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Mutagenic Effects of Gamma Rays on Young, Middle and Old age Stem Cuttings of Poplar (*Populus deltoides*) in Spring Season

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Abstract: ***Three different age group stem cuttings of P.deltoides viz. 6 month (young age), 12 month (middle age), and 24 (old age) months have been selected in the present investigation to investigate their regeneration sensitivity after treated with different continuous and fractionated doses of gamma rays. Treated cuttings have been planted in the first week of March when mother plant has got started to sprout after winter dormancy. Maximum sprouting percentage (66.6%) has been recorded under 2KR-C level in young age stem cuttings than the middle and old age. Under fractionated treatment 500r-F was found more effective in sprouting percentage and it was recorded 60% sprouting. Combination effects of gamma rays with 500ppm showed antagonistic effects for different parameters in all the three age treated cuttings. Different parameters for lateral shoot growth and mortality percentages of the sprouted bud have also been observed during regeneration. At 2KR-C level maximum shoot length 70.5 cm have been recorded and this is followed by 4KR-C level 58.5cm. The area of leaf was favoured higher at lower continuous doses of all the treated cuttings.***

Keywords: Gamma rays, Irradiation, Mutation, Stik, mortality

1. Introduction

Radiation breeding of woody perennials is supposed to be most efficient way to make specific improvements in the commercially cultivated varieties (Nishida et. al. 1967). The capability of ionizing radiation to bring out mutation at much higher frequencies than would occur naturally has been of interest to the plant breeders. This is because radiation can be used to provide material of much wider genetic variability for selection in respect of the breeding programme (Broertjes and Harten, 1978). The cuttings provide a versatile and convenient material for studies of the influence of ionizing radiation and posses vegetative cells essential for mutation breeding studies. For these reason, the effects of ionizing radiations on the seeds have been studied for a number of plants (IAEA,1977). Poplar *(P.deltoides)* has deciduous characters which allow winter cropping with only marginal effects on agricultural crops. It has been introduced in a large scale all over the country from seacoast to Himalaya foot hills because of rapid growth and high yield in stipulated time and ability to survive under wide edapic and climatic condition (Piarelal 1991, Singh et al. 1988). Induction of useful mutation as a means of cultivar improvement has a special appeal in plant breeding and induced mutation has played a proven role in the improvement of different plant species. Present investigation was undertaken to study the effect of gamma rays and its combination effect with Stik 500ppm on cell division of buds and its role in morphogenetic changing in irradiated cuttings.

2. Materials and Method

Three age group of stem cuttings i.e. 6 month (dia.1.5 cm), 12 month (dia. 2.0 cm) and 24 month (dia. 2.5cm) have been taken from the mother plant. Cuttings of *P.deltoides* irradiated with acute gamma rays at 500r, 1KR, 2KR and 4KR level. The doses were given as follows- continuous doses (c) –cuttings were irradiated continuous by given desired dose uninterruptedly. Fractionated doses (f) –this was done by fractionating the dose in equal part at an interval of 24 hours. Gamma irradiation was done at radio isotope laboratory, at Forest Research Institute Dehradun Uttarakhand India. Based on the data belonging to different parameters, reduction percentage over the control was determined and LD-50 values were calculated. Irradiated cuttings were planted in poly pots and only two cuttings were potted in each pots of the size of 28×18 cm approximately and were filled with garden soil. Cuttings were kept with lower ends 8cm. deep at the av. maximum-minimum temperature ranging between 24^0c-14^0c. Treated cuttings have been potted in the month of March. The upper end of cuttings was covered with moist cotton plug to avoid drying specimens received distilled water treated as control. To study the protagonestic and antagonistic effects of gamma rays some irradiated cuttings presoaked with Stik 500ppm for 72 hours and another set of experiment have been carried out simultaneously. Growth hormone Stik have been taken on its 500ppm level for doing combination effects.

