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Induced Genetic Variability for Morphological and Yield Parameters in *Capsicum annum* and *Capsicum frutescens*

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ABSTRACT

Mutations were induced in (*Capsicum annum* var *accuminatum* and *Capsicum frutescens* var *baccatum*) using Fast Neutron Irradiation (FNI) from Americium-Beryllium source with a flux of 1.5×10^4 n cm^{-2} $s^{-1.}$ The pepper seeds were irradiated for 0, 30, 60, 90 and 120 minutes before they were sown – with their respective controls – in order to assess the effects of the different irradiation treatments on the percentage survival, days to maturity, plant height, number of fruits per plant, number of seeds per fruits, height of fruit, weight of fruit and width of fruit in the M_1 generations. Results showed that all irradiation periods caused leaf abnormalities which essentially may affect productivity negatively. There was also pronounced variation in yield and other morphological parameters, suggesting the possibility of evolving higher yield variants of pepper through proper selection.

1. Introduction

Capsicum is a genus of flowering plants in the nightshade family; Solanaceae (GRIN, 2009). The genus consists of over one hundred (100) species and even more botanical varieties (Ado, 1999; Falusi, 2007). These include five domesticated species namely: *Capsicum annuum, C. frutescens, C. baccatum, C. chinense* and *C. pubescens*; all believed to have originated from the New World (Mcleod *et al.*, 1982; Bosland, 1994). Phylogenetic relationships between species have being investigated using biogeographical, morphological, hybridization and genetic data (Walsh and Host, 2001; Tewksbury *et al.*, 2006; Pickersgill, 1971). *C. annuum* and *C. frutescens* are the most recognized species grown in commercial quantities all over Nigeria (Falusi and Morakinyo, 2001; Mady *et al.*, 2005). These two species form an important ingredient in people's diet all over the world (GRIN, 2009), due to the pungency properties of the fruits, resulting from its high concentration of capsaicinoid alkaloid (Bosland and Vostava, 2000). In addition, *Capsicum* is a rich source of vitamins A and C (ascorbic acid) (Gill, 1992; Ado, 1999). *Capsicum* fruits are also popular as food spices, colouring agent, as well as pharmaceutical ingredients (Bosland, 1996). In Afri-

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can medicine, it is used in treating sore throat (Abdullahi *et al*., 2003).

Mutations are known to enhance the genetic variability of crop plants as the variability at species level has reached the ceiling due to high breeding intensity and rapid erosion of genetic resources (Poornananda and Hosakatte, 2009). Since spontaneous mutations occur at very low frequency, induced mutations facilitate the development of improved varieties at a swifter rate (Maluszynski, 1990). Besides the vital role in plant breeding programs, a new role of induced mutations in releasing of gene silencing in transgenic plants has been reported (Bhatia, 1999). 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 Anis, 2001), *Vigna mungo* (Misra *et al*., 2001), *Helianthus annuus* (Elangovan, 2001) *Cajanus cajan* (Ravikesavan *et al*., 2001), *Sesame indicum* (Mensah *et al*., 2007), *Guizotia abyssinica* (Misra, 2001). These reports show that mutagenesis is a potential tool to be employed for crop improvement.

This study aimed to use fast neutron irradiation to induce genetic variability on yield and morphological parameters in *C. annum* var *accuminatum* and *C. frutescens* var *baccatum* .

2. Materials and Methods

Fresh fruit of two pepper varieties (50 fruits each) were obtained from a local farmer in Minna, Niger State, Nigeria. The fruits were kept in separate polythene bags. The varieties were identified as *Capsicum annum* var. *accuminatum* Fingerh and *C. frutescens* var. *baccatum* L. (Table 1) using taxonomic aid provided by Simmond (1976), as well as morphological descriptions of Hutchinson and Dalziel (1963), Schippers (2000) and Abdullahi *et al*., (2003). Each fruit of the *Capsicum* accessions was cut open; the seeds were removed, kept separately and sun-dried for 8 hours. The sun-dried seeds were tested for viability using the floatation method before Fast Neutron Irradiation (FNI) treatment. 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 1.5×10^4 n $cm^{-2} s^{-1}$ for five different irradiation exposure periods (IEPs): 0, 30, 60, 90, and 120 min.

