Radio frequency treatments for insect disinfestation of dried legumes

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Abstract

Dried legumes (chickpeas, green peas or lentils) are valuable export commodities in the US Pacific Northwest. A major problem in the marketing of these products is infestation by insect pests. Typically, chemical fumigants are used to disinfest product, but regulatory issues, insect resistance, environmental concerns and the increase of the organic market have forced the industry to explore non-chemical alternatives. One possible alternative is the use of radio frequency (RF) energy to rapidly heat product to insecticidal levels. To determine the potential of RF treatments to control insect pests in dried pulse products, the heat tolerance of the cowpea weevil (*Callosobruchus maculatus* F.) was evaluated and compared to the tolerance of previously studied insects, and the dielectric properties of both the insect and the products were compared. The most heat tolerant stage of the weevil was found to be the pupal stage, with adults being the most susceptible. Cowpea weevil pupae were fairly heat tolerant; to obtain rapid mortality (exposure <10 min) temperatures of 56-58°C were needed. At frequencies commonly used by industry for RF heating, dielectric loss factors for both adult and larval cowpea weevil was higher than those for legumes, suggesting that cowpea weevils would heat at a faster rate than the product. Previous studies showed that suitable heating uniformity during RF treatments was obtained through the addition of hot air (60°C) and conveyor belt movement. These studies showed that chickpeas, green peas and lentils were able to tolerate RF treatments of 60°C for 10 min without adverse effects on quality. The results suggest that practical large scale RF treatments to disinfest pulses may be possible.

Keywords: Heat treatments, Dried pulses, Cowpea weevil, Radio frequency, Disinfestations

1. Introduction

Chickpea (*Cicer arietinum* L.), green pea (*Pisum sativum* L.) and lentil (*Lens culinaris* Medikus) are three important rotational legumes in the western United States. Infestation by postharvest insect pests can be a major problem in the processing and marketing of dried legumes. Of particular economic importance is the cowpea weevil, *Callosobruchus maculatus* F. (Coleoptera: Bruchidae), a serious internal pest of several legume crops. Another pest of concern is the Indianmeal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), a common pest of many stored products. These insects reduce the quality of products through direct feeding damage and contamination of the product with exuviae, webbing and feces. Legumes infested with cowpea weevils and other internal feeders are not often easily detected by external inspection. Regulatory agencies and importers in many countries have therefore established phytosanitary protocols, often including postharvest disinfestation treatments.

Historically, the legume industry has relied on chemical fumigation (methyl bromide or phosphine) for postharvest insect control. In 2004, India imposed a non-tariff barrier requiring all imported legumes to be fumigated with methyl bromide and certified free of bruchids (USADPLC, 2007). However, most phytosanitary uses of methyl bromide within the U.S. were phased out in 2005 by the U.S. Environmental Protection Agency under the Montreal Protocol (UNEP, 2006). In addition, methyl bromide fumigation is less practical at treatment temperatures <5°C, with lower treatment temperatures requiring higher doses or extended exposure times. Legume processing plants and warehouses in the interior northern states of the U.S. have night temperatures below 5°C for more than 6 months each year (USADPLC, 2007). Therefore, there is a need to develop a practical alternative to methyl bromide for control of insect pests in legumes. Any alternative must also have a minimum impact on product quality and environment.

Heat treatment methods using hot air have been investigated extensively as non-chemical alternatives for disinfesting stored commodities. However, it is difficult to accomplish disinfestation using conventional hot air heating methods without causing deleterious effects to product quality (Armstrong, 1994), and the slow rate of heat transfer due to a high resistance of conduction within bulk materials results in extended treatment times (Evans et al., 1983). Low heating rates also may increase the thermotolerance of the targeted insects (Beckett and Morton, 2003) by causing the induction of heat shock proteins in insects (Yin et al., 2006).

Radio frequency (RF) energy offers the possibility of rapidly increasing temperatures within bulk materials. RF energy directly interacts with commodities containing polar molecules and charged ions to generate heat volumetrically, significantly reducing treatment times when compared to conventional heating methods. Many studies have explored the possibility of using RF energy to disinfest insect pests (Hallman and Sharp, 1994; Nelson, 1996). More recent studies demonstrated the potential of RF treatments for industrial disinfestation of in-shell walnuts with acceptable product quality (Wang et al., 2007a, b). The demonstrated ability of RF treatments to disinfest low moisture products suggests this method for potential applications in dried legumes.

The most important considerations in developing heat treatments using RF energy are the thermotolerance of targeted insects and treated products, and the heating uniformity of the product. In RF treatments, heating uniformity is largely a function of the dielectric properties of the product and the design of the treatment. This paper presents information on the thermotolerance of target insects and the dielectric properties of products and target insects. The treatment effect on product quality and heating uniformity of proposed RF treatments will also be discussed.

