but data in the graphs are back transformed. Number of replicates = 10. Fifteen N_4-♂s within each treatment regimen constituted 1 replicate.

tion of pre-gravid (5–6 d-old) females resulted in 100% egg inviability at 200 Gy or more, whereas 11–12 d-old females irradiated with around 400 Gy produced no viable eggs (complete sterility) (Table 4).

After the dose response test on fertility of females, the $ED_{99.9}$ doses for sterility were computed as 52, 138 and 374 Gy for females irradiated at the ages 0–1d, 5–6d (pre-gravid), and 11–12d (gravid), respectively, ($F = 259.5$; df = 2, 27; $P < 0.05$) (Fig. 8).

Parent (P) Irradiation-Induced Effects on Metamorphosis and Development in F_1 Progeny

Prevention of development of the F_1 N_2 from the N_1 could be considered as a criterion for efficacy of phytosanitary irradiation treatment against this species, as > 400 Gy would be required to prevent egg hatch when the irradiated gravid female contained eggs that were ready to hatch (Table 4). Effective doses for 50%, 90% and 99.9% metamorphic inhibition of F_1 generation N_1 nymphs to N_2 nymphs were computed as 75, 150, and 169 Gy, respectively, when the most radio-resistant (11–12 d) female parents were irradiated (Table 5). The percentages of irradiated F_1 N_1 nymphs that developed to the N_2 stage for 0, 5, 10, 20, 30, 40, 70, 100, and 200 Gy doses applied to 11–12 d-old adult females were 98, 90, 84, 81, 71, 67, 44, 8.6, and 0%, respectively. F_1 adult formation in descendants from sub-sterilized 0–1d male adults also were significantly reduced with increasing doses of radiation as compared to the control ($F = 58.5$; df = 5, 54 for female adults; and $F = 21.2$; df = 4, 45 for male adults; $P < 0.05$).

Differential responses of male and female development due to irradiation were observed. For example, 30 Gy resulted in 10.6% F_1 female adult formation, whereas it completely inhibited F_1 male adult formation (Table 6). F_1 adult formation in descendants from irradiated 5–6 d-old P females, was significantly reduced with increasing doses of radiation (5–70 Gy) as compared to the control ($F = 70.6$; df = 6, 63 for female adult and $F = 25.9$; df = 5, 54 for male adult; $P < 0.05$). Furthermore, in the case of the F_1 progeny of 11–12 d-old female mealybugs, there was a dose-dependent decrease in adult male ($F = 40.0$; df = 6, 63; $P < 0.05$) and adult female ($F = 62.7$; df = 7, 72; $P < 0.05$) development, with an apparently greater radiosensitive response by the males. Thus, for example, 70 Gy resulted in 2.2% F_1 female adult formation, while it completely inhibited F_1 male adult formation.

The estimated effective radiation doses used to treat P generation females for 99.9% inhibition of survival of F_1 progeny up to the F_1 male adult were 28, 35, and 61 Gy when applied to 0–1, 5–6, and 11–12 (gravid) d-old P generation females, respectively. Likewise, the $ED_{99.9}$ of P generation females for 99.9 % inhibition of survival of F_1 progeny up to F_1 female adult were 36, 55, and 79 Gy when treated as 0–1, 5–6, 11–12 d-old P generation females, respectively (Fig. 9). Gravid (11–12 d-old) females were found to require quite high radiation doses to inhibit 50, 90, and 99.9 % of F_1 male and female adult formation ($F = 46.0$ among ED_{50}; $F = 64.8$ among ED_{90}; $F = 66.2$ among $ED_{99.9}$; df = 5, 54; $P < 0.05$). The radio-sensitivity of the parents of with respect to the formation of adult male F_1 progeny was evidently higher than their radio-sensitivity with respect to the formation of adult female progeny.

Impact on F_1 Reproduction Induced by Sub-sterilizing Irradiation of the Parent

The F_1 oviposition rates along with their oviposition periods were affected due to the sub-sterilization of P (parent) generation mealybugs. For instance, in the case of 11–12 d-old females, 5–70 Gy caused 22.6–93.6 % reduction in total eggs laid per F_1 female compared to the

Table 1. Prevention of molting to the subsequent developmental stage in *Phenacoccus solenopsis* by gamma irradiation of the various pre-imaginal stages.