Treated seeds (100 from each treatment) were then planted in nursery trays to obtain seedlings, which were transplanted into 3.5 litre plastic pots containing garden soil, at a rate of three seedlings/pot after 4 weeks in the nursery. No fertilizer was applied although, when the crop began to flower, an insecticide was applied to prevent insect-borne diseases. The planted seeds were watered once daily between 5.00-6.30 pm using borehole water. Each treatment was replicated four times using a completely randomized design (CRD).

Data were collected from 15 plants for each accession 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 M1 (M = first filial mutant) generation. Data was analysed using analysis of variance (ANOVA) and Least Significant Difference (LSD) 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 some selected parameters.

3. Results and Discussion

All the plants produced from the non-irradiated seeds (the control) produced normal leaves (Plates. 1A & B). However, this was not the case among plants whose seeds were irradiated. The different irradiation periods caused different leaf abnormalities such as small leaves with dented margin, leaves with invaginated margins, with inverted margins, leaves that turned bifoliage and with bifurcated apices (Plate 1C-H). These observed leaf morphological abnormalities are indications that FNI may affect productivity negatively. Shah *et al*., (2008), and Islam *et al*., (1994) reported similar leaf morphological abnormalities in *Crotalaria saltiana* and *C. juncea* treated with ionizing radiation.

Pronounced variations were observed in plant height, number of leaves/plant, fruits/plant, and seeds/ fruit, and weight, length and width of fruits for the two varieties (Table 2, 3 and 4). This is an indication that FNI is capable of producing significant changes in the agronomic traits and yield parameters of pepper plants. 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. However, Poornananda and Hosakatee (2009) reported a decrease in plant height of niger treated with gamma rays and sodium azide. FNI may therefore be used to induce beneficial variability which can be incorporated in conventional breeding for the improvement of pepper.

Table 1 Description of pepper (*C. annum* var *accuminatum* and *C. frutescens* var *baccatum*) plants that were used in this study.

LOCAL NAMES	SOURCE	BOTANICAL NAMES	DESCRIPTIONS
'ATA SHOMBO'	MINNA	*C. annuum* var *accuminatum* Fingerh	Medium sized annual plant, long pointed and pendant fruits with hot taste, one pedicel per node.
'ATAWEWE'	MINNA	*C. frutescens baccatum* L.	Large perennial shrub, small pointed fruit with very hot taste, 2-4 pedicels per node.

A

C

B

D

E

F

G **H**

Plate 1: **A** and **B.** Normal leaves of *C annum* var *accuminatum* and *C frutescens* var *baccatum* . **C.** Leaf with bifurcated apex in *C frutescens* var *baccatum* (120 min IEP) **D.** A leaf with another leafy outgrow at the petiole in *C frutescens* var *baccatum* (90 min IEP) **E**. Leaves showing invaginated margins in *C annum* var *accuminatum* (120 min IEP) **F**. The leaf that turned bifoliage in *C annum* var *accuminatum* (30 min IEP) **G**. Leaf with bifurcated apex in *C annum* var *accuminatum* (60 min IEP) **H**. Leaves of *C frutescens* var *baccatum*: From the left, leaf with curved apex, the middle leaf is small with dented margin, the third leaf has invaginated margin. (60 minutes irradiation exposure).

Table 2 Effects of fast neutron irradiation on survival percentage and plant height at four periods after germination of *Capsicum annum* and *C. frutescens.*

	Germination % in pots	**Plant Height (cm) 2 weeks**	**4 weeks**	**8 weeks**	**Maturity**
C. annum* var *accuminatum					
Control	98.00^{c}	12.51^{ab}	19.11^{a}	25.51^{a}	44.20^{a}
30 minutes	50.00^{b}	11.87^{a}	20.77^{a}	30.23^{b}	45.27^{a}
60 minutes	51.00^{b}	13.38^{a}	24.92^{b}	31.83^{bc}	52.86^{ab}
90 minutes	51.00^{b}	13.53^{ab}	25.36^{b}	31.76^{bc}	50.23^{ab}
120 minutes	37.00^{a}	15.51^{b}	28.11^{b}	36.21^{bc}	59.93^{b}
	-0.8156				
C. frutescens* var. *baccatum					
Control	100.00^{d}	9.58^{a}	13.44^{a}	19.61^{a}	41.50^{ab}
30 minutes	44.00^{c}	11.04^{b}	15.84^{ab}	22.88^{ab}	37.77^{a}
60 minutes	13.00^{a}	9.72^{a}	14.69^{a}	21.83^{a}	38.84^{a}
90 minutes	26.00^{b}	9.79^{a}	15.94^{ab}	22.96^{ab}	34.84^{a}
120 minutes	40.00^{c}	11.34^{b}	17.84^{ab}	24.88^{ab}	45.85^{ab}
	r = -0.6525				

Means followed by the same letter (s) within the same column do not statistically differ at 5% level tested by LSD.