3. Results and Discussion

The cell function, structure and the gross effects on chromosomes and on mitosis have been influenced by different continuous and fractionated doses gamma rays. Data on sprouting percentage and subsequent growth of cuttings are presented in Fig. 1,2 and Table-1&2. Under C treatment maximum (66.6%) sprouting have been recorded at 2KR-C level in young age cuttings which is followed by 500r-F (59.5%) in middle age cuttings after 43 days of experimentation but in middle age cuttings lower C and F doses showed better sprouting percentage in comparison to old age cuttings. For analyzing the synergistic effects of

gamma rays with stik500ppm most of the treatments of old age cuttings showed antagonistic effects for the sprouting percentage. Mostly all the lower C and F doses favoured better sprouting percentage than the higher doses. Gamma rays treatments were observed as it breaks the dormancy of the buds at lower doses. Kapoor & Sharma (1983) and Chauhan & Singh (2007) have been made an attempt to justify gamma rays as a source of inducing sprouting of cuttings, rooting and their survival in Poplars. Stik 500ppm with irradiated cuttings showed protagonistic effects for sprouting percentage in young and middle age cuttings. The hormonal control of bud sprouting of in the twig of cuttings was put forth by Michener (1942). All the lower Continuous and Fractionated doses enhanced the growth rate of lateral branch and the leaf formation (Table-1). Fractionated doses showed the better shoot growth behavior than the continuous doses. The maximum length (15.9cm±.31) of lateral branch has been recorded at 1KR-F level in middle age in comparison to young and old age. Continuous doses retarded the growth of lateral branch and it was minimum 3.8cm±.11) at 4KR-C level. In regards to change in various morphological parameters, the lower C and F doses were found to be most effective. All molecules can be altered by a sufficiently large amount of ionizing radiation. It is not surprising that massive doses of radiation can break and destroy the structural components of a cell produce cell death. Exposure of smaller amounts of radiation can result in cell death, but by less obvious mechanisms. In most of the cases, the higher doses failed to bring out any prominent effects as compared to the lower doses. The apical shoots get damaged and its original morphology and color also gets altered. Iqbal (1972) also observed that at all stages of seedling development of *Capsicum annum* and at all the exposure (1-10KR), terminal leaves consistently showed the maximum percentage of damaged cells. The effects of Stik might be due to rapid channeling of mineral nutrients and photosynthesis towards apical meristem. The hormones induce cell division, cell elongation and cell differentiation by regulating the expression of transcription factors and cell type specific genes (Johri & Mitra 2001). At 2KR-C dose in young age cuttings and 4KR-C in old age cuttings have been found more effective in leaf formation but the maximum area (4308.0±13.2cm^2) of the leaf was recorded under 500r-C level in old age cuttings. For combination effects of gamma rays with Stik500ppm in middle age cuttings have favored maximum length (22.5cm±3.97) of lateral branch and leaf area (4626±17.7cm^2) at 1KR-F level (Table-2). Sensitivity of growth pattern of petiole and midrib of the leaf has been observed under the treatments of different C & F doses of gamma rays. All the higher C &F doses exhibited stimulation in the length of petiole and midrib in all the three age groups of cuttings. Combination effects of gamma rays with Stik 500ppm showed the protagonistic effects for the size of the leaf. Lower C & F doses with synergistic effects caused reduction in leaf formation but the area of the leaf was increased by higher doses and it was recorded maximum at 2KR-F (3482.0±9.79). The reduction in leaf numbers, size was also recorded in Gladiolus (Banerji et al. 1994). The decreased leaf area could be attributed to deleterious effects of radiation at different continuous and fractionated doses in different age groups of cuttings. Joel et. al. (1997) tested the effects of physical mutagen on ground nuts and recorded negative trends for leaflet length and width. The morphological abnormalities i.e. unequal pairs of leaves, variegated leaves and leaves with black margins on tips was also recorded in leaves. The pinule formation in irradiated plants under C&F treatments was also found in *Albizia lebbek* (Singh & Paliwal 1987). Significant effects of gamma rays were observed for different parameters i.e. plant height, length of lateral branch and size of the leaf. Significant difference was found among the dose and days of the cuttings in different parameters (ANOVA, Table 3). The radiation treatments caused various growth inhibitions i.e. reduction in plant height, internodes length and leaf size were some of the most conspicuous effects. The vegetative plants raised from irradiated cuttings were reduced vigor as reflected by their stunt growth. In the buds of *Betula*, increase in the inhibition contents are detectable after as few as two short photoperiods and decrease after two long photoperiods (Kawase, 1961). The mortality percentage of the sprouted buds has been recorded. Mutagenic capabilities of radiation when absorbed in the living material, ionization and excitation of molecules of the absorbing tissue take place almost instantly (Sparrow, 1962). Induced mutation contribute primarily by increasing genetic variability and in addition to their direct use, induced mutation is an alternative to introduction as a source of germplasm for tree improvement.

