

## FURTHER OBSERVATIONS ON THE EFFECTS OF FAST NEUTRON IRRADIATION ON THE MORPHOLOGICAL AND YIELD TRAITS OF $M_1$ AND $M_2$ GENERATION OF AFRICAN WRINKLED PEPPER (*Capsicum annum* var *abbreviatum* FINGERH)

Daudu, O. A. Y.,<sup>1</sup> Falusi O. A.<sup>1</sup>, Thomas T.,<sup>1</sup> Mohammed, D. C.,<sup>1</sup> Muhammad, L. M. and Lateef A. A.<sup>1</sup>  
<sup>1</sup>Department of Biological Sciences, Federal University of Technology, Minna, Niger State, Nigeria  
 Correspondence: [dauduoladipupoyusuf@yahoo.com](mailto:dauduoladipupoyusuf@yahoo.com)

### ABSTRACT

The mutagenic effects of fast Neutron irradiation (FNI) from an Am-Be source with a flux of  $1.5 \times 10^{14}$  neutrons/cm<sup>2</sup> on the morphological and yield traits of  $M_1$  and  $M_2$  ( $M$  = filial mutant) generation of African Wrinkled Pepper (*Capsicum annum* var *abbreviatum* Fingerh) were studied. Seeds of the pepper variety were irradiated for 0, 30, 60, 90 and 120 min before they were sown with their respective controls - in order to assess the effect of the different irradiation treatments on the  $M_1$  and  $M_2$  generations of the pepper plants. Results showed both negative and positive shifts of characters as a result of FNI treatments. However, 60 minutes was observed to be the most effective IEP to induce viable and useful mutations for yield parameters in the pepper plants.

**Keywords:** mutagenic effects, IEP, FNI,  $M_1$  and  $M_2$  generations, yield parameters.

The *Capsicum* genus contains numerous species of sweet and hot peppers (Reifschneider, 2000). The most common are *C. annum* L., *C. baccatum* L., *C. chinense* Jacq., *C. frutescens* L., and *C. pubescens* R. & P., and horticulturists have cultivated these species worldwide for long time (Casali and Couto, 1984; Moscone *et al.*, 2007). In Nigeria, the most indigenous are *C. annum* and *C. frutescens* because these species have a large genetic diversity in the region, and they are well adapted to the diverse environmental conditions around the country. In addition, they are the most recognized species grown in commercial quantities all over Nigeria (Falusi and Morakinyo, 2001; Mady *et al.*, 2005; Falusi, 2007). These two species form an important ingredient in people's diet around the world (GRIN, 2009) due to the pungency properties of the fruits resulting from their high concentration of capsaicinoid alkaloid (Bosland and Vostava, 2000). In addition, *Capsicum* is a rich source of vitamins A complex, B1 and B2, C and minerals such as dietary calcium, iron and phosphorus (Bosland, 1992; Gill, 1992; Ado, 1999). The content of vitamin C in the *Capsicum* fruit is higher than in *Citrus*. *Capsicum* fruits are also popular as food spices, as a colouring agent and serve as

pharmaceutical ingredients (Bosland, 1996). In African medicine, it is used to treat sore throats (Abdullahi *et al.*, 2003). These popular uses which *Capsicum* peppers have been put to have fuelled increasing demand for the crop thus necessitating corresponding increased supply of the product. Though, attempts have been made to achieve this increased supply through increased cultivation of the different varieties and species; the successes of such attempts were limited by challenges ranging from dwindling man-power to inadequate farming conditions. Thus, attention is gradually shifting towards improving the genetic quality of the species through plant breeding and selection made possible by radiation-induced genetic variability. This, perhaps, will ensure that the limited acreage of *Capsicum* will go a long way in meeting our needs. Mutations, which render the genetic information of a cell or living creature to be different from that of another, are the tools used to study the nature and function of genes which are the building blocks and basis of plant growth and development. Through producing raw materials for genetic improvement of economic crops (Adams and Aliyu, 2007).