	Estimated dose[a] (Gy) to prevent 50, 90, and 99.9% from reaching the succeeding stage			
Pre-imaginal stage irradiated and the target stage to be prevented	ED_{50}	ED_{90}	$ED_{99.9}$	Linear regression equation
N_1 to N_2	36 ± 3a	146 ± 10a	174 ± 12a	y = 0.363x + 36.86 R^2 = 0.5371
N_2 to N_3-♂	55 ± 4b	212 ± 18b	251 ± 21b	y = 0.254x + 36.07 R^2 = 0.7223
N_2 to N_3-♀	106 ± 8c	229 ± 16c	260 ± 19b	y = 0.323x + 15.87 R^2 = 0.8696
N_3-♂ to N_4-♂	114 ± 8cd	242 ± 16cd	273 ± 18bc	y = 0.314x + 14.01 R^2 = 0.8771
N_3-♀ to ♀adult	140 ± 11d	301 ± 24d	340 ± 27c	y = 0.248x + 15.32 R^2 = 0.8702
N_4-♂ to ♂adult	120 ± 9cd	240 ± 18cd	270 ± 21b	y = 0.333x + 9.94 R^2 = 0.9047
F-value	*F* = 65.9	*F* = 18.9	*F* = 16.4	for slopes: *F* = 9.03

[a] ED_{50}, ED_{90} and $ED_{99.9}$ are the effective doses that prevent molting and induce either 50, 90 or 99.9% mortality, respectively. Means followed by same letter within a column are not significantly different at $P < 0.05$ level (ANOVA, df = 5, 54; followed by LSD post-test). Number of replicates = 10. Fifty insects for N_1, and 12–15 insects each for the N_2, N_3♂, N_3♀ and N_4-♂ instars constituted 1 replicate.

Table 2. Irradiation doses applied to various nymphal stages of *Phenacoccus solenopsis* required to prevent either 50, 90 or 99.9% of the irradiated nymphs from ultimately developing into adults.

Pre-imaginal stage irradiated to prevent adult formation	Estimated dose[a] (Gy) for prevention of 50, 90, and 99.9% from reaching the adult stage			
	ED_{50}	ED_{90}	$ED_{99.9}$	Regression equation
N_1 to ♂ adult	10.7 ± 0.78a	27.7 ± 2.0a	31.9 ± 2.3a	y = 2.3533x + 24.669 R^2 = 0.7653
N_1 to ♀ adult	11.1 ± 0.73a	62 ± 4b	74.6 ± 4.9b	y = 0.7858x + 41.246 R^2 = 0.5656
N_2 to ♂ adult	20.8 ± 1.9b	72 ± 6.7b	84.6 ± 7.9b	y = 0.7817x + 33.712 R^2 = 0.7148
N_2 to ♀ adult	48.3 ± 2.8c	129 ± 7.6c	149 ± 8.8c	y = 0.4958x + 26.023 R^2 = 0.6691
N_3-♂ to ♂ adult	69.4 ± 4.9d	143 ± 10c	161 ± 12c	y = 0.5421x + 12.367 R^2 = 0.8273
N_3-♀ to ♀ adult	140 ± 11e	301 ± 24d	340 ± 27e	y = 0.2484x + 15.322 R^2 = 0.8702
N_4-♂ to ♂ adult	120 ± 9e	240 ± 18d	270 ± 21d	y = 0.3331x + 9.9394 R^2 = 0.9047
F-value	*F* = 217.5	*F* = 120.5	*F* = 115.1	for slope: *F* = 188.5

[a]$ED_{50,}$ $ED_{90,}$ and $ED_{99.9}$ are effective doses that prevent either 50%, 90% or 99.9% from developing into adults, respectively. Means followed by same letter within a column are not significantly different at $P < 0.05$ level (ANOVA, df = 6, 63; followed by LSD post-test). Number of replicates = 10. Fifty insects for N_1, and 12–15 insects each for the $N_{2,}$ N_3♂, N_3♀ and N_4-♂ instars constituted 1 replicate.

control (*F* = 462.4; df = 6, 63; *P* < 0.05). Fertility of F_1 adults derived from P generation 0–1 d female adults was completely inhibited at 30 Gy (*F* = 200.6; df = 4, 45; *P* < 0.05). Furthermore, F_1 adults derived from irradiated P 5–6-d-old and P 11–12-d-old females were completely infertile at 40 Gy and 70 Gy, respectively (Table 7).

The doses used to irradiate 0–1, 5–6, 11–12 d-old P generation females estimated to induce 99.9% F_1 sterility were 26, 35, and 57 Gy, respectively (*F* = 109.1; df = 2, 27; *P* < 0.05) (Fig. 10).

PHYSICO-CHEMICAL AND NUTRITIONAL QUALITY OF POTATOES SUBJECTED TO PHYTOSANITARY IRRADIATION

No significant differences (*P* ≤ 0.05) were observed for weight loss, color, total soluble solids, titratable acidity, pH, vitamin C content, or β-carotene content between non-irradiated potatoes and those irradiated at either 200 or 400 Gy and then stored at 5 °C for 7 d (Table 8).

Discussion

The present radiobiological investigation of *P. solenopsis* was undertaken to understand physiological or developmental responses of each of the ontogenetic stages submitted to doses of irradiation that could be used in phytosanitary irradiation treatments. This study would not only predict the efficiency of using phytosanitary irradiation, but also indicate the carry-over effects of sublethal irradiation that could be apparent due to a logistical limitation. This study might also help in steering overall phytosanitary irradiation strategy in the direction of taking advantage of the considerable sustained impacts of irradiation on pests with diffuse chromosomes, which include Hemiptera, Lepidoptera and Arachnida.

