Gamma irradiation effects on physical properties of squash seeds

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Abstract: In order to study the effects of gamma radiation on some physical properties of squash (*Cucurbit pepo*. L) seed, five irradiation doses (25, 50, 75, 100 and 200 GY) have been used. Some physical properties, including dimensional properties (length, width, thickness, geometric mean diameter, sphericity, volume, surface area, projected area, flakiness ratio and elongation ratio), mass, 1,000 seeds mass, bulk density, true density and porosity of gamma irradiated squash seeds were measured. Statistical indices including maximum, minimum, average, variance, skewness and kurtosis, for dimensional properties and mass of the seeds were calculated. Results revealed a significant raise in hollow seeds number by increasing gamma irradiation dose from 5% for 25 GY to nearly 100% for 100 and 200 GY. On the other hand, length, width, thickness, mass of single seed, 1,000 seeds mass and porosity showed an increase followed by a decrease with the increasing gamma irradiation dose. With the increasing gamma irradiation dose, true and bulk densities were found to decrease from 338.41 kg m⁻³ to 214.01 kg m⁻³ and 420.16 kg m⁻³ to 256.12 kg m⁻³, respectively. In 100 and 200 GY all seeds were hollow and very small, therefore dimensions and mass of these seeds were not measured.

Keywords: gravimetric properties, dimensional properties, squash seeds, irradiation, gamma ray

Citation: Ebrahimzadeh, H., A. H. Mirzabe, M. Lotfi, and S. Azizinia. 2013. Gamma irradiation effects on physical properties of squash seeds. Agric Eng Int: CIGR Journal, 15(1): 131–138.

1 Introduction

In recent years, there has been a growing interest in having safe and natural food, including oils and fats. Over the last few years, many nontraditional vegetable oils produced by mechanical extraction without use of any solvent have emerged and become available to consumers. These oils are obtained from different oilseed fruits. Squash seed oil has an outstanding place in the group of the so-called seed oils (Vujasinovic et al., 2010). Squash seed oil belongs to the group of oils of high nutritive value due to its favorable fatty acid composition and different minor components which have certain beneficial effects on the human organism (Fruhwirth and Hermetter, 2008).

Haploidization techniques facilitate the production of

pure lines from heterozygous plants in a single generation and represent significant advantages for plant breeders and geneticists. Haploid plants can also be obtained spontaneously, but the frequency of this event is very low (Kurtar et al., 2002). At present, haploid plants are produced using in vitro androgenesis (anther-microspore culture) and ontogenesis (ovule-ovary culture), and institute parthenogenesis (pollen irradiation and sample with chemicals, etc.).

Pollen irradiation (*UV*, gamma rays, and *X*-rays) is the most widely used technique to induce institute parthenogenetic haploid plants. Gamma rays are commonly used in haploid production programmes because of their simple application, good penetration, reproducibility, high mutation frequency, and low disposal (lethal) problems (Chahal and Gosal 2002). Irradiated pollen can germinate on the stigma, grow within the style and reach the embryo sac, but cannot fertilize the egg-cell and the polar nuclei. Genetically inactive but germinable pollen can be used to stimulate

Received date: 2012-10-08 Accepted date: 2012-12-30

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the division of the egg cell, and thus induce parthenogenesis or development of parthenocarpic fruit (Kurtar and Balkaya 2010). The irradiated pollen technique is an effective method for the induction of haploid embryos in Cucurbit (Kurtar and Balkaya 2010). Influence of gamma irradiation on pollen viability, germinability and fruit and seed-set of pumpkin and winter squash were studied by Kurtar (2009). Production of haploid and doubled haploid plants of melon (*Cucumis melo* L.) for use in breeding for multiple virus resistance, were investigated by Lotfi et al. (2003).

In order to design and modify equipment of planting, harvesting and post harvesting of haploid production and also for optimum performance of planting, conveying, sorting, sizing, oil extraction machines and other processes, physical and mechanical properties of haploid squash seed must be known. A lot of researches have been conducted on physical and mechanical properties of agricultural seeds. Moisture dependent physical properties of the almond nut and kernel were measured by Aydin (2003). Fracture resistance of sunflower seed and kernel to compressive loading was calculated by Gupta and Das (2000). Altuntas et al. (2005) measured some physical properties such as length, width, thickness, geometric mean diameter, sphericity, mass, porosity, and bulk density of fenugreek seeds. Tabarsa et al. (2011) studied Mechanical and physical properties of wheat straw boards bonded with tannin modified phenolformaldehyde adhesive. But a limited number of researches have been conducted on the physical properties of squash seeds. Moisture dependent physical properties of cucurbit seeds were examined by Milani et al. (2007). Some physical properties of pumpkin seeds (Cucurbita maxima) were investigated by Joshi et al. (1993). Some physical properties of squash seeds at different moisture content were estimated and the role of moisture content was also studied by Paksoy and Aydin (2004).

