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Radio frequency disinfestation of black gram: effect on grain's viability and physico-chemical composition

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Abstract
Radio frequency (RF) has gained popularity in recent years as an effective mode for disinfestation and safe storage of food grains and other perishables. Apart from providing better heating uniformity, it also has an edge on the disinfestation because of its differential heating ability, but still there's a lack of data depicting towards its effect on the germination characteristics of the disinfested grains. The disinfested grain samples with different RF levels (180, 190, 200 and 210 mm) providing complete mortality were considered for the germination studies and to optimize the process accordingly. RF disinfestation at 180 mm electrode level proved to affect the most as least germination energy, around 13%, was observed depicting to loss in viability for most of the grains. Apparently, the disinfestation at 210mm, which was having the least electric field density subjected to the sample, had its viability maintained, with germination energy of around 90%. The temperature of the sample in the former varied from 54 to 60 °C, where as in the later it was observed to be around 45 to 48 °C. This increase in the temperature of the black gram grains may be considered as the reason for the loss in viability. This opens up a wide array of unreported facts which needs to be studied as a part of effect laid down by dielectric heating on the food materials.

Keywords: Dielectric heating, germination, heating uniformity, vigor, legumes

1. Introduction

Indian sub-continent is self-sufficient in growing food crops in order to feed the entire population. The annual production of legumes is over 25.23 MT in the year 2018 (Varghese *et al*., 2019). Still there's a lag that can be observed in the availability to the end user, the consumer. This is mainly due to the prevailing post-harvest losses in the country. It's clear indication towards the need to enhance proper infrastructure for storage and protection of harvested food grains.

Among the post-harvest losses, insect infestation accounts to about 23 per cent of the total losses during storage of food grains (Kumar and Kalita, 2017) [11]. Various chemical methods were conventionally used to prevent the insects from entering the warehouses and to even to enhance the mortality with small doses. But, due to the effect on environment, residues can cause depletion of the ozone layer, due to this all the major insecticides were phased out for circulation by the Montreal Protocol (Porter *et al*., 2010) [14]. There onwards physical methods for the control of insect pests were the prime focus of the researchers working on integrated pest management.

Low and high temperature storage, thermal treatments, inert dust and Diatomaceous earth (D.E.) have been extensively studied for disinfestation, but still there exist some drawbacks for bulk application (Arthur, 2000; Loganathan *et al*., 2011; Subramanyam and Roesli, 2000; Tang *et al*., 2000) [3, 12, 16, 17]. Recent advancements in microwave and radio frequency (RF) treatments have been utilized to cope up with the limitations of the earlier stated techniques (Alfaifi *et al*., 2013; Gao *et al*., 2010; Jiao *et al*., 2017; Wang *et al*., 2010) [2, 7, 10, 7, 8, 20]. The RF disinfestation is principally based on the dielectric heating properties, which causes volumetric heating of the food grains causing a rapid increase in temperature, thus inactivating the insects (Dalmoro *et al*., 2015; Das *et al*., 2013; Vadivambal *et al*., 2007; Yadav *et al*., 2014) [4, 5, 18, 22]. But, there might be some changes in the properties of the food grains too which are not studied extensively till date. It has been reported that microwave has a stimulating effect on the viability and germination capacity of lentils but no data has been submitted regarding the effect of radio frequency on the same.

The present research study aims to provide information regarding the effect of radio frequency disinfestation on the germination capacity and viability of black gram (*Vigna mungo*) grains after its disinfestation.

2. Materials and methods

2.1. Legume sample

Black gram was procured from local market of Thanjavur, Tamil Nadu for the study. These were then sorted and graded accordingly in order to have uniform size and to be free from any foreign (unwanted) materials.

