Effects of X-Ray Irradiation on the Nutritional and Antinutritional Factor Contents of Ten Soybean Cultivars in Wukari, North-Eastern Nigeria

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Abstract

The use of traditional processing methods has fast eroded the nutritive, organoleptic and sensory characteristics of legumes such as soybean. Although soaking, germination, cooking and fermentation have been used to reduce antinutritional factors of legumes, irradiation remains the most reliable and safe method for the improvement and preservation of nutritional values of foods. Study was conducted at the Teaching and Research Farms of the Federal University Wukari, Taraba State between the 2017 and 2018 cropping seasons and harvested seeds were evaluated for trypsin, tannins, phytate, crude protein, crude fiber, carbohydrate, moisture, ash and oil contents. Following the ANF and proximate analyses, ten (10) best performers were selected and irradiated with varied x-ray dosages (40, 50, 60, 70 and 120⁰Rad). Data collected were subjected to Analysis of Variance with mean separated using Least Significant Difference (LSD), at 5% probability level. Results of the study revealed that x-ray dosage 70⁰Rad caused a significant reduction (12% and 7%) in the tannins and phytate contents respectively, whereas trypsin was significantly reduced (11%) in TGX1904-6F at dosage levels between 60⁰Rad and 70⁰Rad.

Introduction

Soybean has many nutritional benefits for man and livestock, as it can as well serve other industrial and commercial uses (Malik *et al.*, 2007). It is classified as an oilseed and it contains significant amounts of all the essential amino acids, minerals and vitamins for human nutrition (Fu *et al.*, 2002). With an average of 40% dietary protein content, 30% carbohydrate and oil content of 20%, soybean is therefore an important source of human diet (Adu-Dapaah *et al.*, 2004). The poultry, pig and fish farming industries

especially, are benefiting tremendously from soybean as a cheap source of high quality protein feed. Soybean oil is the World's most widely used edible oil, as it is low in cholesterol, with a natural taste and imperceptible odour, which makes it the ultimate choice of vegetable oil for domestic and industrial food processing (Mpepereki *et al.*, 2000). In West Africa, soybean has become a major source of high quality and cheap protein for the poor and rural households, as it is used in processing soy cheese, cakes, baby foods and local seasoning product for stews and soups (Malik *et al.*, 2007). It is also used to fortify various traditional foods such as gari, sauces, stew, soups, banku and kenkey to improve their nutritional levels (Ojiako and Igwe, 2008).

Soybean contains some anti-nutritional substances that reduce the nutritional value of the beans and are dangerous to health and therefore, need to be entirely removed, broken down or reduced to the tolerable content before they can be consumed (Messina, 2004). These bioactive compounds, with toxic antinutritional properties can alter the body metabolism of consumers and exert a negative impact on the nutritional quality of the seed protein (Armour *et al.*, 1994). Prominent among the antinutritional factors found in soybean are trypsin inhibitors (protease inhibitors), tannins and phytic acid (Sumner *et al.*, 2003). Tannins cause decreased feed consumption in animals, bind dietary protein and digestive enzymes to form complexes that are not readily digestible (Aletor, 1993a). They also cause decreased palatability and reduced growth rate (Fu *et al.*, 2002). Generally, the bioavailability of the essential nutrients in plant products could be reduced by the presence-of some anti-nutritional factors such as protease, tannins and phytate (Akindahunsi and Salawu, 2005). Also, the nutritional and chemical compositions of soybean may vary extensively, depending on the cultivar and growing conditions (Berk, 1992).