Mutation technology has been used to produce many cultivars with improved economic value.

to advance the study of genetics and plant environmental phenomena (Bortagne-Sagnard *et al.*, 2008; Romanandi and Hozakane, 2009). Genetic variability for desired characters can be induced successfully through mutations, with high practical value in plant improvement programs (Fahad and Salim, 2009).

Induced mutations have been used to generate genetic variability and have been successfully utilized to improve yield components of various crops like *Oryza sativa* (Awan *et al.*, 1980; Singh *et al.*, 1998), *Hordeum vulgare* (Ramesh *et al.*, 2001), *Triticum durum* (Sakin and Yildirim, 2004), *Cicer arietinum* (Wani and Aris, 2001), *Hydrocotyle* (Misra *et al.*, 2001), *Helianthus annuus* (Elangovan, 2001), *Cajanus cajan* (Ravikesavan *et al.*, 2001), *Sesamum indicum* (Mensah *et al.*, 2007), *Guzonias abyssinica* (Misra, 2001), *Solanum tuberosum* (Shin *et al.*, 2011). These reports show that mutagenesis is a potential tool to be employed for crop improvement. Mutation breeding employing fast neutron irradiation (FNI) has been used to develop new varieties (Sodkiewicz and Sodkiewicz, 1999) and is widely used for the induction of mutations (Zhang *et al.*, 2002) resulting in a significant increase in the yield of major crops, including chili (Swaminathan, 1998).

The FAO (2009) reported that 2008 marked the 80<sup>th</sup> anniversary of mutation induction in plants. The application of gamma rays and other physical mutagens such as fast neutrons has generated a vast amount of genetic variability and has played a significant role in plant breeding and genetic studies (David, 2010). The widespread use of induced mutants in plant breeding programmes throughout the world has led to the official release of more than 2700 plant mutant varieties (FAO 2009). Since FNI-induced mutations in pepper could be useful as a new source of altered germplasm, our objective is to assess the impact of FNI on growth and yield parameters of  $M_1$  and  $M_2$  (Mutant generation) generation of African Wrinkled Pepper (AWP), (*Capsicum annum var abbreviatum* Fingerh extending results achieved more recently (Falusi *et al.*, 2012).

#### Materials and Methods

Fifty (50) fresh fruits of AWP accessions were bought from a local farmer in Minna, Niger

State, Nigeria. The fruits were kept in a clean polythene bag. The accessions were identified as *Capsicum annum var abbreviatum* Fingerh using taxonomic aid provided by Simmond (1970), as well as morphological description of Hutchinson and Dalziel (1960), Schippers (2000) and Abdufahri *et al.* (2001). Each fruit of the AWP accessions was cut open, the seeds were removed, kept separately and sun-dried for 8 h. The dry seeds of *Capsicum* were irradiated at the Centre for Energy and Research Training (CERT), Ahmadu Bello University, Zaria with FNI using an Americium-Beryllium source with a flux of  $1.5 \times 10^7 \text{ n cm}^{-2} \text{ s}^{-1}$  for five different irradiation exposure periods (IEPs): 0, 30, 60, 90, and 120 min. The equipment used was a Miniature Neutron Source Reactor (MNSR) designed by the China Institute of Atomic Energy (CIAE) and licensed to operate at a maximum power of 31 kW (SAR, 2005).

The sun-dried seeds were tested for viability using the floatation method before FNI treatment. Treated seeds (100 from each treatment) were then planted in nursery trays to obtain seedlings, which were transplanted into 3.5-l plastic pots containing garden soil, at a rate of three seedlings/pot after 4 weeks in the nursery. The planted seeds were watered once daily between 5.00-6.30 pm using bore-hole water. Each treatment was replicated four times using a completely randomized design (CRD).