BIO-EFFICACY OF RADIATION ON PRE-IMAGINAL STAGES, *PHENACOCCUS SOLENOPSIS*

Metamorphosis is a physiological process implying active cell division. It is, therefore, expected to be highly susceptible to irradiation. In the present study, the mortality of the pre-imaginal stages was assessed in terms of metamorphic inhibition due to irradiation at the different ontogenetic stages. Survival of growing nymphs to the adult stage was observed to decrease with increasing radiation doses. It showed that some irradiated crawlers developed to the adult stage at doses up to 70 Gy range but these adults did not reproduce. Similarly, Ravuiwasa et al. (2009) reported that *Planococcus minor* Maskell (Hemiptera: Pseudococcidae) treated as crawlers with 150–250 Gy survived to adult stage but were unable to reproduce. Another study conducted on *Dysmicoccus neobrevipes* Beardsley (Hemiptera: Pseudococcidae) demonstrated that 1.3–11.3% of adults developed from treated crawlers at 100–200 Gy, but reproduction was inhibited at 200 Gy (Doan et al. 2012).

The present study demonstrated that adult development from irradiated N_2 stage *P. solenopsis* ranged from 61% at 5 Gy to 3.3% at 100 Gy, whereas, oviposition was observed only at doses up to 30 Gy. Furthermore, irradiation with 30 Gy inhibited the hatching of the eggs laid by adults that had developed from treated N_1 and N_2 stages. Doan et al. (2012) reported that 8–27% of *D. neobrevipes* N_3 nymphs survived to the adult stage at all tested irradiation doses (100–250 Gy), and that the adults that developed from N_3 females irradiated with 200–250 Gy were completely sterile. In the present study some adults developed from female N_3 nymphs irradiated at 70–300 Gy, but none laid viable eggs.

Phenacoccus solenopsis male nymphs were found to be more radiosensitive than female nymphs in terms of inhibition of development, which was consistent with the findings of Jacobsen & Hara (2003) and Seth et al. (2009). For example, male adult formation of irradiated crawlers of *P. solenopsis* was completely inhibited at 40 Gy, whereas the corresponding complete inhibition of female adult formation required 100 Gy.

Effective doses for 99.9% prevention of metamorphosis to the succeeding stage were 174 Gy applied to the N_1 stage and 340 Gy applied to the N_3-♀ stage. Radiosensitivity of irradiated pre-imaginal stages of *P. solenopsis* decreased as nymphs grew and developed. The increased radiosensitivity in developing pre-imaginal stages might be attributed to the fact that these ontogenetic stages require radical cell division and differentiation as suggested in Tilton & Brower (1983) and Jacobsen & Hara (2003). Therefore, the third instar female nymph being the penultimate stage to adult development was the most radio-tolerant among various irradiated pre-imaginal stages.

Table 3. Reproductive performance—number of eggs laid and percent that hatched—of *Phenacoccus solenopsis* females that were gamma-irradiated while they were still in various pre-imaginal stages.

Pre-imaginal stage irradiated	Reproductive parameters	Radiation doses applied to parent generation							*F*-value
		0 Gy	5 Gy	10 Gy	20 Gy	30 Gy	40 Gy	70 Gy	
First instar	Eggs laid per female	269.6 ± 19.8a	215.7 ± 16.3a	124.8 ± 7.0b	35.6 ± 2.7c	9.9 ± 1.0d	0e	—	*F* = 782, df = 5,54
	Egg hatch (%)	98.6 ± 0.76a	58.9 ± 2.4b	24.6 ± 2.9c	8.5 ± 1.0d	0e	—	—	*F* = 475, df = 4,45
Second instar	Eggs laid per female	260.2 ± 12.7a	224.4 ± 11.9b	154.3 ± 7.3c	49.5 ± 2.6d	21.4 ± 1.1e	0f	—	*F* = 842, df = 5,54
	Egg hatch (%)	96.9 ± 1.2a	69.9 ± 2.5b	28.7 ± 1.4c	10.9 ± 0.85d	0e	—		*F* = 668, df = 4,45
Third instar female	Eggs laid per female	242.5 ± 14.3a	215.2 ± 16.2a	157.3 ± 8.0b	119.5 ± 4.4c	80.3d ± 7.3d	60.3 ± 4.3e	0f	*F* = 672, df = 6,63
	Egg hatch (%)	97.5 ± 0.31a	82.3 ± 5.2b	38.7 ± 1.0c	18.3 ± 1.2d	6.9 ± 0.44e	0f	—	*F* = 279, df = 5,54

Means ± SE followed by same letter within a row are not significantly different at *P*< 0.05 level (ANOVA followed by LSD post-test). Percentage data were arcsine transformed before ANOVA, but data in the table are back transformations. Number of replicates = 10. Average data of 7 parthenogenetic females within each treatment regimen constituted 1 replicate.