Information on the effect of different processing techniques on the anti-nutritional factors in human foods and animal feeds attract interests by human and animal nutritionists, dieticians, veterinarians, public health and food regulatory authorities on the exploitation of these techniques so that the nutritive values of foods and feeds could be appropriately preserved. According to Gharaghani *et al.* (2008), glucosinolate content of canola meal for broiler chickens was reduced to 40, 70 and 89% at gamma irradiation dosage levels of 10, 20 and 30 kGy, respectively (p<0.01). Other investigators reported that anti nutritional factors, such as protease inhibitors (El-Morsi *et al.*, 1992; Farag, 1998), a-amylase inhibitors (Abu-Tarboush, 1998; Al-Kahtani, 1995), phytohamagglutinins (Farag, 1989; Mahrous, 1992; Farag, 1998), oligosaccharids (Ghazy, 1990) and tannin (Abu-Tarboush, 1998) were significantly inactivated by irradiation. Second only to Gamma photons, x-ray is moderately penetrating, posing minimal hazard and have been successfully used to achieve breeding breakthrough by Getha (1994) and Pavadai *et al.* (2010).

This research endeavor was intended to assess the effect of x-ray irradiation (at varying dosage levels) on the nutritional and antinutritional contents in the commonly cultivated soybean varieties around Wukari and environment.

Keywords: trypsin, phytate, variations, tannins, irradiation.

Materials and Methods

Seeds of commonly cultivated varieties of soybean were planted during the 2017/2018 growing season, at the Teaching and Research Farms of the Federal University Wukari, Wukari, Nigeria. Wukari is situated on latitude 7^o 52'17.00^oN and longitude 9^o 46'40.30^oE. It falls within the guinea savannah of North-eastern Nigeria with the annual rainfall of 1058mm-1300mm and the relative humidity dropping to about 15%, alongside with the annual temperature of 28^oC and 30^oC. Its characteristic alfisol soil is clay enriched, with subsoil that has relatively high native fertility is suitable for the cultivation of many crops such as yam, soybean, sorghum, maize, rice and other assorted fruit and vegetables (Franke and Rufino, 2010).

Collection of experimental materials

Twenty five well identified and certified soybean varieties, popularly grown in the Wukari Local Government area of Taraba state, were obtained from the College of Agricultural Research, Yandev, Benue state, Institute for Agricultural Research, Samaru, Kaduna state, and the International Institute for Tropical Agriculture, Ibadan, Oyo state.

Field layout and experimental design

The experimental field was laid out in Randomized Complete Block Design (RCBD), with three replications, for each of the 25 soybean varieties. Beds of dimensions $2\times5m$ were horizontally aligned, with 50cm distance between beds, 1m demarcation between replications and 2m border row across the length and breadth of the experimental field. At the experimental site, planting distance of 50cm intrarow and 75cm inter-row spacing was used, thus there were 35 soybean plant stands per bed. Thus, the dimension of each of the experimental fields was $96m\times26m$ ($2,496m^2$) and the total number of soybean plant stands per field was 5,180. Soybean seeds were sown directly into the raised bed, using the drilling planting method, by placing seeds inside the shallow dug holes of about 2cm depth and covered with soil.

Field management

Plants were raised following the standard agronomic and cultural practices recommended for soybean cultivation, as contained in the IITA handbook of Soybean production.

Analysis and quantification of antinutritional factors content of soybean

The determination, analysis and quantification of the antinutritional factors in the different soybean varieties used for the purpose of this research work was carried out through the following procedures;

Analysis of trypsin inhibitor, (Prokopet and Unlenbruck, 2002)

Materials: Grinder/blender, conical flasks, Centrifuge and Spectrophotometer

Reagents: Trypsin inhibitor standards, Sodium Chloride (NaCl)

Procedure:

- 1. 1g of dry well blended dried soybean sample was weighed into a flask
- 2. 50ml of 0.5M NaCl was added to the blended sample
- 3. The solution thus obtained was then stirred for 30 minutes and centrifuged at 1500rpm for 5 minutes
- 4. The solution was thereafter decanted and the filtrate kept. 10ml of filtrate was pipette and put into another flask
- 5. Next, 2ml of standard trypsin solution of known concentration (2mg/l) was added to the 10ml filtrate
- 6. Absorbance was measured at 410nm using 10ml of same substrate (the sample filtrate) as blank
- 7. Also prepare 1mg, 2mg, 4mg, 6mg, 8mg, and 10mg/l standard trypsin inhibitor were also prepared and their absorbencies measured at 410nm.
- 8. A standard graph of absorbance against concentration was then plotted
- 9. Extrapolation was achieved by tracing the absorbance of the sample down the concentration axis to obtain the trypsin inhibitor concentration of the sample