Data were randomly collected from 15 plants for germination percentage, number of leaves/plant at maturity, height of plant at maturity (number of days to 50% flowering) and yield/plant in each  $M_1$  and  $M_2$  ( $M_1$  = filial mutant) generation. Data was analyzed using analysis of variance (ANOVA) and Least Significant Difference test was used to separate the means with significant differences detected at  $P = 0.05$ . Pearson's correlation analysis was used to find the relationship between treatments and selected parameters.

#### Results and Discussion

##### For $M_1$ Generation

The FNI treated seeds showed a negative correlation between irradiation period and seedling survival percentage, implying that as irradiation period increased, percentage seedling survival decreased (Table 2). Similar



result was observed in chemical mutagen treated *Sesamum indicum* (Mensah *et al.*, 2007); *Lycopersicon esculentus* (Adamu and Aliyu, 2007). Plant heights were increased in the FNI treated plants; these were significantly different from the control (Table 3). Similarly, total number of leaves/plant including withered leaves was also increased in the FNI treated plants; these were significantly different from the control. Other yield parameters such as number of fruits/plant, number of seeds per fruit, length of fruit (cm), width of fruit (cm) and weight of fruit (g) also increased as the IEP increased. Although, for number of fruit/plants, there were no significant difference between the control and all other treatments. For the length of fruit and weight of fruit, 60 minutes IEP was the best periods among the exposure periods used to induce beneficial mutants; whereas the 120 minutes IEP was the best for the production of heavy fruits. Similar effects of ionizing radiation on reproductive and other yield parameters has been reported for tomato exposed to sodium azide with a concentration between 1 and 4 mM (Adamu and Aliyu, 2007) and also for Okra exposed to Gamma irradiation doses between 300 and 500 Gray (Hegazi and Hamideldin, 2010). Asmahan and Nada (2006), Fahd (2009) and Hegazi and Hamideldin (2010) reported that an increase in irradiation dose tended to increase certain morphological traits such as plant height.

**For M<sub>1</sub> Generation:**

Seedling survival percentage also showed a negative strong correlation with the IEP (Table 2); this implies that as IEP increased, the seedling survival percentage reduced. Plant heights were decreased in 30 and 90 minutes IEP and were significantly lower than the control. However, the 60 and 120 minutes IEP were significantly higher than the control. Total number of leaves/plant including withered leaves was significantly lower in the 30 minutes IEP than the control. However, 30, 60 and 120 minutes IEP showed significant increases in number of leaves/plants. Yield parameters such as number of fruits/plant, number of seeds/fruit, length of fruit, width of fruit and weight of fruit increased significantly as IEP increases (Table 2). The 60 minutes IEP seemed to be consistent in producing better yields amongst other IEPs. Comparing the M<sub>1</sub> and M<sub>2</sub> plants, all the morphological and yield traits studied were lower in the M<sub>2</sub> plants than the M<sub>1</sub> plants. However, higher weight of fruits was recorded in the M<sub>2</sub> generated plants. 60 minutes FNI was observed to be the most effective IEP to induce viable and useful mutations for yield parameters for both M<sub>1</sub> and M<sub>2</sub> generations in the pepper plants. A similar result was reported by Adamu *et al.*, (2004) and Falusi *et al.*, (2012). Thus, 60 minutes IEP of FNI may be utilized to increase variability that will ultimately increase the possibility of isolating mutants for improvement of pepper.

Table 1 Description of the pepper (*Capiscum* spp.) accessions that were used in this study

Code number	Source	Local name	Botanical name	Description
AWP	Minna	Ata Rodu	<i>C. annum</i> var. <i>abbreviatum</i> Fingerh	Medium-sized annual plant small orange and wrinkled fruits with hot taste. 100 pericarp per fruit.



Table 2 Correlation (r) between the treatments and percentage of seeds that germinated per 100 seeds sown for AWP accessions in M<sub>1</sub> and M<sub>2</sub>.