2. Materials and methods

2.1. Insect thermotolerance

The relative heat tolerance of cowpea weevil stages was determined using a computer-controlled heat block system designed by Washington State University (Ikediala et al., 2000; Wang et al., 2002). Test insects were from a culture of cowpea weevils maintained at the San Joaquin Valley Agricultural Research Center, Parlier CA, on black-eyed peas, *Vigna unguiculata* (L.), at 28 ± 0.5°C and a photoperiod of 14:10 (L:D) h. Cowpea weevils were treated in mung beans, *Vigna radiata* (L.) Wilczek, a small legume that fit within the heat block and provided relatively rapid heat transfer. Adult cowpea weevils were allowed to oviposit on clean mung beans for 24 h. Infested mung beans were held under rearing conditions and treated 2, 7, 15, and 21 d after infestation when developing cowpea weevils were in the egg, young larval, old larval and pupal stage, respectively. All stages were treated at 50°C for 50, 100 and 150 min and 54°C for 7, 14, and 21 min. A second series of tests treated old larvae and pupae at 54°C for 12, 16 and 20 min, and 56°C for 5, 8 and 11 min. All infested mung beans were held for adult emergence at rearing conditions. Emerging adults were counted and treatment survival estimated by comparing emergence from treated beans with emergence from untreated controls. About 100 adult weevils in flat nylon screen bags were also treated at the above temperature/time combinations. Adult mortality was evaluated 24 h after treatment. When adults proved to be much less tolerant than the immature stages, adults were also treated at a series of less extreme temperature-time combinations. For all experiments each treatment was replicated three times.

Preliminary estimates for the thermal tolerance of cowpea weevil were made for both the most tolerant immature cowpea weevil stage (as determined above) and cowpea weevil adults. Thermal death time (TDT) curves based on the observed minimum exposure time that resulted in 100% mortality were compared with those previously determined for Indianmeal moth, red flour beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae) and navel orangeworm, *Amyelois transitella* (Walker) (Lepidoptera: Pyralidae) in Johnson et al. (2003), Johnson et al. (2004), and Wang et al. (2002), respectively.

2.2. Dielectric property measurements

Immature stages (late larvae and pupae) were dissected from black-eyed peas, while adult weevils were collected from laboratory cultures. Both immature stages and adults were killed and stored at -20°C until they could be shipped to Washington State University for measurement. Dielectric measurements of cowpea weevils were made using the open-ended coaxial probe technique with an impedence analyzer (model 4291B, Hewlett-Packard, Santa Clara, CA, USA) over a frequency range of 10-1800 MHz (Wang et al., 2003). Measurements of the dielectric constant (ε ') and loss factor (ε '') were made at 20, 30, 40, 50 and 60°C. Insect and legume moisture contents were determined on wet basis in aluminium moisture dishes in a vacuum oven (ADP-31, Yamato Scientific America Inc., Santa Clara, CA, USA) at 130 ºC for one hour (AOAC, 2002).The dielectric properties were compared to similar values obtained for Indianmeal moth (Wang et al., 2003). Of the four legumes previously studied (chickpea, green pea, lentil and soybean) in Guo et al. (2010), lentils were chosen as a representative legume for comparing dielectric properties.

3. Results

3.1. Insect thermotolerance

The survival of cowpea weevil eggs, young larvae, old larvae and pupae exposed to 50 and 54°C by the heat block method is given in Figure 1. Eggs and young larvae were consistently the least tolerant to the treatment in the heat block, while old larvae and pupae were the most tolerant. Complete mortality occurred in all adult cowpea weevil exposed to the temperature-time treatments in Figure 1.

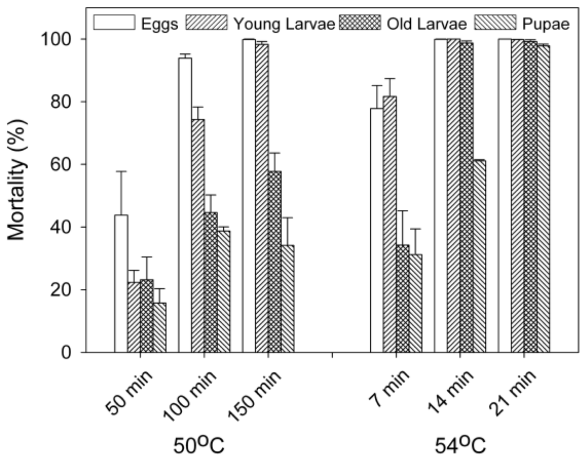

Figure 1 Mortality of immature stages of the cowpea weevil at 50°C for 50, 100 and 150 min, and 54°C for 7, 14 and 21 min.

