Study on disinfestation of pulses using microwave technique

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Abstract Mortality of the pulse beetle (*Callosobruchus chinensis* L.) exposed, continuously, to microwave radiation (2450 MHz) was evaluated as a function of exposure time and percent power level, at adult stages. The microwave exposure time to attain 100% insect mortality at 100 %, 80%, 60%, 40%, and 20% power levels for Chickpea, Pigeon Pea and Green Gram was optimized. Effect of optimized microwave exposure time on viability, germination, cooking and milling characteristics of Chickpea, Pigeon Pea and Green Gram was also evaluated. Adult stage study was characterized by a distinct dose–exposure curve. The mortality curve was following third degree polynomial equation. The seed viability and germination of Chickpea, Pigeon Pea and Green Gram was affected by microwave exposure time and power level. It was observed that as the power level is decreasing the germination and viability of all the pulses are increasing. The effect on cooking and milling characteristics are not affected by microwave exposure time and power level. The insects in the mobile state were observed to move towards the surface from inside the nutrient medium during microwave exposure. They also curled up and in some cases aggregation was observed.

Keywords Disinfestation · Microwave · Insect mortality · Germination · Seed viability · Cooking and milling characteristics

Microwaves are very similar to high frequency electric fields, but utilize much higher frequencies, 0.5–3 GHz. This enables rapid heating (72–83 °C) to be achieved with much lower field intensities. However, the penetration depth becomes more

important, as the microwave intensity diminishes with increased penetration. Also, the electrical properties of the pest and host are much closer at these frequencies, which make differential heating less effective. Unlike electric field frequencies (10–100 MHz), the grain is heated close to the same temperature as the insects. The effectiveness also depends upon the moisture content of the grain. A higher moisture content yields higher temperatures and therefore higher mortality for a given treatment time. As the grain is heated, the moisture content of the grain will drop, reducing the rate of temperature increase, and potentially reducing mortality. This is unlikely to be a factor due to the short treatment times (seconds). One hundred percent mortality of confused flour beetle in wheat at 15.6% moisture content was obtained after a 105 s exposure to 8.5 GHz with a pulse frequency of 4 MHz (Watters 1976). As frequency increases grain temperatures must increase to generate complete mortality. In a comparison between a 39 MHz electric field and a 2.45 GHz microwave on the mortality of rice weevils in wheat, the 39 MHz frequency achieved complete mortality at 40 °C wheat temperatures, compared to 80 °C for complete mortality at 2.45 GHz. At 39 MHz, the insects absorb the energy at a greater rate than the wheat. Therefore the temperature of the individual insects within the grain will be greater than the grain bulk, i.e. while the grain bulk is at 40 °C, the insects within the grain may be at 80 °C. Because the rate of energy absorption is similar for the insects and grain at 2.45 GHz, both are heated at the same rate, and would therefore reach the same temperature of 80 °C. This accounts for the difference in bulk grain temperatures for the different treatments. The use of microwave energy to control insects was initiated by Webber et al. (1946), and since then the literature prior to 1980 has been reviewed (Watters 1976; Tilton and Vardell 1982). Insect control and other applications such as grain drying, seed treatment, and moisture content measurements with radiofrequency and microwave

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energy have also been reviewed thoroughly (Nelson 1986). Wilkin and Nelson (1987) recommended 15 min exposures to microwaves, which increased temperature to 60 °C to kill all insects in confectionery walnuts without adversely affecting flavour. However, temperature greater than 80 °C was required to disinfest rice of Lepidoptera and Coleoptera infestations with microwaves (Locatelli and Traversa 1989). Thus, lethal temperatures were found to vary considerably not only with species, but also with the stage of development, acclimation, and relative humidity (Fields 1992).

Although flour and flat grain beetles were shown to be economically controlled with continuous microwave irradiation (Langlinais 1989), no information was available on the use of intermittent energy. Moreover, the generation of ‘hot spots’ during industrial microwave heating has recently become of great concern to scientists involved in microwave research (Hill and Jennings 1993; Zhu et al. 1995). A hot spot can be defined as a local area of very high temperature that results from the temperature dependence of material properties. Although hot spots are desirable to increase insect mortality, detrimental effects on the medium are not. However, our review of the literature did not reveal any entomological concern over hot spot generation during microwave control of insects. Insect behavior in a microwave field has also not been reported. The study was undertaken with the objective to study different optimum microwave exposure time and percent power level combinations for 100 % insect mortality and the affect of optimized microwave exposure time on dal recovery and cooking time characteristics of chickpea, pigeon pea and green gram.