2.2. Radio frequency (RF) Sterilizer

A 10 kW RF sterilizer (40.68 MHz; Make: Lakshmi industries, Coimbatore) was used to treat the black gram grains at various load densities which can be modulated by changing the electrode height of the equipment. It is equipped with a conveyer which can ease to move the sample under the electrodes. The sterilizer can have a minimum of 180mm to a maximum to 300mm of gap between the electrode and the conveyer which can be changed depending on the height of the sample.

2.3. *Callasobruchus maculatus* adults

Progenies of *C. maculatus* were taken from stock culture maintained at insect culture lab at IIFPT, Thanjavur. Twenty pairs were released in an insect cage with black gram (kept in petri dish) for the purpose of egg laying. It was monitored for 28 ± 3 days at 28 ± 0.5 °C for emergence of new adults in order to have insects with uniform age for the ease of disinfestation experiment (Fatima *et al*., 2016; Sewsaran *et al*., 2019) [6, 15].

2.4. RF disinfestation treatment

The samples (1Kg) were packed in cloth bag and treated at four electrode gaps of heights 180, 190, 200, 210 mm at a belt speed of 10 m/hr. this will allow the electric field to treat the sample uniformly and the grains will be subjected to same density of the electric field. The bags were then then checked after 24 hours of treatment for assessing the mortality caused.

2.5. Physiochemical properties

Black gram grains were analysed for thousand kernel weight, volume and bulk density (Wani *et al*., 2013) [21]. Protein, ash, fat, fibre and moisture content were also determined using standard AOAC (1990) methods.

2.6. Germination study

Germination test was conducted after each treatment to test effect on viability of the grains. The analysis was performed at a temperature 25 ± 2 °C and the observations were taken after 2nd and 5th day after treatment. Grains were kept on Whatmann filter paper no. 1in 9cm diameter petri dishes. Approximately 4 ml of distilled water was poured into each petri dish and were covered. For each treatment, 25 black gram grains were used and the test was conducted in triplicate. After accessing the germination values, germination energy was calculated as the number of grains germinated after two days divide by total number of grains per replicate (Maneemegalai and Nandakumar, 2011) [13].

2.7. Statistical analysis

The observed data was analysed using Fischer's LSD test to check the significance between the treatment means and the significant level was reported at $p<0.05$ using IBM SPSS (Version 25.0).

3. Results and discussion

3.1. Mortality assessment

The experiments were conducted based on the preliminary studies on the disinfestation. It was found that, four combinations of electrode gap and time were effective against the control of pulse beetle adults. At all these levels complete mortality was achieved (Table 1). It was observed that there was an increase in time required for mortality with increase in electrode height. Similar results were stated by Jiao *et al*. (2012) in the case of lentils in order to develop an industrial scale treatment for the same. In some other studies related to microwave disinfestation, the sample size was comparatively smaller than the present study, that was the sole reason for their treatment time to be less as microwave will be having less penetration depth in comparison to RF.

Table 1: Different level RF treatment for complete mortality of *C. maculatus* adults

Sample	Electrode Gap (mm)	Treatment time (min)	Mortality (%)
A	180	4.27 ± 0.20	100
B	190	5.33 ± 0.25	100
C	200	6.17 ± 0.24	100
D	210	7.58 ± 0.11	100

Values are depicted as mean ± SD; n=3

3.2. Physicochemical analysis

The RF treated and the untreated black gram grains were subjected to physicochemical analysis for crude protein, crude fat, ash content, crude fibre and moisture content. Thousand kernel weight, volume and bulk density of the grains were also reported for all the treatments (Table 2). The moisture content of untreated sample was 11 per cent whereas it decreased to 10.2, 9.6, 8.6 and 7.7 in the RF treated samples at different treatment levels (Fig. 1). The crude protein, crude fat, ash content and carbohydrate ranged from 27.1 to 27.6, 5.2 to 5.6, 6.2 to 6.4 and 50.4 to 52.9, respectively. There was a significant change in the composition of the treated samples. This might be because of the reduction in moisture content after the treatment. Whereas, the moisture loss followed a trend which supported the results reported by Guo *et al*. (2010) [8] after subjecting to this form of dielectric heating.