Treatments	MN/AR/002/%	
	M <sub>1</sub>	M <sub>2</sub>
Control	100	90
30 min	30	83
60 min	21	70
90 min	16	51
120 min	16	51
R	-0.8736	-0.9700

Table 3. LSD of the effects of Fast neutron irradiation on agronomic traits of AWP accessions in the M<sub>1</sub> and M<sub>2</sub> generations.

Characters	0	30	60	90	120
(MN/AR/002) M <sub>1</sub>					
Plant height (cm)	21.68±3.68 a	29.94±7.67 b	32.79±5.11 b	29.32±7.05 b	33.70±10.62 b
Number of leaves/plant	67.08±19.68 a	99.20±32.80 b	113.00±2.12 b	99.09±32.31 b	120.10±52.95 b
Number of fruits/plant	15.90±7.56 a	17.80±12.85 a	19.00±7.58 a	19.70±8.55 a	17.50±9.11 a
Number of seeds/fruit	54.90±21.22 a	66.10±11.08 ab	71.90±16.64 b	73.5±19.76 b	72.00±23.87 ab
Length of fruit (cm)	3.95±0.86 a	4.28±0.65 ab	5.31±0.67c	4.32±1.09 a b	4.89±1.16 bc
Width of fruit (cm)	2.99±0.71 a	3.59±0.43 b	3.86±0.29 b	3.72±0.54 b	3.67±0.32 b
Weight of fruit (g)	10.10±3.81 ab	9.60±3.17 a	11.00±1.70 ab	12.20±3.55 ab	12.90±2.42 b
(MN/AR/002) M <sub>2</sub>					
Plant height (cm)	20.08±2.17 <sup>b</sup>	13.76±2.73 <sup>a</sup>	27.63±4.34 <sup>c</sup>	13.76±3.42 <sup>a</sup>	27.41±5.02 <sup>c</sup>
Number of leaves/plant	108.00±23.83 <sup>b</sup>	125.60±173.03 <sup>ab</sup>	158.40±35.85 <sup>ab</sup>	77.70±43.71 <sup>a</sup>	190.50±91.76 <sup>c</sup>
Number of fruits/plant	14.9±6.45 <sup>a</sup>	16.8±12.54 <sup>b</sup>	18.0±6.57 <sup>c</sup>	18.5±7.58 <sup>c</sup>	17.5±8.10 <sup>b</sup>
Number of seeds/fruit	33.40±11.24 <sup>a</sup>	39.10±13.14 <sup>a</sup>	48.30±14.41 <sup>b</sup>	42.60±21.43 <sup>ab</sup>	44.10±17.57 <sup>ab</sup>
Length of fruit (cm)	3.80±0.81 a	4.13±0.51 ab	5.01±0.67c	4.11±0.99 ab	4.71±1.09 bc
Width of fruit (cm)	2.66±0.41 a	3.26±0.33 b	3.53±0.22 b	3.49±0.49b	3.34±0.32 b
Weight of fruit (g)	48.63±16.09 <sup>a</sup>	55.20±10.39 <sup>a</sup>	63.37±12.29 <sup>b</sup>	49.69±22.49 <sup>a</sup>	56.65±15.17 <sup>a</sup>

Values are mean±SD. Values followed by the same letter(s) within the same row do not statistically differ at the 5% level according to LSD, analysed for the accession.

## REFERENCES

- Abdullahi, M., Muhammad, G. and Abdulkadir, N.U. (2003). *Medicinal and economic plants of Nupeland*. Jube-Evans Books and Publications, Bida, Nigeria, 276 pp.
- Adamu, A.K., Clung, S.S. and Abubakar, S. (2004). Effects of ionizing radiation (gamma-rays) on tomato (*Lycopersicon esculentum* L.). *Nigerian Journal of Experimental and Applied Biology*, 5(2): 185-193.
- Adamu, A.K. and Aliyu, H. (2007). Morphological effects of sodium azide on tomato (*Lycopersicon esculentum* Mill.). *Science World J.* 2(4), 9-12.
- Ado, S.G. (1999). Potentials of native and exotic pepper germplasm in Nigeria: An exploitable resource in the next millennium. *Commemorative publication on the Silver Jubilee of the Genetic Society of Nigeria*. pp. 22-36.
- Asmahan, A.M. and Nada, A. (2006). Effect of gamma irradiation and sodium azide on some economic traits in Tomato. *Saudi J Biol Sci* 13(1), 44-49.