Calculation:

Trypsin inhibitor content (mg/kg) = Conc. obtained in mg/l x volume of sample x DF Sample weight

DF: Dilution factor. If not diluted, then DF = 1

Analysis of tannin, using the Folin Ceocalteu Method

Materials: Blender, Conical flasks, Centrifuge

Reagents: Folin Ceocalteous reagent, Na₂CO₃ (saturated), Tannic acid standard

Procedure:

1. 1g of dry well blended soybean sample was weighed into a conical flask

- 2. 10ml of distilled water was then added, the mixture agitated and left for 30 minutes at room temperature
- 3. The mixture was centrifuged at 2500rpm for 15min
- 4. 2ml of supernatant was measured into a 10ml volumetric flask and 1ml of folin-ceocalteu reagent was added to it
- 5. Then 2ml of saturated Na₂CO₃ solution was added to the mixture and the solution was diluted to 10ml with distilled water
- 6. The solution was then incubated for 30min at room temperature

Preparation of standard tannic acid:

- 7. The procedure 1 to 6 was repeated for tannic acid standards 20, 40, 60, 80, 100, 120mg/l from a stock of 500ppm (50mg of Tannic acid standard dissolved in 100ml of distilled water) excluding centrifugation (procedure 3)
- 8. Absorbance of the above Tannic acid concentrations was read off at a wavelength of 725nm and a calibration curve for the tannic acid standards was drawn. That is, absorbance against concentration
- 9. Extrapolation was done by tracing the absorbance of the sample down the concentration axis to obtain the tannic acid concentration of the sample

Calculation

Tannic Acid content (mg/kg) = Conc. obtained in mg/l x volume of sample x DF Sample weight

DF: Dilution factor. If not diluted, then DF = 1

Determination of phytate (phytic acid), following the Eskin's methologies.

Materials: Conical flask, Filter paper, Pipette, Beaker and Titrating apparatus

Reagents: Hydrochloric acid (HCl), Ammonium thiocyanate and Iron iii chloride (FeCl₃₎

Procedure

- 1. 2g of dry finely ground sample of soybean was weighed into a 250ml conical flask
- 2. 100ml of 2% concentrated HCl was added and allowed to soak for 3 hours and then filtered
- 3. 50ml of the filtrate was pipette into a 250ml beaker and 107ml of distilled water was added to improve acidity

- 4. 10ml of 0.3% ammonium thiocyanate solution was added as indicator
- 5. Titration with standard iron iii chloride (FeCl3) solution which contain 0.00195g iron/ml until a brownish yellow colour appear and persist for 5min
- 6. The phytic acid content was calculated as shown below

Phytic acid g/kg = 0.00195 x volume of FeCl3 consumed x DF

Sample wt

DF: Total volume of extraction solvent added/volume of aliquot taken for the titration.

Determination of nutritional content using methods of Association of Analytical Chemists

Moisture content

One gram of sample in pre-weighed crucible was placed in an oven (at 105^oC) for 24 hours, allowed to cool and then re-weighed. The percentage moisture was thus calculated as follows;

Moisture content (%) = $\frac{W_2 - W_3 \times 100}{W_2 - W_1}$

Where: W₁ is the weight of the crucible

 W_2 is the weight of the crucible after drying at 105^0 C and sample and

W₃ is the weight of the crucible and the sample after cooling in airtight desicators

Determination of ash content

Two grams of sample was added into a pre-weighed crucible and incinerated in muffle furnace at 600° C, with the value calculated as follows;

Ash content (%) =
$$\frac{W_2 - W_3}{W_2 - W_1} \times 100$$

Where: W1 is the weight of cleaned, dried, ignited and cooled crucible

 W_2 is the weight of the crucible and sample after incinerating at 600^{0} C and

 W_3 is the weight of the crucible and the sample after cooling in airtight homogenized vessel