4. Conclusion

Present study indicate that the gamma rays is a potential mutagen for inducing different morpho-physiological changes, breaking dormancy of the buds and changes in chromosomes number or DNA structure which are not physically visible but lead to mutations. It also concluded that Radiation technology can provide an important source of morphogenetic variability so that the plants become more desirable economically and agronomically.

References

[1] Banerji BK, Datta SK & Sharma SC, 1994. Gamma irradiation studies on *Gladiolus* cv. white friendship. J. Nuclear Agric. Bio. 23:127-133.
[2] Broertjes C &Van Harten AM, 1978. Application of mutation breeding methods in the improvement of vegetatively propagated crops. An interpretive literature review (Elsevier Scientific publishing company, Amsterden Oxford, New York.
[3] Chauhan PS & Singh SS, 2007 Sensitivity of *Populus deltoides* young aged stems cuttings (Six months) to gamma rays and its synergistic effects with Kinetin. Indian J. of Agroforestry, Vol.9 no. 1:47-52.
[4] IAEA, 1977. Manual of mutation breeding. Tech report series no. 199, IAEA, Vienna.
[5] Iqbal J, 1972. Effects of acute gamma irradiation on the survival, growth and radio sensitivity of the special meristem of *Capsicum annum* L at different stages of seedling development. Radiat. Bot. 12: 197-204.
[6] John Joel A, Selvarani S, Sumathy P & Rangaswamy M, 1977. Effects of physical mutagen (gamma-rays) on ground nut (*Arachis hypogaea* L). J. Nuclear agric. Bio. 26(2); 116-124.
[7] Johri MM & Mitra Doyal, 2001. Action of plant hormone. Cur. Sci. 80(2):199-205.

[8] Kapoor ML & Sharma VK, 1983. Effects of gamma rays on cuttings of *Populus* species XV International conger. genetics, Dec. 12-12, New Dehli. India.
[9] Kswase M, 1961. Growth substances related to dormancy in *Betula*, proc. Am. Soc. Hort. Sci 78:532-544.
[10] Michener HD, 1942. Dormancy and apical dominance in *Potato* tubers. Am. J. Bot. 29:558-562.
[11] Nishida T, Nakajima K, Obha K, Takata S, 1967. "Radio sensitivity and induction of somatic mutations in woody perennials under chromic gamma ray irradiation". Use of chronic irradiation in mutation breeding, gamma field sym. No. 6 Ohmiyamichi 19-45.
[12] Piarelal, 1991. Improved package of practices for *Popular* under agroforestry. Indian forester 117: 168-177.
[13] Singh K, Ram P, Singh AK & Hussain A, 1988. Poplar (*Populus deltoides* Bartram e x marshall) in forest and agroforestry system. Indian forester 114:814-818.
[14] Singh SS & Paliwal GS, 1987. Sensitivity of *Albzia lebbek* seeds to gamma rays. Indian forester 113:490-500.
[15] Sparrow AH, 1962. The role of cell nucleus in determine radio-sensitivity. Book haven lecture series, no. 17, BNL 766(T-287).