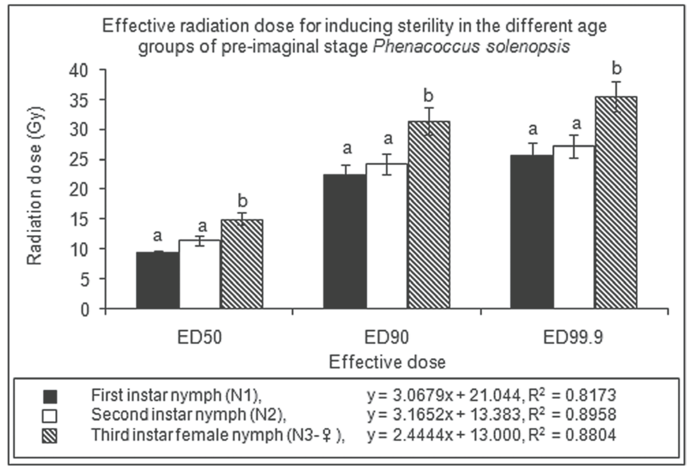

Fig. 7. Estimated doses to induce 50, 90, and 99.9% sterility in *Phenacoccus solenopsis* irradiated in different pre-imaginal stages. Means followed by same letter within a level of control are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Number of replicates = 10. Average of the data of 7 females within each treatment regimen constituted 1 replicate.

Phytosanitary irradiation differs from all other commercial treatments in 1 important technical consideration that the end point of the treatment may be prevention of further biological development and/or reproduction, rather than acute mortality (Hallman 2011). Irradiation protracted the developmental period of male and female mealybugs proportionately to the doses applied. However, radiation-mediated delay in development of male mealybugs was greater than in female mealybugs. This protraction of the developmental period due to irradiation could have a physiological effect on its later development, possibly resulting in sterility of subsequent adults (Ravuiwasa et al. 2009). In agreement with this suggestion, the present study concluded that reproductive sterilization was induced in adults that developed from irradiated pre-imaginal stages at estimated doses of 25 to 36 Gy.

BIO-EFFICACY OF RADIATION ON IMAGINAL STAGES, *PHENACOCCUS SOLENOPSIS*

The target of a phytosanitary treatment must be the most tolerant life stage of quarantine pests that can infest the shipped commodity,

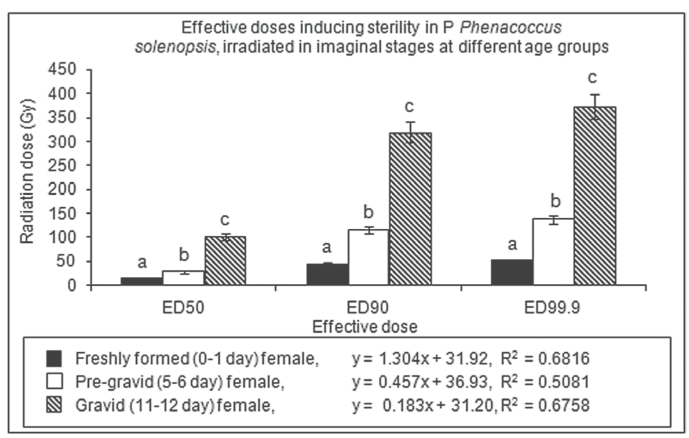

Fig. 8. Estimated doses to induce 50, 90, and 99.9% sterility in *Phenacoccus solenopsis*, irradiated as adults in 3 age groups. Means followed by same letter within a level of control are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Number of replicates = 10. Average of the data of 7 females within each treatment regimen constituted 1 replicate.

Table 4. Reproductive performance—number of eggs laid and percent that hatched—of *Phenacoccus solenopsis* females gamma-irradiated as adults in different age groups.