Fat and oil content

The fat and oil content was estimated using Tecatoy Soxtec (Model 2043[20430001]; Hilleroed, Denmark). A quantity of 1.5g sample mixed with 2.3g anhydrous sulfate was weighed into a thimble and



covered with absorbent cotton, while 40ml of petroleum ether $(40 - 60^{\circ}\text{C Bpt})$ was added to a pre-weighed cup. Both thimble and cup were attached to the Extraction Unit. The sample was extracted using ethanol for 30 minutes and rinsed for 90 minutes. Thereafter, the solvent was evaporated from the cup to the condensing column. Extracted fat in the cup was then placed in an oven at 105°C for 1 hour, allowed to cool and weighed. Percentage Fat/oil was then calculated as;

 $Lipid = \underline{Initial \ cup \ weight} - \underline{Final \ cup \ weight} \times 100$ Weight of sample

Crude protein

The crude protein content was determined using the micro-Kjeldahl method, as described by Pearson (1976). A volume of 10mL H₂SO₄ added to 3g of sample was digested with a digestor (model Bauchi 430) for 90 minutes. A volume of 40mL water was added and distilled using a Kjeldahl distillation Unit (model unit B - 316) containing 40% concentrated sodium hydroxide and Millipore water. Liberated ammonia was collected in 20mL boric acid with bromocresol green and methyl red indicators and titrated against 0.04N H₂SO₄. A blank (without sample) was likewise prepared. Percentage protein was thus calculated as;

Crude protein (%) = $\frac{\text{Sample titer} - \text{blank titer} \times 14 \times 6.25}{\text{Sample weight}} \times 100$

Where: 14 is the molecular weight of nitrogen and 6.25 is the nitrogen factor

Crude fiber content

A weighed crucible containing 1g of defatted sample was attached to the extraction unit (in Kjeldahl, D-40599; Behr Labor-Technik GmbH, Dusseldorf, Germany) and into this 150mL of hot 1.25% H₂SO₄ was added and digested for 30 minutes, then the acid was drained and sample washed with hot distilled water for 90 minutes. The crucible was removed and oven-dried overnight at 105⁰C, allowed to cool down, weighed and incinerated at 550⁰C in a muffle furnace (MF-1-02; PCSIR Labs, Lahore, Pakistan) overnight and reweighed after cooling. Percentage extracted fiber was thus calculated as;

Crude fiber (%) = <u>Weight of digested sample – Weight of ashed sample</u> \times 100 Weight of sample

Carbohydrate content

The carbohydrate content was determined by the difference, that is, addition of all the percentages of moisture, fat, crude protein, ash and crude fiber was subtracted from 100%. This gave the amount of nitrogen free extract otherwise known as carbohydrate. The value is calculated as; Carbohydrate (%) = 100 - (% moisture + % Fat + % Ash + % crude fiber + % crude protein)



Exposure of soybean varieties to irradiation substances

Seeds of the soybean varieties were exposed to physical mutagen (x-ray), at the x-ray unit of the General Hospital, Wukari, Taraba State. The doses were 0, 40, 50, 60, 70 and 120⁰ Rad.

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Procedure

- 1. The ten best soybean performers were selected and prepared for exposure to the x-ray
- 2. 200 seeds from each of the selected varieties were wrapped in moistened tissue paper and enveloped separately
- 3. Samples (following steps 1 and 2) were prepared in accordance with the intended varying dosage levels of exposure; 40, 50, 60, 70 and 120^{0} Rad respectively, tagged accordingly and then sealed together in envelopes, one for each dosage
- 4. The envelopes were conveyed to the x-ray chambers of the x-ray unit of the General Hospital where the irradiation was conducted



Results

Comparison of Soybean Seed Nutritional Content Prior to and after Irradiation

The yield and proximate nutritional contents of the ten selected soybean varieties were compared prior to and after exposure to the x-ray (Tables 1 and 2). The effect of exposure to x-ray was least for the carbohydrate and protein contents of the soybean varieties under study, with significant difference recorded for TGM120, TGM574 and TGM584 in carbohydrate content. For protein, significant differences were recorded for TGM555 and TGN954 only, and the higher values were recorded after irradiation. Also, irradiation was seen to produce a slightly significant increase in the ash content of TGM951, TGX1904-6F and TGM120, with no pronounced effect on TGM555, TGM577, TGM553, TGM111, TGM574, TGM584 and TGM954.