The results of further tests with old larvae and pupae at 54 and 56°C are given in Figure 2. Mortality of old larvae was consistently higher than that for pupae. Based on these results, pupae were selected as the most tolerant cowpea weevil stage for more detailed thermal death studies. Preliminary results from these studies, given in Figure 3, show that cowpea weevil pupae are quite temperature tolerant when compared to navel orangeworm, Indianmeal moth and red flour beetle. Figure 3 also shows that cowpea weevil adults, the least tolerant of the cowpea weevil stages using the heat block method, were also more tolerant than all the other insects previously tested.

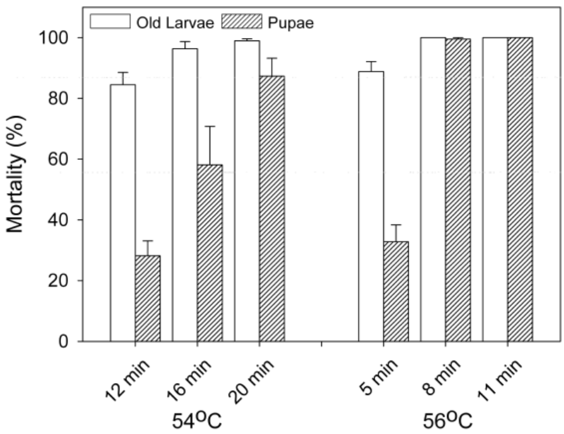

Figure 2 Mortality of cowpea weevil older larvae and pupae at 54°C for 12, 16 and 20 min, and 56°C for 5, 8 and 11 min.

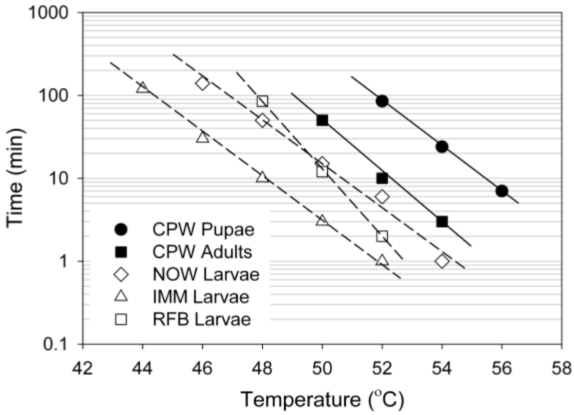

Figure 3 Thermal death time curves for cowpea weevil pupae and adults, navel orangeworm larvae, Indianmeal moth larvae and red flour beetle larvae.

3.2. Dielectric property measurements

Figure 4 shows dielectric constant (ε′) and dielectric loss (ε″) for cowpea weevil immature stages, cowpea weevil adults, Indianmeal moth larvae and lentil (8% m.c.) as a function of frequency at 20°C. The ε′ and ε″ for both cowpea weevil immature stages (larvae and pupae) and adults were comparable to those found for Indianmeal moth larvae (Wang et al., 2003). Both ε′ and ε″ for all insect stages were higher than those for lentil due to the higher moisture (70.8%) of insects. This was consistent for all temperatures tested. Figure 5 shows the effect of temperature on dielectric loss for cowpea weevil immature stages and lentil at two different moisture contents (8 and 22%). The difference between the cowpea weevils and the product increased with increasing temperature. These results suggest that both cowpea weevils and Indianmeal moth would heat at a faster rate than the product.

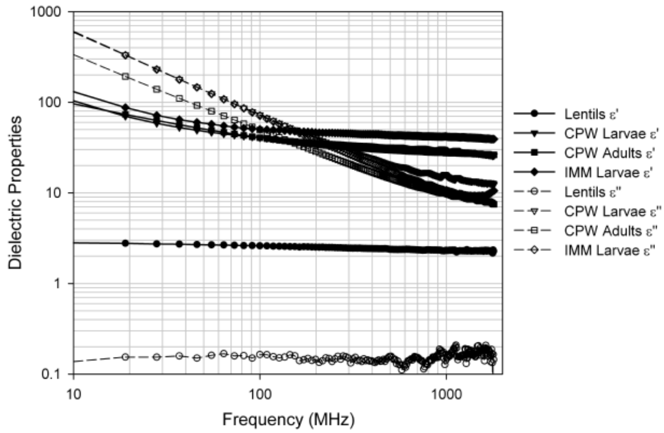

Figure 4 Frequency dependence of the dielectric constant (ε′) and loss (ε″) for cowpea weevil immature stages (large larvae and pupae), adults, Indianmeal moth larvae and lentil (8% m.c.) at 20°C.