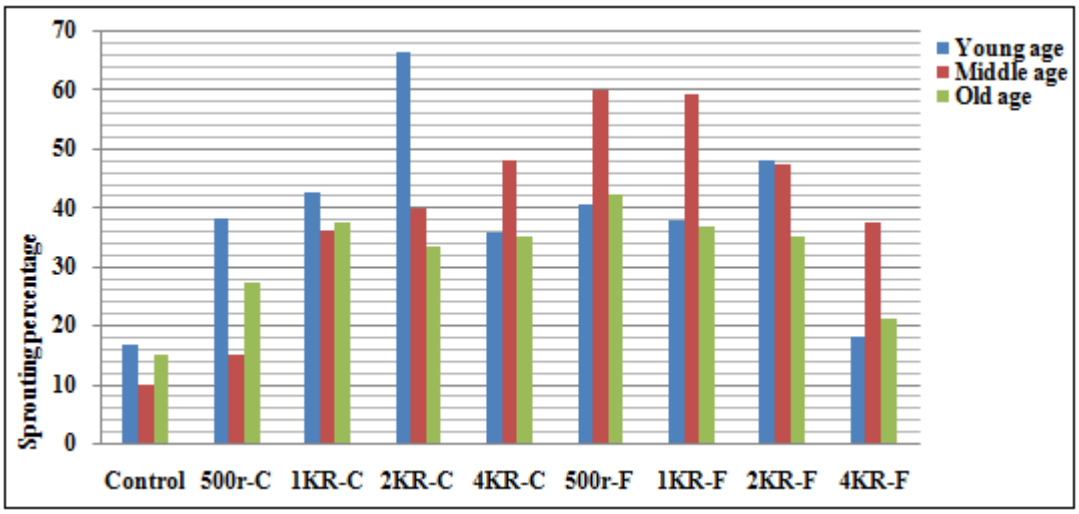
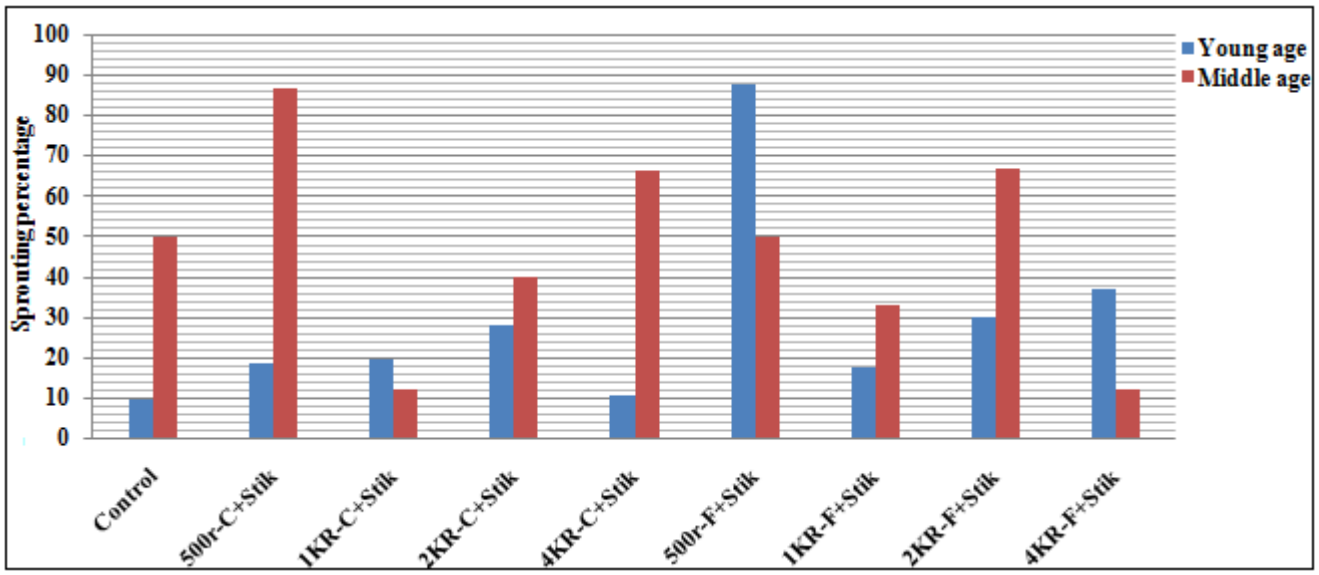


Figure 1: *P. deltoides*-influence of different continuous and fractionated doses of gamma rays on sprouting percentage in young, middle and old age stem cuttings after 43 days.



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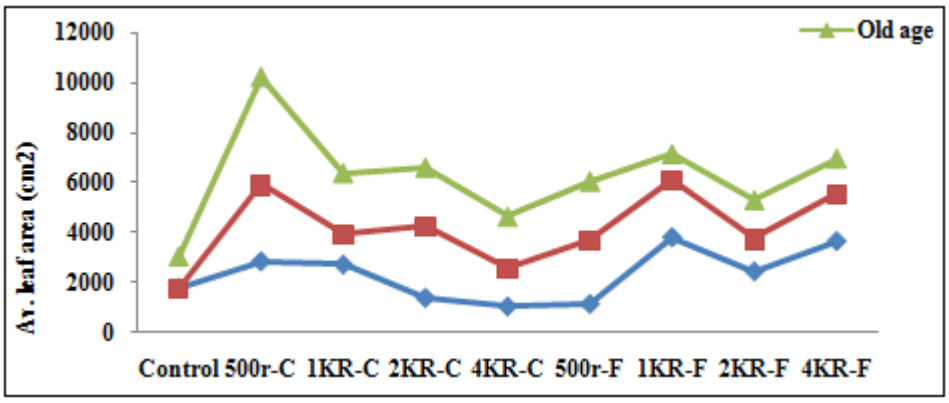