Age group of adult females when irradiated	Reproductive parameters	Radiation doses												*F*-value
		0 Gy	5 Gy	10 Gy	20 Gy	30 Gy	40 Gy	70 Gy	100 Gy	200 Gy	300 Gy	400 Gy	500 Gy	
Freshly formed female (0–1 d-old)	Eggs laid per female	224.8 ± 17.1a	220.7 ± 8.8a	187.7 ± 14.6ab	185.4 ± 17.1ab	190.1 ± 9.8ab	166.7 ± 14.6bc	132.4 ± 14.2c	141.3 ± 7.3c	121.5 ± 13.8c	96.6 ± 7.8cd	62.3 ± 12.1d	60.6 ± 6.6d	F=478.3 df=11,108
	Egg hatch (%)	97.5 ± 0.3a	60.2 ± 3.8b	20.8 ± 2.0c	13.6 ± 1.1d	8.2 ± 0.9e	1.4 ± 0.1f	0g	—	—	—	—	—	F = 512.3 df = 6,63
Pre-gravid female (5–6 d-old)	Eggs laid per female	256.9 ± 24.7a	248.3 ± 17.1a	199.9 ± 15.6ab	202.2 ± 14.7ab	209.3 ± 18.8ab	173.0 ± 15.3b	159.6 ± 9.1bc	139.6 ± 11.5c	108.5 ± 10.6d	91.2 ± 5.9d	95.6 ± 7.0d	85.0 ± 5.1d	F = 734.8 df=11,108
	Egg hatch (%)	97.8 ± 0.4a	87.4 ± 5.7ab	75.0 ± 5.1b	51.5 ± 2.8c	12.3 ± 0.8d	6.4 ± 1.4e	2.7 ± 0.6f	1.2 ± 0.3g	0h	—	—	—	F = 238.3 df = 8,81
Gravid female (11–12 d-old)	Eggs laid per female	268.6 ± 28.4a	267.5 ± 10.5a	244.1 ± 12.3b	225.7 ± 28.5bc	247.1 ± 9.6bc	240.8 ± 18.6bc	239.1 ± 24.3bc	202.1 ± 13.9bc	209.6 ± 16.3bc	181.3 ± 15.7bc	180.2 ± 15.1bc	155.2 ± 12.5c	F = 112.8 df=11,108
	Egg hatch (%)	97.6 ± 0.5a	92.4 ± 1.9ab	84.6 ± 5.9b	69.4 ± 2.7c	60.9 ± 2.8d	43.7 ± 3.7e	28.1 ± 2.6f	15.9 ± 1.9g	4.9 ± 1.2h	2.4 ± 0.9i	1.1 ± 0.6i	0j	F = 191.0 df=11,108

Means followed by the same letter within a row are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test); percentage data were arcsine transformed before ANOVA, but data in table are back transformed. Number of replicates = 10. Average data of 7 parthenogenetic females within each treatment regimen constituted a replicate.

regardless of whether that stage is less commonly found than others. The radiotolerance of an arthropod species increases progressively as the successive immature stages develop (Hallman et al. 2010). When the adult stage can occur in the commodity, then the measure of treatment efficacy should be the prevention of reproduction of adult. The adult female mealybug is a serious surface quarantine pest. As thelytokus parthenogenesis was observed to be the main reproductive mode in some mealybug species including *P. solenopsis*, the female adults necessarily were the primary focus of the present study. Furthermore, our elucidation of development and reproduction of the F_1 progeny due to sub-sterilizing doses applied to P generation adults—together with our findings on radiosensitivity—are useful for developing PI treatments that would be efficacious for large consignments of various host commodities.

In similar investigations involving parthenogenetic female adults, *D. neobrevipes* (Doan et al. 2012) and *P. minor* (Ravuiwasa et al. 2009) were studied to determine the most tolerant stage and the minimum dose to control these mealybug species. The measure of efficacy for a PI treatment may be prevention of adult emergence, induction of adult sterility, prevention of further F_1 development, or F_1 sterility. The present study quantified the degree of sterility induced in female mealybugs by irradiation of females of different age groups. As the females aged, their radio-tolerance increased. Hence, the $ED_{99.9}$ doses were 52, 138, and 374 Gy for 0–1, 5–6, and 11–12 d-old females, respectively.

The present study demonstrated that 4.8% crawlers emerged from eggs laid by gravid female of *P. solenopsis* irradiated with 200 Gy, but none of them developed into second instar nymph (N_2). Hence, 200 Gy might serve as a phytosanitary treatment against this mealybug. Large-scale confirmatory testing in which many thousands of gravid adult females are irradiated at a single dose that allows no F_1 N_2 nymphs to be produced would normally be required to substantiate a confirmatory test for commercial approval and use (Schortemeyer et al. 2011).

The studies conducted on another parthenogenetic species, *P. minor*, found that a dose of 100 Gy caused complete sterility in the parent female mealybug (Ravuiwasa et al. 2009). The higher radioresistance of *P. solenopsis* might be attributed to its ovoviviparous nature, exhibiting embryonic development within the female body; therefore, egg hatch could be observed within 1–2 h of egg laying.

In a similar finding, 200–250 Gy for inducing adult sterility was reported in asexually reproducing and ovoviviparous mealybug species, *D. neobrevipes* (Doan et al. 2012). Irradiation affected the physiology of insects as was reflected in their impaired reproductive behavior—viz., prolonged pre-oviposition period, shortened oviposition period and enhanced degree of sterility—and reduced survival, which would result in successful phytosanitary radiation treatments. Furthermore, irradiation was observed to delay the onset of oviposition in adults developed from pre-imaginal and imaginal stages of *P. solenopsis*. Similarly, Doan et al. (2012) indicated that female survivors that developed from irradiated N_1, N_2, N_3-♀ nymphs, and irradiated adult females of *D. neobrevipes* showed delayed reproduction.