However, the effect of exposure to x-ray was significant in the fiber content, resulting in lower statistically significant values for most of the varieties studied, except for TGM951, TGX1904-6F and TGM111, that recorded higher values after irradiation. Effect of irradiation (x-ray, between 50rad and 70rad) was notable in almost all soybean varieties under study for moisture, ash, oil and fiber contents. Moisture content was influenced by irradiation, resulting in a significant increase in TGX1904-6F, TGM555 and TGM111. Conversely, irradiation caused a significant moisture reduction in TGM951, TGM577, TGM574 and TGM954, whereas there was no significant difference in moisture content of TGM553, TGM120 and TGM584 prior to and after irradiation.

Increase in oil content of TGM951, TGM1904-6F, TGM555, TGM577, TGM120, TGM574 and TGM954, due to irradiation was significant, meanwhile it had no significant effect on TGM553, TGM111and TGM584. Irradiation caused significant decrease in the fiber content of TGM555, TGM577, TGM120, TGM574, TGM584 and TGM954, while it resulted in significant increase in TGM951, TGX1904-6F and TGM111. However, irradiation produced no significant effect in the fiber content of TGM553.

SN	Variety	Moisture (%)	Ash (%)	Oil (%)	Protein (%)	Fiber (%)	Carbohydrate (%)
1.	TGM951	7.79 ^{cd}	3.05 ^{ab}	21.33 ^g	39.45 ^e	2.46 ^b	25.93ª
2.	TGX1904-6F	7.82°	3.05 ^b	21.38^{f}	39.50 ^d	2.46 ^b	25.80 ^b
3.	T555	7.85 ^b	3.09 ^a	21.52 ^c	39.59°	2.48 ^{ab}	25.49 ^d
4.	T577	7.89 ^a	3.08 ^a	21.46 ^e	39.66 ^{ab}	2.49 ^a	25.44 ^d
5.	T553	7.89 ^a	3.07 ^a	21.52°	39.66 ^{ab}	2.47 ^b	25.43 ^d
6.	T111	7.77 ^d	3.08 ^a	21.49 ^d	39.57°	2.43°	25.67°
7.	T120	7.88 ^a	3.07 ^a	21.55 ^{ab}	39.66 ^{ab}	2.47 ^b	25.39 ^e
8.	T574	7.90 ^a	3.08 ^a	21.56 ^a	39.67 ^a	2.46 ^b	25.36 ^f
9.	T584	7.89 ^a	3.09 ^a	21.53 ^{bc}	39.63 ^b	2.49 ^a	25.38 ^{ef}
10.	Т954	7.89 ^a	3.06 ^a	21.53 ^{bc}	39.63 ^b	2.45 ^{bc}	25.44 ^d

 Table 1:
 Comparison of Soybean Seed Nutritional Content prior to Irradiation

Means within each column followed by the same alphabet are not significantly different from one another based on the 0.05 probability level of DMRT