Table 3: Effects of fast neutron irradiation on the number of leaves per plant at four periods after germination of *Capsicum annum* and *C. frutescens*.

	2 weeks	**4weeks**	**8 weeks**	**maturity**
C. annum* var *accuminatum				
Control	13.43[a]	18.64[a]	33.57[a]	129.93[a]
30 minutes	14.90[a]	19.90[a]	77.90[b]	132.70[a]
60 minutes	14.50[a]	26.10[b]	86.20[b]	173.00[b]
90 minutes	17.60[b]	23.90[b]	83.60[c]	161.80[b]
120 minutes	20.88[c]	31.50[c]	80.36[b]	230.80[c]
C. frutescens* var *baccatum				
Control	10.80[a]	13.80[a]	38.10[a]	112.00[a]
30 minutes	12.90[ab]	18.00[ab]	52.80[b]	103.80[a]
60 minutes	10.82[a]	14.64[a]	48.18[b]	105.64[a]
90 minutes	12.10[a]	19.60[ab]	53.50[b]	100.10[a]
120 minutes	12.50[ab]	20.80[ab]	63.40[c]	156.90[b]

Means followed by the same letter (s) within the same column do not statistically differ at 5% level tested by LSD.

Table 4: LSD of the of the effects of Fast neutron irradiation on the mean performance of some yield parameters of the two varieties of pepper

	Irradiation time (min)				
Characters	**0**	**30**	**60**	**90**	**120**
C annum* var *accuminatum					
Plant height at maturity(cm)	44.20[a]	45.27[a]	52.86[ab]	50.23[ab]	59.93[b]
Number of leaves per plant at maturity	129.93[a]	132.70[a]	173.00[b]	161.80[b]	230.80[c]
Number of fruits per plant	8.00[a]	15.3[b]	14.8[b]	14.3[b]	15.8[b]
Number of seeds per fruit	120.80[a]	105.20[a]	108.20[a]	110.30[a]	125.80[b]
Length of fruit (cm)	7.26[a]	7.54[a]	6.94[a]	7.67[a]	8.20[ab]
Width of fruit (cm)	1.93[a]	1.97[a]	1.90[a]	2.15[a]	2.28[ab]
Weight of fruit (g)	7.70[a]	8.40[a]	8.70[a]	9.10[ab]	10.20[ab]
***C. frutescens* var baccatum**					
Plant height at maturity (cm)	41.50[ab]	37.77[a]	38.84[a]	34.84[a]	45.85[ab]
Number of leaves per plant at maturity	112.00[a]	103.80[a]	105.64[a]	100.10[a]	156.90[b]
Number of fruits per plant	30.60[a]	40.80[a]	33.90[a]	53.00[ab]	48.10[ab]
Number of seeds per fruit	31.00[a]	32.70[a]	24.00[a]	35.80[ab]	32.40[a]
Length of fruit (cm)	5.21[a]	5.33[a]	5.73[a]	5.25[a]	4.45[a]
Width of fruit (cm)	0.80[a]	0.88[a]	0.80[a]	0.89[a]	0.99[a]
Weight of fruit (g)	1.30[a]	0.90[a]	1.01[a]	1.00[a]	1.15[a]

Means followed by the same letter (s) within the same row do not statistically differ at 5% level tested by LSD.