The order of radio-tolerance among different life stages in terms of induced sterility in the adult stage of *P. solenopsis* was $N_1 < N_2 < N_3$ ♀< adult. This pattern of radioresistance was consistent with the other parthenogenetic mealybugs (Ravuiwasa et al. 2009; Doan et al. 2012).

Unlike other disinfestation techniques, irradiation does not need to kill the pest immediately to provide quarantine security; therefore, live but inviable insects may occur with the exported commodity, eliminating the value of inspection for live target pests and placing reliance on verification of treatment application and efficacy.

Studies on mechanisms behind tolerance to ionizing radiation in mealybugs were reported by Chandra (1963a, b). Cytological examina-

Table 5. Estimated gamma radiation doses (Gy) applied to P generation *Phenacoccus solenopsis* adult females of different age groups to prevent the metamorphosis of F_1 first instars (N_1) into second instars (N_2).

Age group of P generation adult females when irradiated	Estimated dose (Gy) to prevent the molt of F_1 N_1 to N_2			Regression equation
	ED_{50}	ED_{90}	$ED_{99.9}$	
Freshly formed female (0–1 d-old)	21.1 ± 1.2a	37.5 ± 2.1a	41.6 ± 2.3a	y = 2.436x – 1.47 R^2 = 0.96
Pre-gravid female (5–6 d-old)	34.2 ± 1.9b	61.1 ± 3.4b	67.7 ± 3.8b	y = 1.490x – 1.05 R^2 = 0.99
Gravid female (11–12 d-old)	74.7 ± 4.9c	150 ± 10.1c	169 ± 11.6c	y = 0.528x + 10.57 R^2 = 0.88
F-value	*F* = 218.1	*F* = 247.3	*F* = 270.6	for slope: *F* = 279.4

ED_{50}, ED_{90}, and $ED_{99.9}$ are effective doses that prevent 50%, 90%, 99.9%, of F_1 N_1 to molt to N_2, respectively. Means followed by same letter within a column are not significantly different at $P < 0.05$ level (ANOVA, df = 2, 27; followed by LSD post-test).

Number of replicates = 10. Fifty F_1 N_1 nymphs from each treatment regimen constituted 1 replicate.

Table 6. Survival to the adult stage of the F_1 progeny of *Phenacoccus solenopsis* P generation adult females that were gamma-irradiated in different age groups.

Age group of P generation adult females when irradiated	Sex of F_1 adults formed	Percent survival to adulthood[a] of the F_1 progeny of a parent irradiated as an adult								*F*-value
		0 Gy	5 Gy	10 Gy	20 Gy	30 Gy	40 Gy	70 Gy	100 Gy	
Freshly formed female (0–1 d-old)	Male	21.9 ± 2.0a	18.0 ± 2.6a	8.6 ± 0.49b	5.2 ± 0.36c	0d	—	—	—	*F* = 21.2 df = 4,45
	Female	62.8 ± 2.9a	50.0 ± 2.2b	32.6 ± 1.8c	17.6 ± 1.3d	10.6 ± 0.83e	0f	—	—	*F* = 58.5 df = 5,54
Pre-gravid female (5–6 d-old)	Male	17.1 ± 1.5a	16.1 ± 1.2ab	14.2 ± 1.4ab	12.6 ± 0.99b	3.3 ± 0.24c	0d	—	—	*F* = 25.9 df = 5,54
	Female	72.1 ± 3.4a	56.6 ± 3.13b	39.9 ± 2.1c	28.0 ± 1.4d	14.5 ± 0.81e	3.2 ± 0.21e	0f	—	*F* = 70.6 df = 6,63
Gravid female (11–12 d-old)	Male	19.4 ± 1.4a	18.7 ± 1.4a	15.7 ± 1.2ab	11.3 ± 1.2b	8.0 ± 0.45c	3.3 ± 0.24d	0e	—	*F* = 40.0 df = 6,63
	Female	74.5 ± 5.8a	58.6 ± 2.5b	46.6 ± 2.5c	35.3 ± 2.6d	22.0 ± 1.72e	9.6 ± 0.82f	2.2 ± 0.17g	0h	*F* = 62.7 df = 7,72

[a]Means followed by the same letter within a row are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Data were arcsine transformed before ANOVA, but data in table are back transformed. Number of replicates = 10. Fifty F_1 eggs from each treatment regimen constitute 1 replicate.

Table 7. Reproductive performance—number of eggs laid and percent that hatched—of the F_1 female progeny of *Phenacoccus solenopsis* P generation adult females that were gamma-irradiated in different age groups.