The main objective of present work is to identify gamma radiation influences on physical properties of squash seeds such as length, width, thickness, mass, geometric mean diameter, sphericity, surface area, volume, projected area, flakiness ratio, elongation ratio, 1,000 seeds mass, bulk density, true density and porosity.

2 Materials and method

2.1 Plant material

The experiment was carried out on squash (*Cucurbita pepo* L.). The seeds were bought from the Nickerson-Zwaan company of Netherlands. The seeds were sown in plastic flats (cell volume 150 cm³ and 50 cells per flat) containing a mixture of peat-moss: perlite (2:1 v/v) on April 4th, 2011. Seedlings were raised in unheated glasshouse, and 15 seedlings from each genotype were planted at 3-4 true leaf stage with spacing of 150×80 cm on April 24th.

2.2 Irradiation conditions

Female flowers were isolated with paper bags (150×100 mm) and male flower buds were collected one day before anthesis: they were placed in Petri-dishes (100 mm in diameter, 20 mm in height) after the removal of the petals (Figure 1). Then they were gamma irradiated at 25, 50, 75, 100 and 200 GY using a Cobalt-60 gamma-ray sources at the Institute of Agriculture and Nuclear Medicine of Iran (Figure 2).



Figure 1 Extracted anthers for irradiation



Figure 2 Gamma cell unit used to irradiation

The experiments were performed at different durations (May 23rd, Jun. 10th and 13th) to evaluate the effects of irradiation duration on physical and mechanical properties of seeds. Irradiated anthers were then incubated at room temperature overnight. Female flowers were pollinated by irradiated pollen in the morning of the following day at 8:00 to 10:00. Pollinated flowers were isolated to avoid undesired pollen contamination. Paper bags were removed at the second or third day after pollination.

2.3 Properties

2.3.1 Dimensional properties

The seeds were cleaned manually to remove externals along with broken seeds. To determine the average seed size, 100 squash seeds were selected randomly from the bulked sample. For each single seed, three principal perpendicular dimensions including length (L), width (W) and thickness (T) were measured using a digital caliper to an accuracy of 0.01 mm.

The geometric mean diameter (D_g) and sphericity (φ) of the seeds were determined using the following equations given by Perez et al. (2007); Dash et al. (2008); Gholami et al. (2012) and Mirzabe et al. (2012). *L* is the length, *W* is the width and *T* is the thickness in mm in the following Equation (1) and Equation (2):

$$D_g = \sqrt[3]{LWT} \tag{1}$$

$$\varphi = \left(\frac{\sqrt[3]{LWT}}{L}\right) \times 100 \tag{2}$$

The volume of the squash seeds was calculated by the relationship (Equation (3)) given by Burubia et al. (2007), Khazaei et al. (2006) and Sadeghi et al (2010):

$$V = \left(\frac{\pi D_g^3}{6}\right) \tag{3}$$

The surface area (S, Equation (4)) in mm² of seeds was measured by obtaining the value sphere of its geometric mean diameter (Milani et al., 2007; Koocheki et al., 2007; Fathollahzadeh et al., 2009):

$$S = \pi D_{\sigma}^{3} \tag{4}$$

The projected area (A_p) is an important parameter for determining aerodynamic properties. This parameter was calculated based on Equation (5) (Khazaei et al., 2008; Mirzabe et al., 2012):

$$A_p = \left(\frac{\pi WL}{4}\right) \tag{5}$$

The flakiness ratio (F_r) and elongation ratio (E_r) of single squash seed were determined using the following Equation (6) and Equation (7) (Mora and Kwan, 2000; Khazaei et al., 2008):

$$F_r = \frac{T}{W} \tag{6}$$

$$E_r = \frac{L}{W} \tag{7}$$

The skewness and kurtosis were calculated using the following Equation (8) and Equation (9) cited by Lucian, (2006) and Khazaei et al. (2008):

$$Skewness = \frac{n}{(n-1)(n-2)} \sum_{i=1}^{n} \left(\frac{x_i - x_{avg}}{s} \right)^3$$
(8)
$$Kurtosis = \left\{ \frac{n(n-1)}{(n-1)(n-2)(n-3)} \sum_{i=1}^{n} \left(\frac{x_i - x_{avg}}{s} \right)^4 \right\} - \frac{3(n-1)^2}{(n-2)(n-3)}$$
(9)

where, n = number of occurrence; s = standard deviation; $x_{avg} =$ mean seed size; $x_i =$ midpoint of each class interval in metric.