Table 2:	Comparison	of Soybean	Seed Nutrit	ional Content af	ter Irradiation
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SN	Variety	Moisture (%)	Ash (%)	Oil (%)	Protein (%)	Fiber (%)	Carbohydrate (%)
1.	TGM951	7.75 ^f	3.07 ^a	21.36 ^f	39.48 ^e	2.50 ^a	29.83 ^a
2.	TGX1904-6Fi	7.83 ^d	3.07 ^a	21.41 ^e	39.55 ^d	2.48^{ab}	25.68 ^b
3.	T555i	7.87 ^{bc}	3.08 ^a	21.65 ^a	39.65 ^b	2.46 ^{bc}	25.40^{d}
4.	T577i	7.87 ^c	3.08 ^a	21.57 ^b	39.68 ^{ab}	2.47 ^{bc}	25.35 ^d
5.	T553i	7.90^{a}	3.07 ^a	21.65 ^c	39.66 ^{ab}	2.48 ^{bc}	25.36 ^d
6.	T111i	7.80 ^e	3.07 ^a	21.51 ^d	39.61°	2.46 ^{bc}	25.56 ^c
7.	T120i	7.89^{a}	3.08 ^a	21.57 ^b	39.67 ^{ab}	2.44 ^c	25.36 ^d
8.	T574i	7.86 ^{ab}	3.08 ^a	21.58 ^b	39.69 ^a	2.45 ^c	25.33 ^d
9.	T584i	7.90^{a}	3.09 ^a	21.55 ^{bc}	39.66 ^{ab}	2.47 ^{abc}	25.35 ^d
10	T954i	7.86 ^c	3.08 ^a	21.56 ^b	39.67 ^b	2.44 ^c	25.42 ^d

Means within each column followed by the same alphabet are not significantly different from one another based on the 0.05 probability level of DMRT

Values of Tannins, Trypsin and Phytate in 10 Selected Soybean Varieties at Different Dosages of Irradiation

The mean values of antinutritional factors content of the selected soybean varieties (Table 3) showed a significant difference in the values of ANF contents among the soybean varieties studied and also, their responses to the different dosages of x-ray radiation were significantly different. The mean value of tannins in the analyzed varieties, prior to irradiation was 691.51g/kg. Upon exposure to different dosages (40^{0} , 50^{0} , 60^{0} , 70^{0} and 120^{0} Rads) of x-ray, the result obtained showed that TGM954 recorded the least value (415.53g/kg), at 70Rad, as against 629.81g/kg, being the value of its initial content. Exposure to x-ray at 70^{0} Rad resulted in the least trypsin content in TGM584, whereby the initial quantity (457.08g/kg) was reduced to 333.33g/kg. However, the average trypsin content obtained in the soybean seed lot, at 70^{0} Rad was 454.80g/kg. 70^{0} Rad produced a significant reduction in the phytate content of the studied soybean seeds, recording the least value (26.21g/kg) in TGM951, as against the initial value (31.88g/kg), prior to exposure.

SN	Dosage	Mean (g/kg)	M. Square	Min. (g/kg)	Max. (g/kg)
TANNINS					
1.	0	691.51 _{±39.8931}	158.07*	580.13	939.81
2.	40	$658.01_{\pm 40.3989}$	16320.73*	562.36	931.37
3.	50	$639.29_{\pm 27.3381}$	7473.74*	509.71	784.65
4.	60	$648.07_{\pm 21.4907}$	4618.49*	583.14	794.66
5.	70	$607.86_{\pm 39.7510}$	15801.41*	415.53	875.73
6.	120	$601.62_{\pm 16.6458}$	2770.84*	473.04	660.09
TRYPSIN					
1.	0	565.61 _{±32.8999}	10824.01*	410.50	694.61
2.	40	$477.47_{\pm 27.6502}$	7645.32*	382.84	616.81
3.	50	$487.03_{\pm 29.3560}$	8617.77*	365.00	621.08
4.	60	$454.80_{\pm 33.6140}$	11296.59*	333.33	668.50
5.	70	$489.10_{\pm 37.7104}$	14220.77*	353.85	735.29
6.	120	$508.52_{\pm 25.4331}$	6468.44*	379.41	639.71
PHYTATE					
1.	0	$34.24_{\pm 1.4799}$	21.90*	28.40	42.79
2.	40	$56.83_{\pm 3.7626}$	141.58*	41.56	77.72
3.	50	$51.85_{\pm 4.4858}$	201.23*	29.73	77.24
4.	60	$48.19_{\pm 4.1101}$	168.93*	34.46	65.68
5.	70	$50.38_{\pm 4.0160}$	161.28*	26.21	66.91
6.	120	$46.30_{\pm 4.4371}$	196.88*	31.81	77.24