Age group of P generation adult females when irradiated	Reproductive parameters	Radiation doses							*F*-value
		0 Gy	5 Gy	10 Gy	20 Gy	30 Gy	40 Gy	70 Gy	
Freshly formed female (0–1 d-old)	Eggs laid per female	264 ± 18.5a	188 ± 14.7b	120 ± 8.9c	87 ± 6.8d	24 ± 1.0e	—	—	*F* = 562.3, df = 4, 45
	Egg hatch (%)	98.3 ± 0.29a	67.4 ± 3.1b	26.5 ± 2.5c	13.3 ± 2.4d	0e	_	_	*F* = 200.6, df = 4,45
Pre-gravid female (5–6 d-old)	Eggs laid per female	291.0 ± 17.9a	253.2 ± 21.9a	143 ± 8.4b	110 ± 7.5c	88 ± 5.8d	49 ± 2.3e	0f	*F* = 365.7, df = 5, 54
	Egg hatch (%)	98.4 ± 0.40a	81.7 ± 3.2b	34.2 ± 2.1c	28.1 ± 1.1d	11.0 ±0.98e	0f	_	*F* = 175.5, df = 5,54
Gravid female (11–12 d-old)	Eggs laid per female	266 ± 23.2a	206 ± 25.5ab	202 ± 20.8ab	164 ± 19.9bc	125 ± 8.5c	89 ± 4.2d	18 ± 2.1e	*F* = 462.4, df = 6,63
	Egg hatch (%)	96.2 ± 0.54a	81.9 ± 1.7b	73.4 ± 2.5c	56.3 ± 3.2d	19.3 ± 1.4e	8.4 ± 1.0f	0g	*F* = 287.7, df = 6,63

Means followed by same letter within a row are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test); percentage data were arcsine transformed before ANOVA, but data in table are back transformed. Number of replicates = 10, and the average data of 7 parthenogenetic females within each treatment regimen comprised 1 replicate.

tion of the progeny obtained from paternal irradiation with 1.2 kGy revealed very small chromosome fragments that appeared to retain their individual chromosome identity such as replication, metaphase alignment, and anaphase segregation. Mohan et al. (2012) suggested that there was highly efficient protection of radiation-induced chromosome ends (telomeric repeats) that would prevent the onset of fusion-bridge-breakage-fusion cycles, and that this machinery was complemented by the diffuse centromere property in mealybug chromosomes. This property of the mealybug centromere is distributed throughout the chromosomes.

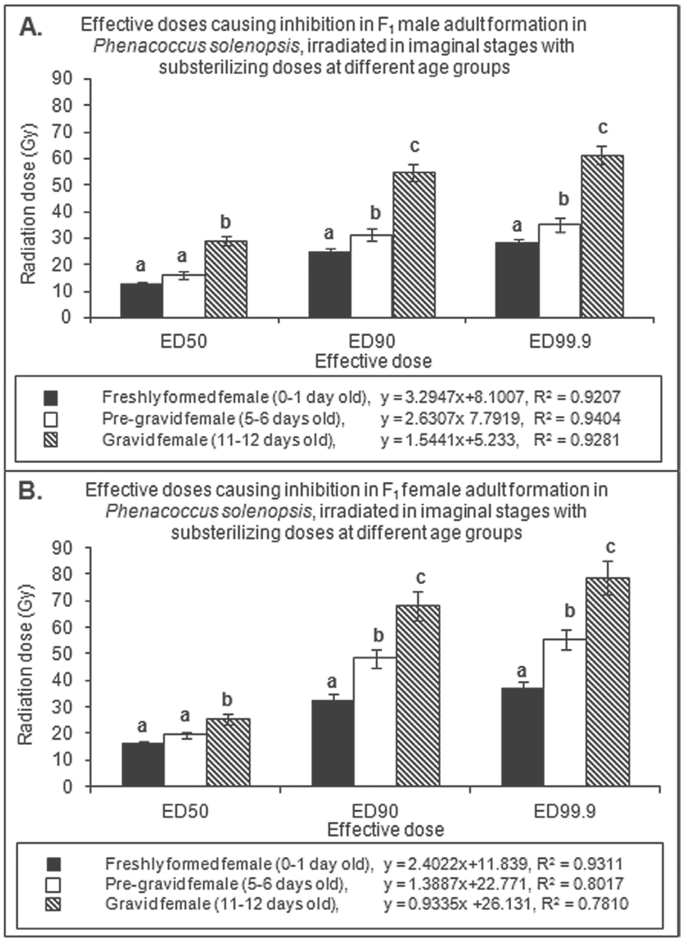

Fig. 9. Estimated doses of gamma radiation applied to 3 different age groups of *Phenacoccus solenopsis* females for preventing their offspring from developing to the adult stage, i.e., prevention of F_1 adult formation: A. Doses for preventing F_1 male adults, and B. Doses for preventing F_1 female adults. Means followed by same letter within a level of control are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Number of replicates = 10. Average of the data of 7 females within each treatment regimen constituted 1 replicate.