2.3.2 Gravimetric properties

2.3.2.1 Mass properties

In order to determine the mass (*M*) of the seeds, 100 seeds were selected randomly. Each seed was weighed by a digital balance reading to 0.0001 g. For calculating 1000 seeds mass (M_{1000}), 250 seeds were selected from bulk sample randomly; then seeds were divided randomly to five bins so that in each bin 50 seeds were placed. The weight of each bin was measured. Then the average weight value of bins was calculated and then multiplied by 20 to give one thousand seeds mass.

The fraction by mass of kernel and hull in a seed is the ratio of the average mass of seed part, to the average mass of seed. In order to calculate the kernel and hull fraction for each dose, 100 seeds were selected randomly and mass of the 100 seeds were measured. Each seed was hulled manually. Then mass of the 100 kernels and 100 hulls were measured. Average mass of the seeds, kernels and hulls were calculated. Finally ratio of the average mass of kernel and hull, to the average mass of seed, were calculated.

Moisture content of squash seeds for doses of 25, 50, 75, 100, 200 and control sample were found to be 9.60, 10.60, 9.50, 9.77, 9.65 and 10.70% (w.b.), respectively.

2.3.2.2 Densities

The bulk density (ρ_b) of the squash seeds was determined using the standard test weight procedure (Dash et al., 2008) by filling a plastic cylinder of 500 mL with the seed from a height of 150 mm at a constant rate and then weighing the content.

The true density (ρ_t) of seeds was determined using the water displacement method. To avoid water absorption by seeds, Toluene was used. The porosity of squash seed was calculated from bulk and true densities using Equation (10) cited by Isaac Bamgboye and Adejumo (2009) as follows:

$$\varepsilon = \left(1 - \frac{\rho_b}{\rho_t}\right) \times 100 \tag{10}$$

where, ε is porosity; ρ_b is bulk density; ρ_t is true density.

For all measurement of bulk density and true density, moisture content of the squash seeds under different doses of gamma irradiated ranged from 5.55 to 6.36% (w.b.). Moisture content of squash seeds for doses of 25, 50, 75, 100, 200 and control sample were found to be 5.60, 5.68, 5.55, 6.20, 6.1 and 6.36% (w.b.), respectively.

3 Results and discussion

3.1 Dimensional properties

Mean values for both measured and calculated variables including length, width, thickness, mass, and other physical properties of the squash seeds in control sample are presented in Table 1. The length, width, thickness and mass of seeds ranged from 14.71 mm to 17.41 mm, 8.47 mm to 10.82 mm, 1.61 to 3.36 mm and 0.0251 to 0.1919 g, respectively. 1,000 seeds mass was found to be 137 g. In control sample, the seed is hollow if the mass of the seed was less than 0.08 g. Therefore 5% of seeds were hollow.

Values of length, width, thickness and mass of the squash seeds irradiated using dose 25 ranged from 16.32 to 19.5 mm, 10.19 to 17.50 mm, 1.74 to 3.76 mm and 0.0265 to 0.3022 g respectively (Table 2). The

corresponding values for dose 50 were found to be 15.25 to 19.15 mm, 9.36 to 11.75 mm, 1.13 to 3.41 mm, and 0.0183 to 0.2716 g respectively (Table 3).

 Table 1 Dimensional properties and mass of squash seeds for control sample

Parameters	Max/mm	Min/mm	Mean/mm	Variance	Skewness	Kurtosis
L / mm	17.41	14.71	15.918	0.354	0.097	-0.360
W/mm	10.82	8.47	9.629	0.161	0.072	0.263
T/mm	3.36	1.61	2.415	0.061	0.032	2.066
M/gram	0.192	0.025	0.137	0.001	-1.686	3.650
D_g/mm	8.111	6.079	7.170	0.075	-0.133	2.763
arphi / %	50.918	40.365	45.083	4.074	0.029	0.220
V/mm^3	279.406	117.147	193.793	487.969	0.373	2.514
S / mm^2	206.683	115.780	161.715	151.111	0.126	2.512
A_p/mm^2	133.411	55.935	92.532	111.250	0.373	2.514
F_r	0.337	0.174	0.251	0.001	-0.159	0.772
E_r	1.800	1.480	1.655	0.005	-0.229	-0.672

Table 2Dimensional properties and mass of squash seedsusing irradiation under dose 25