Figure 3: *P. deltoides*-influence of different continuous and fractionated doses of gamma rays on av. leaf area (cm2) in young, middle and old age stem cuttings after 58days.

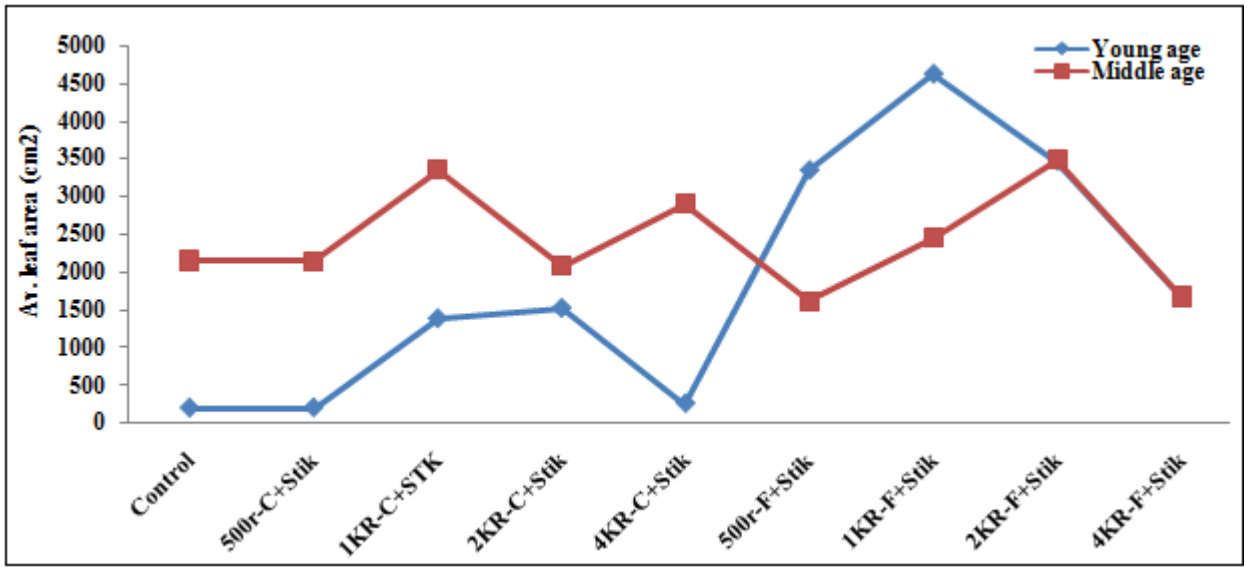

Figure 4: *P. deltoides*- influence of different continuous and fractionated doses of gamma rays on av. leaf area (cm2) in young and middle age stem cuttings after 58days.