4. References

1. Abdullahi, M., Muhammad, G. and Abdulkadir, N.U. (2003). *Medicinal and economic plants of Nupeland*. Jube-Evans Books and Publications, Bida, Nigeria, 276 pp.
2. Adamu, A.K. and Aliyu, H. (2007). Morphological effects of sodium azide on tomato (*Lycopersicon esculentum* Mill.). *Science World J*. 2(4), 9-12.
3. 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.
4. 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.
5. Awan, M. A., Konzak, C. F., Rutger, J. N. and Nilan, (1980). Mutagenic effects of sodium azide in rice. *Crop Sci*., 20: 663-668.
6. Bhatia, C. R. (1999). Release of gene silencing in transgenics-a new role for induced mutations. *Mutat. Breed. Newslett*. 44: 3-5.
7. 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.
8. 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.
9. Bosland, P.W. and Vostava, E.J. (2000). Peppers: Vegetable and Spice *Capsicum*. CABI Publishing, Wallingford, UK, UK. 204 pp.
10. Elangovan, M. (2001). Gamma radiation induced mutant for improved yield components in sunflower. *Mutat. Breed. Newslett*. 45: 28-29.
11. Fahad, A., (2009). Effects of sodium azide on growth and yield traits of *Eruca sativa* (L.). *World Applied Sci J*. 7(2), 220-226.
12. Falusi, O.A. (2007). Germplasm collection of peppers (*Capsicum* spp.) in Nigeria. *Res. Crops* 8(3), 765-768.
13. 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.
14. Germplasm Resources Information Network (GRIN). (2009). *Capsicum* L. United StatesDepartment of Agriculture. http://www.ars.grin.gov.cgi (last accessed 21 August, 2011).
15. Gill, L.S., 1992. *Ethnomedicinal uses of plants in Nigeria*. Uni. Ben. Press Benin City,
16. 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 Hortic For* 2(3), 38-51.
17. Hutchinson, J. and Dalziel, J.M. (1963). *Floral of west tropical Africa II* Crown Agents, London, 533 pp.
18. Islam, S.M.S., Kabir, G., Shahiquzzaman, 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.
19. Mady, E.A., Uguru, M.I. and Ugwoke, K.I. (2005). Interrelations of growth and disease expression in pepper using principal component analysis. *Proc. 30th Annual National Conference of Genetic Society of Nigeria*. pp. 55-59.
20. Maluszynski M (1990). Gene manipulation in plant improvement. Vol. 2. Gustafsson JP (eds) Plenum press, New York.
21. Mcleod, M.T., Sheidon, I.G. and Eshbaugh, W.H. (1982). Early evolution of chilli peppers. *Econ. Bot.*, 36, 361-368.
22. 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.
23. 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.
24. Misra, R. C. (2001). Productive mutants of niger. *Mutat. Breed. Newslett*. 45: 42-43.
25. Pickersgill, B. (1971). Relationships Between Weedy and Cultivated Forms in Some Species of Chili Peppers (Genus *Capsicum*). *Evolution* 25 (4): 683-691.
26. 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.
27. Ramesh, B., Prasad, B. K., and Singh, V. P. (2001). Semi dwarf, high yielding and high protein mutants

in barley. *Mutat. Breed. Newslett.* 45: 26-27.

28. Ravikesavan, R., Kalaimagal, T., Rathnaswamy, R. (2001). An extra early mutant of pigeonpea. *Mutat. Breed. Newslett.* 45: 19-20.
29. Sakin, M. A., Yildirim, A. (2004). Induced mutations for yield and its component in durum wheat (*Triticum durum* Defs.). *Food Agric. Environ*. 2(1): 285-290.
30. 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. 122-133.
31. Shah, A. B. S., Ud-Deen, M. M., Naz, A., Sarker, J. K. and Kabir, G. (2008). Post-irradiation ageing effect on morphological characters of *Crotalaria saltiana*. *Journal of Biological Sciences*, 16: 89-93.
32. Simmond, N.W. (1976). Evolution of Crop Plants. Longman Co. Ltd, London. pp. 265-68.
33. Singh, S., Richharia, A. K., Joshi, A. K. (1998). An assessment of gamma ray induced mutations in rice (*Oryza sativa* L.). *Indian J. Genet.* 58(4): 455-463.
34. Tewksbury, J. J., Manchego, C., Haak, D. C., and Levey, D. J. (2006). Where did the chilli get its spice? *Journal of Chemical Ecology*, 32 (3): 547-564.
35. Wani, A. A. and Anis, M. (2001). Gamma rays induced bold seeded high yielding mutant in chickpea. Mutat. Breed. Newslett. 45: 20-21.
36. Walsh, B. M. and Hoot, S. B. (2001). Phylogenetic Relationships of *Capsicum* (Solanaceae) Using DNA Sequences from Two Noncoding Regions: The Chloroplast atpB-rbcL Spacer Region and Nuclear waxy Introns. *International Journal of Plant Sciences*, 162 (6): 1409-1418.