A dose range of 250 Gy has been suggested for phytosanitary irradiation of mealybug species (Hofmeyr et al. 2016). A generic treatment comprising at least these 4 families (Aleyrodidae, Coccidae, Diaspididae, and Pseudococcidae) of Hemiptera might be ~250 Gy (Hallman et al. 2010). Many aleyrodids, coccids, diaspidids, and pseudococcids are quarantine pests of commodities currently treated by the APHIS (2016) generic dose of 400 Gy and would be prime candidates for research aimed at lowering the doses applied to host commodities.

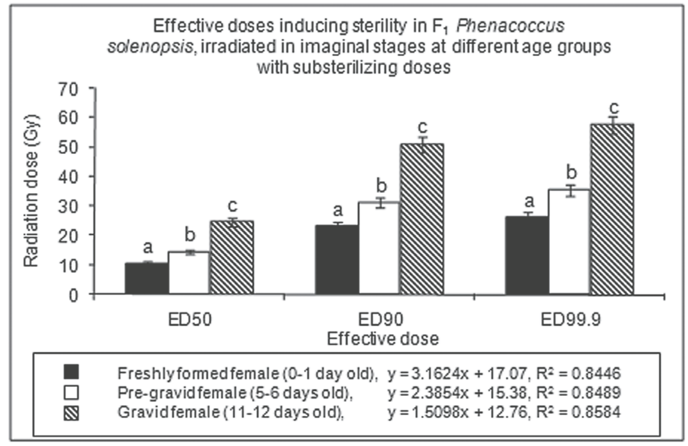

Fig. 10. Estimated doses of gamma radiation applied to 3 different age groups of P *Phenacoccus solenopsis* females to induce 50, 90, and 99.9% F_1 sterility, i.e., prevent 50, 90, and 99.9% of F_2 egg hatch. Means followed by same letter within a level of control are not significantly different at $P < 0.05$ level (ANOVA followed by LSD post-test). Number of replicates = 10. Average of the data of 7 females within each treatment regimen constituted 1 replicate.

SENSORY AND NUTRITIONAL QUALITY OF IRRADIATED POTATOES

Present study showed that irradiation at 200 and 400 Gy did not affect the quality of treated potato. Janave & Thomas (1979) reported a slight decrease in β-carotene content in irradiated potatoes. Blessington et al. (2007) noted that storage itself had a much greater detrimental effect on nutritive value of potato than did irradiation at 200 Gy.

As per earlier reports on phytosanitary irradiation on various mealybug species, > 200 Gy has been suggested as an effective quarantine dose for *Pseudococcus comstocki* Kuwana (Hemiptera: Pseudococcidae) (Dohino & Masaki 1995) *Maconellicoccus hirsutus* Green (Hemiptera: Pseudococcidae) (Jacobson & Hara 2003), *P. minor* (Ravuiwasa et al. 2009) and *D. neobrevipes* (Doan et al. 2012), whereas ≤ 200 Gy was reported as an effective phytosanitary treatment dose for the solenopsis mealybug (Huang et al. 2014). Our data to some extent coincide with those of Huang et al. (2014), with respect to mortality, adult formation, and adult reproductive performance. The results indicated that at least 374 Gy would be required to completely sterilize (prevent F_1 egg hatch) when applied to the gravid female of *P. solenopsis* (the most tolerant among all stages). However, the phytosanitary dose could be reduced to around 170 Gy if the measure of efficacy was the prevention of development of F_1 N_1 into F_1 N_2 nymphs. Large scale testing is necessary to confirm the final PI dose.

Acknowledgments

This work was part of the FAO/IAEA Coordinated Research Project D62008 on Development of Generic Irradiation Doses for Quaran-

Table 8. Physiochemical and nutritional parameters of irradiated and non-irradiated potatoes after 7 d of storage at 5 °C. Values within each column were not significant at $P < 0.05$ (ANOVA, n = 5; df = 2,12).

	Quality parameters								
		Color characteristics							
Dose (Gy)	Weight loss (%)	'L' value	'a' value	'b' value	Total soluble solids (%)	Titratable acidity (%)	pH	Vitamin C (mg/100g)	β-carotene (mg/100g)
0	5.2 ± 0.67	51.6 ± 0.15	5.60 ± 0.26	17.6 ± 0.32	6.87 ± 0.33	29 ± 0.50	6.2 ± 0.03	3.0 ± 0.12	0.091 ± 0.008
200	5.3 ± 0.83	51.9 ± 2.03	5.50 ± 0.32	18.3 ± 1.13	6.53 ± 0.17	27 ± 1.2	6.3 ± 0.08	3.1 ± 0.13	0.077 ± 0.009
400	5.9 ± 0.76	51.3 ± 1.69	5.73 ± 0.57	19.0 ± 0.53	6.83 ± 0.09	28 ± 0.9	6.3 ± 0.03	3.1 ± 0.21	0.079 ± 0.007
F-value	0.26	0.99	0.27	3.0	2.37	2.42	1.18	0.34	0.98

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