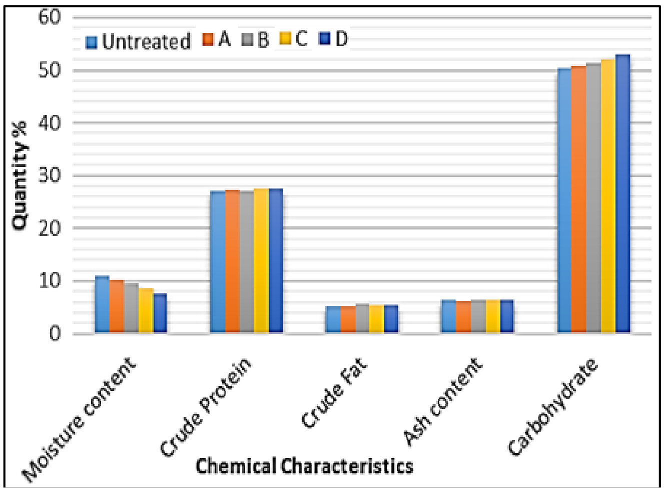

Fig 1: Chemical composition of black gram

Table 2: Physical characteristics of black gram

Sample	Thousand Kernel Weight (g/1000 grain)	Grain Volume (ml/1000 grain)	Bulk Density
Untreated	35.6 ± 2.24[a]	40.3 ± 2.5[b]	0.90 ± 0.03[b]
A	34.8 ± 0.50[a]	40.6 ± 2.5[ab]	0.06 ± 0.06[e]
B	34.2 ± .3.23[a]	42.1 ± 2.6[ab]	1.04 ± 0.01[a]
C	33.1 ± 2.50[a]	42.3 ± 2.0[ab]	0.79 ± 0.01[d]
D	32.2 ± 2.33[a]	43.1 ± 1.0[a]	0.80 ± 0.02[c]

Values in the column are mean ± SD; n=3; mean with different superscripts differ significantly

3.3. Germination energy

The germination energy of the treated and untreated black gram grains were calculated based on germination of grains. It was observed that there was no significant change in the germination energy in the treatment D when compared to the control. There was a notable change in the germination energy for treatment A, B and C. This might be due to the long exposure of the grains to highly dense electric field which in turn increase the temperature of the grains due to volumetric heating making it loose its germination capability.

Table 3: Effect of RF Treatment on germination energy of black gram grains

Sample	Germination Energy
Untreated	99.3±1.67[a]
A	12.5±2.5[d]
B	46.5±5.3[c]
C	75.0±8.7[b]
D	90.0±5.0[a]

Values in the column are mean ± SD; n=3; mean with different superscripts differ significantly

Vadivambal *et al*. (2007) [18] defined microwave energy lethal for the germination characteristics of the wheat at a power level of 500 W when exposed for 28 sec for a sample size of 50 g. The present study also depicts similar trend at three of the treatment combinations but also points towards the decrease in the density of electric field to be vital for maintaining the germination power of the black gram grains.

A study related to pre-soaked lentils was reported by Aladjadjiyan (2010) [1], microwave treatment at 450W for 30secs enhanced the germination and vigor which indicates towards the difference in absorbance of energy due to which the grains were not affected in the water. But the present study is aimed for the disinfestation of black gram which cannot be done with water so decreasing the electric field density by increasing the electrode gap is the solution for the same.

4. Conclusion

In order to maintain the quality of the food grain, RF can prove vital with its volumetric heating concept. The present study helps us to optimize the effect of RF on the viability of the grains and this calls for conducting extensive studies related to changes in amino acids present in the legumes which are essential for human health. Radio frequency can thus be helpful in minimizing the post-harvest losses by disinfestation and moderating the moisture content if found high before considering a food grain for storage. Radio frequency treatment being one of the potent mode of disinfestation, holds scope to be considered as a quarantine method.

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