Table 3: Mean Values of Tannins, Trypsin and Phytate in 10 Selected SoybeanVarieties at Different Dosages of Irradiation

* = significant at $P \le 0.05$

Discussions

Irradiation was seen to have influenced a slight but significant increase in the oil and protein content of soybean seeds, while its effect did not show pronounced changes in the moisture, ash and fiber contents. Meanwhile, irradiation caused a significant reduction in the carbohydrate content of the soybean seeds after exposure to the x-ray. The most significant reduction in the tannin content of the soybean was seen to be produced by irradiation dosages around 70° and 120° Rad. At this range, the tannin content of TGM954 and TGM553 were reduced from 629.81g/kg and 799.02g/kg to 415.53g/kg and 473.04g/kg respectively. Mahdi et al. (2003) also reported that the activity of trypsin inhibitor in broad bean was reduced by 4.5%, 6.7%, 8.5% and 9.2% at 2.5kGy, 5.0kGy, 7.5kGy and 10kGy of gamma irradiation respectively. Similarly, irradiations at 10.2, 12.3, 15.4 and 18.2kGy reduced the phytic acid content. Exposure to x-ray dosages between 60° Rad and 70° Rad gave the best result, yielding a significant reduction in the trypsin content of TGM584 and TGX1904-6F. In TGM584, the trypsin content was reduced from the initial 457.08g/kg, to 333.33g/kg, while in TGX1904-6F, its value was reduced to 353.89g/kg, from the initial 538.89g/kg. Irradiation produced significant effect in the phytate content of soybean seeds analyzed in agreement with the finding of Pavadai et al. (2010).

Though exposure to 70^{0} Rad of x-ray resulted in the least value for phytate in TGM951, 50^{0} Rad also produced a similar effect in TGM574, in which case the phytate content was reduced from 36.03g/kg to 29.73g/kg. Generally, exposure of soybean seeds to x-ray produced significant effect in their ANF content. In some cases, depending on the dosage, the effect was negative, causing an increase in the quantity of ANF contained in the sampled seeds, while the effect was positive for most of the varieties, especially at relatively higher dosages, ranging from 60^{0} Rad to 70^{0} Rad and also 120^{0} Rad. Value of soybean is primarily determined by the protein and oil content of the seed, while the value to the producer is determined by the yield (Orf and Helms, 1994). During the last 30 years, it has been observed that about 30 cultivars of soybean have been produced by x-rays, thermal neutron and gamma irradiation in different countries, especially in China (Sagel *et al.*, 1995). While most treatments (soaking, heating, fermentation and de-hulling) used to reduce the ANF content of legumes adversely affect the sensory characteristics of the final product, irradiation has been recognized as a reliable and safe method for improvement of the nutritional value of food (Diehl, 2002; Al-Kaisey *et al.*, 2002).