Materials and methods

Insect

The insect *Callosobruchus Chinensis* has been reported from U.S.A. Taiwan, Africa, China, Philippines, Japan, Indonesia & India. It has been reported on pigeonpea, Chickpea,

cottonseed, sorghum and maize. The larva causes the damage by feeding inside the grain. This insect causes very high level of damage to the grain and quite often each and every grain is infested. The female starts laying small, oval and scale like eggs, which are glued to the grain. In this species more than one egg may be laid on a grain. A single female insect may lay 34 to 113 eggs at the rate of 1–37 per day. The highest egg production is in May and October. The egg hatch in 4 to 18 days in different seasons.

Insect culture preparation

A quarter of jar was filled with the pulses. Fifteen to twenty insects of *Callosobruchus chinensis* were placed in every jar and jar was covered with cloths for proper aeration. The jar was held at 27 +5 °C and 60+5% RH in the humidity control chamber for 4 weeks. After 4 weeks insects were removed from the jar and was used in the experiment.

Insect's collector

An apparatus was made for collecting the adults. A 250 ml conical flask was used. Two holes were made on the beaker cork and two 8 mm glass pipes were inserted into them. Two numbers of 8 mm pipe were inserted on the upper glass tube. One of the pipes were placed in the jar and the mouth sucked other pipe. Thus, this apparatus collected insects. Collected insects were used for the experiments on mortality of adults.

The disinfestation procedure

For experiments to assess the mortality of adults, five microwave power output levels were taken. The sample for disinfestation of insects was prepared by adding 15–20 adults of *Callosobruchus chinensis* in 200 g samples each. The microwave oven was set for five-power level i.e (100%, 80%, 60%, 40%, and 20%) with varied exposure time combination. The infested sample was spread and placed in the microwave oven tray for disinfestation after it

Fig. 1 Effect of microwave exposure time on mortality of chickpea

Fig. 2 Effect of microwave exposure time on mortality of pigeon pea

attained the desired microwave radiation. After a pre-determined exposure time, the sample was removed from the tray and allowed to cool in ambient temperature. These microwave-exposed samples were taken for analysis of insect mortality seed germination, seed viability, milling characteristics and cooking characteristics of dal.

Analysis of microwave exposed disinfested pulses

Microwave exposed pulses were analyzed for insect mortality by visual observation. The number of dead and live insects were recorded, and expressed in percent mortality.

Insect Mortality(percent)

$$= \frac{\text{No. of dead in sec ts after microwave exposure}}{\text{Total no. of in sec ts before microwave exposure}} \times 100$$

Seed germination

Seed germination is the ability of seed to germinate under ideal condition. It was done by germinating the number of

seeds inside the laboratory seed germinator. Fifty seeds in three replications were taken at random and were placed in wet muslin cloth. The spacing between seed to seed was kept as five times the width of seed. The relative humidity inside the germinator was maintained close to saturation point while temperature was maintained at 25 °C. After 4–5 days the germinated seeds were counted and germination percentage was calculated as:

$$\text{Germination, \%} = \frac{\text{No. of germinated seeds}}{\text{Total no. of seeds}} \times 100$$

Seed viability

One hundred seeds in three replications were taken at random for viability test. The seeds were soaked in water for 24 h and outer cover of seeds was removed. Cotyledons were dissected longitudinally through embryo in two parts. Then all dissected part was kept in a petri dish and a half was soaked in 1 % tetrazolium chloride solution. To prevent reaction through light, petri dish was covered with piece of paper, and is kept for 4 h. Respiring and viable seeds

Fig. 3 Effect of microwave exposure time on mortality of green gram

Table 1 Effect of optimized microwave exposure time on viability and germination characteristics of chickpea, pigeon pea and green gram

Power W	Temperature, °C	Seed Viability, %			Germination, %		
		Chick pea	Pigeon pea	Green gram	Chick pea	Pigeon pea	Green gram
700	80	57	53	62	54	64	78
560	70	65	59	67	65	73	66
420	65	77	76	78	78	76	82
280	50	86	69	87	87	86	87
140	40	92	94	87	94	92	91
Control		98	97	98	97	98	98
Stat.		S	S	S	S	S	S

S-Significant at 5 % level of significance

changed color to red. Dead and non-respiring seeds were colorless. Percent viability was calculated as follows.