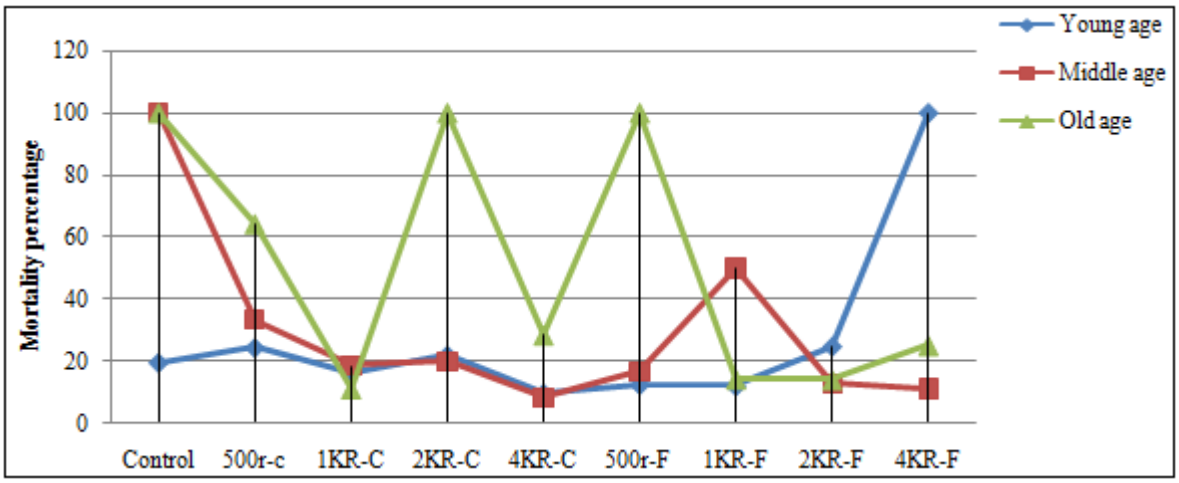

Figure 5: *P. deltoides*-influence of different continuous and fractionated doses of gamma rays on mortality percentage in young, middle and old age stem cuttings after 58days.

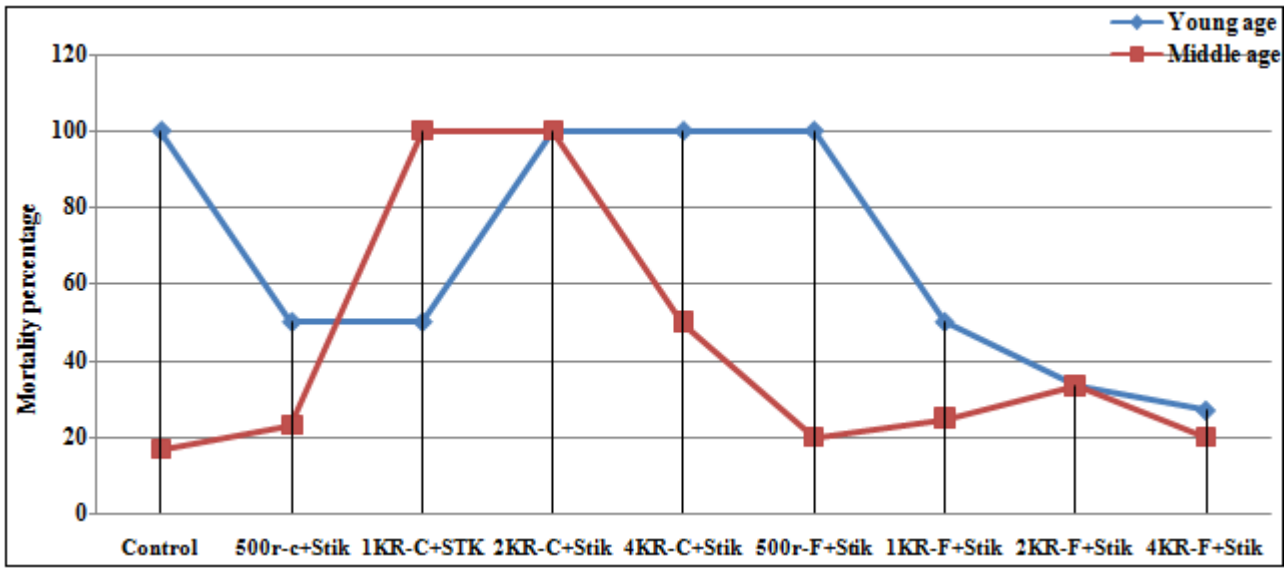

Figure 6: *P. deltoides*-influence of different continuous and fractionated doses of gamma rays on mortality percentage in young and middle age stem cuttings after 58days

Table 1: ***P.deltoides***- Influence of different continuous and fractionated doses of gamma rays on shoot growth behavior in young, middle and old age cuttings after 58 days of potting