- Awan, M. A., Konzak, C. F., Rutger, J. N. and Nilan, (1980). Mutagenic effects of sodium azide in rice. *Crop Sci.*, 20: 663-668.
- Bertagne-Sagnard, B., Fouilloux, G. and Chupeau, Y., (1996). Induced albino mutations as a tool for genetic and cell biology in flax (*Linum usitatissimum*). *J. Exp Bot* 47, 189-194.
- Bosland P.W. (1992). Chiles: a diverse crop. *Hort. Techn.* 2: 6-10.
- Bosland, P.W. (1996). *Capsicum*: Innovative uses of an ancient crop. In: *Progress in New Crops*, Janick, J. (ed.) ASHS Press, Arlington, VA, pp. 479-87.
- Bosland, P.W. (1994). Chiles: history, cultivation, and uses. In: Charalambous, G. (ed.), *spices, herbs and edible fungi*. Elsevier Publ., New York, pp. 347-366.
- Bosland, P.W. and Vostava, E.J. (2000). Peppers: Vegetable and Spice *Capsicum*. CABI Publishing, Wallingford, UK, UK. 204 pp.
- Casali, V.W.D. and Couto, F.A. (1984). Origem botânica de *Capsicum*. *Inf. Agrop.* 113: 8-10.
- David, T. (2010). All you wanted to know about induced mutations in crop breeding. *Bulletin of Biofortified*.
- Elangovan, M. (2001). Gamma radiation induced mutant for improved yield components in sunflower. *Mutat. Breed. Newslett.* 45: 28-29.
- Fahad, A., (2009). Effects of sodium azide on growth and yield traits of *Eruca sativa* (L.). *World Applied Sci J.* 7(2), 220-226.
- Fahad, A. and Salim K., (2009). Mutagenic effects of sodium azide and its application in crop improvement. *World Applied Sci J* 6 (12). 1589-1601.
- Falusi, O.A. (2007). Germplasm collection of peppers (*Capsicum* spp.) in Nigeria. *Res. Crops* 8(3). 765-768.
- Falusi OA, Daudu OA, Teixeira da Silva JA (2012). Effects of fast neutron irradiation on agronomic traits of Nigerian pepper (*Capsicum annum* L.). *Eur. J. Hort. Sci.*, 77(1): 41-45.
- Falusi, O.A. and Morakinyo, J.A. (2001). Pollen and hybridization studies in some Nigerian species of peppers. *Nigerian J. Tech. Edu.* 1 & 2, 40-43.
- Food and Agricultural Organization. 2009: Induced plant mutations in the genomic era. Publication of the Food and Agricultural Organization of the United Nations (FAO) Rome.
- Germplasm Resources Information Network (GRIN). (2009). *Capsicum* L. United States Department of Agriculture. <http://www.ars.grin.gov/cgi> (last accessed 21 August, 2011).
- Gill, L.S., (1992). *Ethnomedicinal uses of plants in Nigeria*. Uni. Ben. Press Benin City.
- Hegazi, A.Z. and Hamildeldin, N. (2010). The effect of gamma irradiation on enhancement of growth and seed yield of okra [*Abelmoschus esculentus* (L.) Moench]] and associated molecular changes. *J Hort. For* 2(3), 38-51.