References

- Adu-Dapaah, H. K., Asafo-Adjei, B., Owusu-Akyaw, M. and Amoah, S. (2004). Sustainable Soybean Production in Ghana. Paper presented at Program on Soybean in Ghana, pp 6.
- Abu-Tarboush, H. M. (1998). Irradiation inactivation of some antinutritional factors in plant seeds. *Journal of Agriculture and Food Chemistry* 46, 2698-2702.
- Akindahunsi, A. A. and Salawu, S. O. (2005). Phytochemical screening and nutrient and antinutrient composition of selected tropical grean leafy vegetables. *African Journal Biotechnol 4, 497-501.*
- Aletor, V. A. (1993). Allelochemicals in Plant Foods and Feeding Stuffs. Part I. Nutritional, Biochemical and Physiopathological aspects in animal production. *Veterinary* and Human Toxicology 35(1), 57-67.
- Armour, J. C., Chanaka Perera, R. L., Buchan, W. C. and Grant, G. (1998). Protease Inhibitors and Lectins in Soybeans and Effect of Aqueous Heat-Treatment. *Journal of Science, Food and Agriculture* 78, 225-231.
- Al-Kaisey, M. T., Mohammed, M. A., Alwan, A. K. H. and Mo-hammed, M. H. (2002). The Effect of Gamma Irradiation on the Viscosity of Two Barley Cultivars for Broiler Chicks. *Radiation Physics Chemistry* 63, 295–297.
- Berk, Z. Food and Agriculture Organization of the United Nations (1992). Technology of Production of Edible Flours and Protein Products from Soybeans. *FAO agricultural services bulletins No* 97. 1992.
- El-Morsi, E. A., Abdel-Salam, H. S., Ismail, H. A., Aboul-Fetouh, S. A. and Fawzay, A. M. (1992). Effect of Gamma Irradiation on Trypsin Inhibitor and Subunit Structure of Proteins in Field Bean (*Vicia faba*) Seeds during Germination". *Minia J. Agric. Res. Develop. 14, 849-871.*
- Diehl, J. F. (2002). Food Irradiation-past, Present and Future. *Radiat. Phys. Chem.* 63, 211-215.
- Farag, M. D. E. H. (1998). The Nutritive Value for Chicks of Full-fat Soybeans Irradiated at up to 60kGy. *Animal Feed Science Technology*. 73, 319-328.
- Fu, P. P., Xia, Q., Lin, G. and Chou, M. W. (2002). Genotoxic Pyrrolizidine Alkaloids-Mechanisms Leading to DNA Adduct Formation and Tumorigenicity. *International Journal of Molecular Science* 3,948-964.

- Franke, A. C., Rufino, M. C. (2010). Characterization of the Impact Zones and Mandate areas in the N2Africa Project, pp 142.
- Gharaghani, H., Zaghari, M., Shahhoseini, G. and Moravej, H. (2008). Effect of Gamma Irradiation on Antinutritional Factors and Nutritional Value of Canola Meal for Broiler Chickens. Asian-Australian Journal of Animal Science 21, 1479-1485.
- Gatel, F. (1994). Protein Quality of Legume Seeds for Non-Ruminant Animals; a review. *Animal Feed Science Technology* 45, 317–348.
- Ghazy, M. A. (1990). Effect of Gamma-Irradiation on some Antinutritional Factors in Kidney Bean (*Phaseolus vulgaris L*) Seeds. *Minia Journal of Agric. Resources* Development 12, 1965-1980.
- Mahdi, T., Al-Kaisey, Abdul-Kader, H. A., Manal, H. M. and Amjed, H. S. (2003). Effect of Gamma Irradiation on Antinutritional Factors in Broad Bean. *Journal of Radiation Physics and Chemistry* 67, 493-496
- Mpepereki, S., Javaheri, F. and Davis, P. (2000). Soybean and Sustainable Agriculture: Promiscuous Soybean in Southern Africa. *Field Crops Resource 63, 137 – 149*.
- Messina, M. (2004). Nutritional Value of Soyfood. *Ph.D. Thesis*. Department of Nutrition Michigan State University, *United State of America*, 6. 215-223.
- Malik, M. F. A., Ashraf, M., Qureshi, A. and Khan, M. R. (2011). Investigation and Comparison of some Morphological Traits of the Soybean Populations using Cluster Analysis. *Pakistan Journal of Botany* 43(2), 1249-1255.
- Ojiako, O. A. and Igwe, C. U. (2008). The Nutritive, Anti-nutritive and Hepatotoxic Properties of *Trichosanthes anguina* (Snake Tomato) Fruits from Nigeria. *Pakistan Journal* 7(1), 85-89.
- Orf, J. H. and Helms, T. C. (1994). Selection to Maximize Gross value per hectare within three Soybean Populations. *Crop Science*, *34*, *1163-1167*.
- Pavadai, P., Girija, M., and Dhanavel, D. (2010). Effect of Gamma Rays on some Yield Parameters and Protein Content of Soybean in M2, M3 and M4 Generation. *Journal of Expt. Science 1(6)*, 8-11.
- Sagel, Z., Atila, A. S. and Tutluer, M. I. (1995). Characteristics of Improved Mutant Varieties in Soybean. *IAEA*. *Vienna 21, 160-164*.