Dose/ Treatment	Young age				Middle age				Old age			
	Av. length of lateral branch (1)	Av. no. of leaves formed (2)	Av. length of petiole (3)	Av. length of midrib (4)	(1)	(2)	(3)	(4)	(1)	(2)	(3)	(4)
Control	8.5	11.0	3.0	5.0	6.5	-	-	-	5.2	10.5	2.0	5.0
500rC	4.3	11.0	3.5	7.0	7.1	19.8	5.0	10.5	6.0	19.0	4.5	9.0
1KR-C	8.4	16.1	5.0	7.0	6.2	15.4	3.0	5.5	4.1	16.5	3.5	5.5
2KR-C	5.0	21.7	2.5	5.3	4.5	15.5	3.5	5.0	4.4	2.0	2.5	5.5
4KR-C	3.8	11.8	2.0	2.5	4.6	14.5	3.0	5.0	4.3	21.0	3.0	5.0
500r-F	5.1	15.0	3.0	5.0	5.6	14.8	7.2	10.0	6.6	15.0	4.0	6.0
1KR-F	4.0	7.7	4.5	7.5	15.9	19.5	3.0	6.5	4.7	21.0	3.0	4.5
2KR-F	6.8	11.2	3.6	5.7	9.9	15.5	3.0	4.5	5.9	13.5	3.0	5.5
4KR-F	8.5	14.0	3.5	6.5	4.1	17.0	3.0	6.0	6.8	9.5	3.0	6.0

Table: 1 ***P.deltoides***- combination effects of stik 500ppm with different continuous and fractionated doses of gamma rays on shoot growth behavior in young and middle age cuttings after 58 days of potting.

Dose/ Treatment	Young age				Middle age			
	Av. length of lateral branch (1)	Av. no. of leaves formed (2)	Av. length of petiole (3)	Av. length of midrib (4)	(1)	(2)	(3)	(4)
Control	1.5	3.0	1.5	1.0	4.1	22.0	3.0	5.0
500rC+Stik	2.5	3.0	1.6	1.5	4.2	15.0	3.0	5.2
1KR-C+Stik	8.5	11.5	3.5	5.0	7.5	13.0	4.5	7.2
2KR-C+Stik	4.5	14.5	3.0	6.0	3.5	11.5	3.0	5.5
4KR-C+Stik	2.5	3.0	1.0	2.0	5.2	16.5	3.7	6.2
500r-F+Stik	5.7	23.5	3.5	7.7	11.2	19.0	3.0	4.0
1KR-F+Stik	22.5	17.0	4.2	8.0	8.0	15.5	3.7	6.5
2KR-F+Stik	16.8	12.0	3.0	8.7	5.0	16.0	4.5	8.0
4KR-F+Stik	3.8	13.5	3.0	5.0	5.0	9.0	3.2	5.0

Table 3: (a) Analysis of variance for different parameters (ANOVA). Young, middle and old age cuttings.

Source of variation	SS	DF	MSS	Calculated VR	Tabulated VR
Av. length of lateral branch					
Dose	38.4	8	4.8	0.679	2.09
Days	15.18	2	7.59	1.32	3.24
Error	113.10	16	7.06	-	-
Av. no. of leaves formed					
Dose	201.51	8	25.18	0.793	2.09
Days	9.05	2	4.52	0.142	3.24
Error	507.65	16	31.72	-	-
Av. length of petiole					
Dose	20.26	8	2.53	1.974	2.09

Days	0.3429	2	0.71	0.133	3.24
Error	20.523	16	1.282	-	-
Av. length of midrib					
Dose	60.92	8	7.61	2.530	2.09
Days	0.129	2	0.064	0.021	3.24
Error	48.157	16	3.009	-	

(b) Analysis of variance for different parameters (ANOVA). Young and middle age cuttings.

Source of variation	SS	DF	MSS	Calculated VR	Tabulated VR
Av. length of lateral branch					
Dose	285.151	8	35.643	3.118	2.44
Days	11.842	1	11.842	1.036	5.32
Error	91.427	8	11.428	-	-
Av. no. of leaves formed					
Dose	219	8	27.375	0.718	2.44
Days	74.013	1	74.013	1.943	5.32
Error	304.612	8	38.076	-	-
Av. length of petiole					
Dose	7.824	8	0.978	1.670	2.44
Days	2.960	1	2.960	5.055	5.32
Error	4.684	8	0.585	-	-
Av. length of midrib					
Dose	49.46	8	6.182	1.639	2.44
Days	3.293	1	3.293	0.873	5.32
Error	30.171	8	3.771	-	-