Parameters	Max/mm	Min/mm	Mean/mm	Variance	Skewness	Kurtosis
L / mm	19.50	16.32	18.237	0.460	-0.63	0.556
W/ mm	17.50	10.19	11.848	0.586	3.867	29.544
T/mm	3.76	1.74	2.653	0.130	-0.225	0.747
M/g	0.302	0.026	0.213	0.004	-1.010	0.156
D_g/mm	9.926	6.832	8.288	0.250	-0.195	1.361
arphi / %	52.574	39.456	45.458	5.705	0.112	0.821
V/mm^3	512.056	166.987	301.331	2906.225	0.42	2.212
S/mm^2	309.522	146.645	216.589	671.794	0.105	1.642
A_p/mm^2	244.496	79.733	143.879	662.579	0.423	2.212
F_r	0.313	0.146	0.224	0.001	-0.068	0.636
E_r	1.706	1.079	1.543	0.006	-2.007	10.670

 Table 3 Dimensional properties and mass of squash seeds

 using irradiation under dose 50

Parameters	Max/mm	Min/mm	Mean/mm	Variance	Skewness	Kurtosis
<i>L</i> / mm	19.15	15.25	17.520	0.513	-0.220	0.090
W/mm	11.75	9.36	10.877	0.215	-0.571	0.224
T/mm	3.41	1.13	2.589	0.161	-0.524	1.011
M/g	0.272	0.018	0.175	0.005	-0.929	-0.403
D_g/mm	8.721	5.838	7.880	0.256	-1.125	2.984
arphi / %	50.189	32.454	44.992	6.524	-1.240	5.067
V/mm^3	347.329	104.202	259.213	2166.963	-0.494	0.844
S / mm^2	238.950	107.086	195.846	585.523	-0.784	1.709
A_p / mm^2	165.842	49.754	123.769	494.037	-0.494	0.844
F_r	0.318	0.115	0.238	0.001	-0.123	0.255
E_r	1.892	1.431	1.613	0.007	0.776	0.999

For irradiated seeds using dose 25, the seed was hollow if the mass of the seed was less than 0.11 g. Therefore 12.5% of seeds were hollow. The corresponding values for dose 50 were found to be 0.87 g and 16.5%, respectively.

Table 4 shows that, for irradiated seeds using doses 75 GY, value of length, width, thickness and mass of seeds ranged from 13.2 mm to 18.82 mm, 6.54 mm to 11.01 mm, 0.220 to 2.61 mm and 0.0092 to 0.2072 g respectively. For irradiated seeds using dose 75 GY, the seed was hollow if the mass of the seed was less than 0.077 g and 73% of seeds were hollow.

Table 4Dimensional properties and mass of squash seedsusing irradiation under dose 75

Parameters	Max/mm	Min/mm	Mean/mm	Variance	Skewness	Kurtosis
L / mm	18.82	13.2	16.867	1.389	-0.967	0.769
W/mm	11.01	6.54	9.966	0.648	-2.033	5.285
T / mm	2.61	0.22	1.621	0.325	-0.605	-0.285
M/g	0.207	0.009	0.064	0.003	1.111	-0.080
D_g/mm	7.877	2.932	6.387	1.204	-1.1848	1.020
arphi / %	45.257	19.486	37.734	28.564	-1.401	1.661
V/mm^3	255.856	13.193	147.562	3649.440	-0.417	-0.568
S / mm^2	194.899	27.000	131.884	1593.740	-0.762	-0.070
A_p / mm^2	122.166	6.299	70.458	832.022	-0.417	-0.568
F_r	0.253	0.025	0.160	0.003	-0.594	-0.206
E_r	2.243	1.325	1.699	0.018	0.476	2.679

Most of the seeds irradiated using dose 100 and 200 GY were hollow and very small; therefore, dimensions of these seeds cannot be measured. Irradiated seeds using doses 100 and 200 GY of gamma ray are shown in Figure 3.



a. Dose 100 GY

Figure 3 Irradiated seeds using gamma ray

b. Doses 200 GY

A comparison between the average of three dimensions and mass of seeds for control and irradiated seeds indicated that, average dimensions of irradiated seeds under 25 GY were greater than the dimensions of control sample and doses 50 and 75 GY. Dimensions and mass of the irradiated seeds under 50 GY were greater than the corresponding values of control. For seeds irradiated using dose 75 GY, mass of the seeds was less than the control sample, but dimensions of the seeds were greater than the dimensions of control sample.

Maximum values of flakiness ratio and elongation ratio for control sample and dose 75 GY were to be found, respectively. Minimum values of flakiness ratio and elongation ratio for dose 75 and 25 GY were to be found, respectively. Value of flakiness ratio for control sample more than the other cases, were to be found. Value of elongation ratio for control sample more than dose 25 and 50 GY, were to be found.