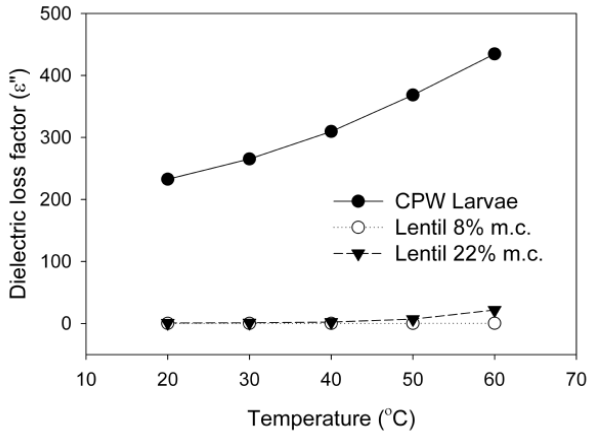

Figure 5 Temperature dependence of dielectric loss (ε″) at 27 MHz for cowpea weevil immature stages (large larvae and pupae) and lentil at 8 and 22% m.c.

4. Discussion

The cowpea weevil is the first internal seed feeding insect to be tested using the heat block system designed by Washington State University. Because of the carefully controlled and rapid heating rates obtained with the heating block system, it has proven a valuable tool in determining thermal death kinetics for several target insect pests in previous radio frequency feasibility studies (Wang et al., 2002; Johnson et al., 2003, 2004). However, there are some difficulties inherent in using this method on an internal seed feeder. Dissecting the necessary number of test insects without causing extreme control mortality was difficult, so test insects were treated within small mung bean to minimize the effect of insulation by the bean. Because test insects were treated in bean, it was difficult to be certain of the exact number of insects and stage being treated, resulting in sometimes variable results.

The current study indicates that the pupal stage is the most heat tolerant stage of the cowpea weevil. However, care must be taken in applying this information. Eggs and adults were the least tolerant stages tested, and both these are external stages. It is difficult to compare these stages directly with the internal larval and pupal stages, due to the insulation of the bean. The heating rate experienced by internal stages is much slower than that experienced by the external stages, but so is the cooling rate, making the estimation of the actual accumulative temperature effect very difficult. Because of this uncertainty, it is recommended that studies to confirm the relative heat tolerance of cowpea weevil stages under RF heating be conducted before large-scale confirmatory studies using industrial RF units are attempted.

Regardless of the relative tolerance of the internal stages, it is clear that the cowpea weevil is heat tolerant when compared to other species, as the least tolerant cowpea weevil stage (adult) was more tolerant than any of the other species tested with the heat block. Protocols for RF treatments will likely need to raise products to 56°C or higher for more than 7 min to obtain adequate control of cowpea weevil, and any such treatment should also control Indianmeal moth.

The comparison of the dielectric properties of the target insects with those of the products indicates that the target insects should heat faster than the product when treated with RF energy. Theoretical differential heating has been reported for several other products (Wang et al., 2003) and was demonstrated experimentally in almond (Wang and Tang, 2007). The occurrence of differential heating improves the likelihood of developing a successful RF treatment protocol, resulting in increased insect mortality at relatively low product temperatures that cause no harm to the product.

Guo et al. (2010) calculated penetration depths of various legumes from their dielectric properties, noting that penetration decreased as frequency increased. It was determined that the penetration depths in 27 MHz industrial RF systems should provide sufficient throughput of product to be practical. Wang et al. (2010) suggested an RF treatment protocol for legumes using 27 MHz RF to rapidly heat product in a 10-cm-deep bed to 60°C, then holding the product at 60°C for 10 min through the application of hot forced air. Afterwards, the product would be reduced to a 1-cm-deep bed to allow for rapid cooling through forced ambient air. In this treatment, the heating uniformity of the product was maximized by the addition of 60°C forced hot air and movement along a conveyor belt. Mixing of the product between successive RF treatments did not improve heating uniformity, as had been found with RF treatments of inshell walnut (Wang et al., 2007a). The results of the current study indicate that this treatment should be sufficient to control cowpea weevil and Indianmeal moth, but additional tests are needed to confirm the treatment protocol.

The effect of the proposed treatment protocol on product quality was also examined by Wang et al. (2010). No significant differences in weight loss, moisture content, color or germination of chickpea, green pea or lentil were observed between RF treatments and unheated controls. RF treatments, therefore, should provide a practical, effective and environmentally friendly method for disinfestation of postharvest legumes while maintaining product quality.

❖ This paper represents the results of research only. Mention of a proprietary product or trade name does not constitute a recommendation or endorsement by the US Department of Agriculture.

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