- Hutchinson, J. and Dalziel, J.M. (1963). *Floral of west tropical Africa II* Crown Agents, London, 533 pp.
- Islam, S.M.S., Kabir, G., Shahquzzaman, G.M. and Khan, M.R. (1994). Effects of gamma rays and temperature treatment on germination, plant height and morphological abnormalities of *Crotalaria juncea* L. *Journal of Biological Science*, 2: 31-37.
- Khan, M.A. and Aslam, S. (1982). Effects of gamma rays on the morphology and growth of *Crotalaria juncea* and *C. sericea*. *Journal of Asiatic Society Bangladesh (SC)*, 8: 71-77.
- Mady, E.A., Uguru, M.I. and Ugwoke, K.I. (2005). Interrelations of growth and disease expression in pepper using principal component analysis. *Proc. 30<sup>th</sup> Annual National Conference of Genetic Society of Nigeria*. pp. 55-59.
- Mensah J. K., Obadoni, B. O., Akomeah, P. A., Lkhajiagbe, B., and Ajibolu J. (2007). The effect of sodium azide and colchicine treatments on morphological and yield traits of sesame seed (*Sesame indicum* L.). *Afr. J. Biotechnol.* 6(5): 534-538.
- Misra, R. C., Mohapatra, B. D., and Panda, B. S. (2001). High yielding mutants of blackgram variety PH-25. *Mutat. Breed. Newslett.* 45: p. 39.
- Misra, R.C. (2001). Productive mutants of niger. *Mutat. Breed. Newslett.* 45: 42-43.
- Moscone, E.A., Scaldaferrro, M.A, Grabiele, M. and Cecchini, N.M. (2007). The evolution of chilli peppers (*Capsicum* - Solanaceae): a cytogenetic perspective. *Acta Hort.* 745: 137-170.
- Poornananda, M.N. and Hosakatte, N.M. (2009). The effect of gamma and ethylmethyl sulphonate treatments on agronomical traits of niger (*Guizotia abyssinica* Cass.). *African J Biotech* 8(18), 4459-4464.
- Reifschneider, F.J.B. (2000). *Capsicum: Pimentas e Pimentões no Brasil*. Embrapa Comunicação para Transferência de Tecnologia. *Embrapa Hortaliças*, Brasília.
- SAR (2005). Final Safety Analysis Report of Nigeria Research Reactor-1. CERT Technical Report- CERT/NIRR-1/FSAR-01.
- Schippers, R.R. (2000). African indigenous vegetables: An overview of the cultivated species. Natural Resources Institute/ ACP-EU Technical Centre for Agricultural and Rural Cooperation, Chathan, UK, pp. 122133.
- Shah, T.M., Mirza, J.I., Haq, M.A and Atta, B.A. (2008). Induced genetic variability in chickpea (*C. arietinum*). Comparative mutagenic effectiveness and efficiency of physical and chemical mutagens. *Pakistan J. Bot.* 11 (2): 173177.
- Shin, J.M., Kim, B.K., Seo, S.G., Jeon, S.B., Kim, J.S., Jun, B.K... & Kim, S.H. (2011). Mutation breeding of sweet potato by gamma-ray radiation. *African J. Agric. Res.* 6, 14471454.
- Simmond, N.W. (1976). *Evolution of Crop Plants*. Longman Co. Ltd, London, pp. 265268.



- Sodkiewicz T, Sodkiewicz W (1999). Effectiveness of fast neutron irradiation for the stimulation and induction of genetic changes in soybean (*Glycine max*) genome. *Int. Agrophysics*, 13: 503-507.
- Swaminathan, M.S. (1998). *Crop production and sustainable food security. In: Crop Productivity and Sustainability. Shaping the future* (ed V.L. Chopra, R.B. Singh, and A. Varma) New Delhi: IBH, p. 318.
- Zhang, W., Endo, S., Ishikawa, M., Ikeda, H., and Hoshi, M. (2002). Relative biological effectiveness of fission neutrons for producing micronuclei in the root-tip cells of onion seedlings after irradiation as dry seeds. *J. Radiat. Res. (Tokyo)*, 43(4): 397-403.