3.2 Gravimetric properties

A comparison between mass of the control and irradiated seeds indicated that the number of hollow seeds increased using gamma irradiation. The number of hollow seeds increased with the increasing dose of irradiation. A slight decrease in the percentage of hollow seeds was seen between control and 25 GY dose samples and then an obvious raise was shown to over 70% in 75 GY (Figure 4).



Figure 4 Effect of the gamma irradiation on number of hollow seeds

Also mass measurement indicated that, there was an increase in 1,000 seeds mass 25 GY from nearly 150 to 200 g in comparison to control and it starts to dramatically fall by increasing gamma irradiation doses to 75 GY (Figure 5). The variation of 1000 seeds mass was found to be exponential with the gamma irradiation and can be expressed as follows.



Figure 5 Effect of the gamma irradiation on one thousand seeds mass

For irradiated seeds using doses 100 and 200 GY, 100 seeds were selected randomly. Mass of each seed was measured. For irradiated seeds using dose 100 GY, mass of seeds ranged from 0.006 to 0.058 g. The seed was hollow if the mass of the seed was less than 0.052 g and 98% of seeds were hollow. For irradiated seeds using dose 200 GY, mass of seeds ranged from 0.003 to 0.028 g and all seeds were hollow.

Results of kernel and hull fraction are shown in Table 5. Results indicated that, maximum value of kernel fraction were to be found in dose 25 GY. Values of kernel fraction for dose 25 GY and dose 50 GY were more than the control sample. Value of hull fraction for doses 200 GY was more than the other samples. Values of hull fraction for dose 25 GY was less than the other samples.

 Table 5
 Kernel and hull mass fraction for control and irradiated seeds using different doses

Dose/GY	Seed mass/g	Kernel mass/g	Hull mass/g	Kernel fraction	Hull fraction
Control	0.1390	0.0946	0.0444	68.0576	31.9424
25	0.2120	0.1532	0.0588	72.2642	27.7358
50	0.1770	0.1220	0.0550	68.9266	31.0734
75	0.0650	0.0104	0.0546	16.0000	84.0000
100	0.0350	0.0004	0.0346	1.1429	98.8571
200	0.0180	0	0.0180	0	100.0000

The experimental results of the bulk and true densities for squash seed at different irradiation levels are presented in Table 6. It is clear that by increasing radiation level from 0 to 75 GY, the bulk and true densities decreased from 338.41 kg m⁻³ to 214.01 kg m⁻³ and 420.16 kg m⁻³ to 256.12 kg m⁻³, respectively.

Results showed that, porosity of 25 GY, was found to be greater than the other doses. The increase in value of porosity of 25 GY may have resulted from increase in seed size in this radiation level which gives rise to porosity. The same conclusion can be made for 75 GY which has seeds with the lowest size and therefore the less porosity.

 Table 6
 Bulk density, true density and porosity of irradiated seeds in different doses of gamma ray

Dose of Gamma ray	$ ho_b/\mathrm{kg}\mathrm{m}^{-3}$	$ ho_t/\mathrm{kg}~\mathrm{m}^{-3}$	Porosity/%	
0	338.410	420.155	19.456	
25	317.354	416.666	23.835	
50	303.558	386.920	21.545	
75	214.014	256.120	16.440	

Difference between the true and bulk density of dose 75 GY and true and bulk density of other cases very great; because of large number of hollow seed produced in this dose. Also bulk and true density of control sample were more than the other cases.

Bulk density of dose 100 and 200 GY were measured. Values of bulk density were found to be 12.48 kg m⁻³ and 8.05 kg m⁻³, respectively.

4 Conclusions

In sum, the results of the present study can be summarized as follows:

1) The average values of width, thickness, mass of single seed and 1,000 seeds mass of dose 25 GY are greater than the other doses and increased with the increase in radiation level;

2) Bulk density and true density of control is greater than the other cases;

3) Porosity decreased with the increase in radiation level;

4) There was an obvious increase in the number of hollow seeds by increasing gamma irradiation doses;

5) In 100 and 200 GY doses the seeds are very small, for doses 100 and 200 GY, 98% and 100% seeds were hollow.

Acknowledgement

The authors would like to thank the University of

Tehran and Institute of Agriculture and Nuclear Medicine of Iran for providing technical support for this work. We would also like to thank Mr. Mohammad Hassan Torabi Ziaratgahi, Dr. Mohammad Hossein Kianmehr, Mr. Ebrahim Sharifat, Mr. Mostafa Kabiri, Mr. Mohammad Hossein Amir Pour, Ms. Shirin Moradi and Ms. Delnya Pazirofte for their technical help and support while writing the paper.

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