Seed viability, %

$$= \frac{\text{No. of seed part change into red color}}{\text{Total no. of seed parts}} \times 100$$

Milling of microwave treated pulses

The milling study of pulses was performed in rice polisher (Make satake Engineering Co. Ltd. No 553498, Japan). About 200 g of microwave exposed pulses were taken for milling process. The pigeon pea was milled for 5 s and chickpea and green gram were milled for 10 s. The milled mass was separated from broken and husk. The percentage of dhal yield was calculated using following formula.

$$\text{Dal yield, \%} = \frac{\text{dal recovery}}{\text{Total amount of pulses}} \times 100$$

Cooking time of microwave-treated pulses

About 100 ml distill water was brought to the boiling point in a 250 ml beaker. 20 g of microwave exposed and milled

pulses were poured in the boiled water. The sample was stirred at every 2 min interval. After 15 min 2 to 3 dal grains were taken out with the help of spoon without interrupting boiling process. Degree of cooking was tested by pressing boiled dal between two glass slides. Evenly distribution of dal between slides represents complete cooking and subsequently time for cooking was noted.

Specification of Microwave oven

Model: Kelvinator *Megicook*.

Input Volt: 240 Volts AC, 50 Hz.

Input Current: 6 Amperes, AC.

Frequency: 2450 MHz

Power Output: 700 Watts

Statistical analysis

The experiments were conducted in duplicate on three replications of each microwave treatment of pulses. The two-way ANOVA test for the Block design with replication was done with JAVA script based computer program to determine significant differences ($P \leq 0.05$) between treatments and blocks.

Table 2 Effect of microwave exposure time on dal recovery and cooking time characteristics of chickpea, pigeon pea and green gram

Power W	Temperature, °C	Dal recovery, (%)			Cooking time, (min)		
		Chick pea	Pigeon pea	Green gram	Chick pea	Pigeon pea	Green gram
700	80	76.2	72.1	76.2	33.1	21.9	15.2
560	70	72.2	73.7	73.2	34.6	18.3	13.4
420	65	71.4	69.3	76.1	34.5	20.1	12.5
280	50	75.1	72.0	74.4	36.6	20.7	13.3
140	40	75.9	69.0	76.7	42.6	19.2	14.2
Control		73.4	72.8	72.8	39.6	22.3	14.1
Stat.		NS	NS	NS	NS	NS	NS

NS-Non-significant at 5 % level of significance

Results and discussion

Insect mortality of pulses

The mortality curve was following third degree polynomial equation. In case of chickpea, the different optimum microwave exposure time and percent power level combinations for 100 % insect mortality were found as 100 s at 700 W, followed by 160 s at 560 W, 200 s at 420 W, 240 s at 280 W and 300 s at 140 W power levels (Fig. 1). However, in case of pigeon pea, the different optimum microwave exposure time and percent power level combinations for 100 % insect mortality were found as 90 s at 700 W, followed by 140 s at 560 W, 180 s at 420 W, 240 s at 280 W and 280 s at 140 W power levels (Fig. 2). Whereas, in case of green gram, the different optimum microwave exposure time and percent power level combinations for 100 % insect mortality were found as 70 s at 700 W, followed by 120 s at 560 W, 160 s at 420 W, 240 s at 280 W and 300 s at 140 W power levels (Fig. 3).

Viability and germination characteristics of Chickpea, Pigeon pea and Green gram

The effect of optimized microwave exposure time on viability and germination characteristics of chickpea, pigeon pea and green gram were examined. The results are given in Table 1. The seed viability varied between 57–92 % for pigeon pea, 53–94 % for chickpea, and 62–87 % for green gram. In all the cases, as the power level of microwave exposure is decreasing, an increase in seed germination was observed and differs significantly at 5 % level. Similarly, exposure power levels affect germination of the entire three pulses sample. As the power is decreasing, the germination of pulses is increasing and vice-versa. It was also concluded that the microwave power level effect germination quality of all the three pulses sample significantly at 5 % level.

Dal recovery and cooking time characteristics of chickpea, pigeon pea and green gram

The affect of optimized microwave exposure time on dal recovery and cooking time characteristics of chickpea, pigeon pea and green gram were examined. The results are given in Table 2. The dal recovery varied between 76.2–71.4 % for chickpea, 69.0–73.7 % for pigeon pea and 72.8–

76.7 % for green gram. However, the cooking time varied between 33.1–42.6 min for chickpea, 18.3–22.3 min for pigeon pea and 13.3–15.2 min for green gram. In all the cases, the power level of microwave exposure did not affect the dal recovery and cooking time. The power level of microwave exposure on dal recovery and cooking time differ non-significantly at 5 % level.

Conclusions

In case of chickpea, pigeon pea and green gram, it was observed that at certain constant power levels, the mortality of insect's increases as the microwave exposure time elongated and vice-versa. Seed viability and germination of chickpea, pigeon pea and green gram was effected by microwave exposure time and power level at 5% level of significance. It was also concluded that, as the power level is decreasing the germination and seed viability of all the pulses are increasing. The effect on cooking and milling characteristics of all the three pulses are not affected by microwave exposure time and power level.

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