http://www.pjbs.org

PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Pakistan Journal of Biological Sciences 4 (6): 670-671, 2001
© Asian Network for Scienfitic Information 2001

Effect of Gamma Irradiation on the Callus, Developed from Indica Rice

M.F. Hossain and M.S. Alam
Department of Genetics and Plant Breeding,
Bangladesh Agricultural University, Mymensingh, Bangladesh

Abstract: Induced mutation holds promise for effective improvement and have high potential for bringing about genetic variability. Regenerable calli of four rice varieties were exposed to 0 to 6 Gy of gamma rays to determine their effect on growth and plant regeneration capability. Both growth and regeneration capacity decreased with increasing levels of gamma rays ; however, plant regeneration capacity was more sensitive to gamma rays than growth. The 50% inhibition dose for callus growth and plant regeneration was approximately 5.0 Gy of gamma radiation in Binnatoa variety. But in remaining varieties 4.0 Gy was recorded for callus growth and 2.0 Gy for regeneration.

Key words: *Indica rice*, gamma radiation, callus growth, plant regeneration.

Introduction

Rice is a cereal crop that belongs to family Graminae. It is a primary food source for more than one third of world's population. Hence it is desirable to increase the yield and improve the quality of rice. Tissue culture techniques offer the great possibilities for selection of mutants through the use of new and expanded genetic variability. The variability in the population leads the chance of effective selection and increase the scope of improvement (Khanduja and Goel, 1986). Induced mutagenesis serves a source of variability for better selection. Many research workers have attempted to exploit somaclonal variation for crop improvement particularly treated with gamma radiation. Considerable work has been done on induced mutation in rice callus by Zhang *et al.* (1994), Sakila *et al.* (1999) and Pongtongkam *et al.* (1994). Min *et al.* (1991) applied low dosage treatment of gamma rays to callus in rice breeding. Gao *et al.* (1992) observed that 2.5-5.0 Kr is the optimal dose range for mature embryos of rice. A similar observation was noted in callus cultures of rice subjected to 20 Gy radiation (Lu *et al.*, 1997). In rice, Basmati–370 treated with 20 Kr triggered green plant production, lowered the callus production and albino plant production (Zapata *et al.*, 1986). The present study was undertaken to investigate the extent of variability on callus production and plant regeneration of four rice cultivars at different dosage of gamma radiation.

Materials and Methods

Rice (*Oryza sativa*) cv. Binnatoa, Pokkali, BR-16 and BR-26 (collected from Bangladesh Institute of Nuclear Agricultural, Mymensingh) were used. Mature and healthy seeds of these varieties were dehusked and surface sterilized in 0.1% $HgCl_2$ for 20 minutes followed by 4-5 rinses in autoclaved distilled water to remove traces of $HgCl_2$. Sterilized seeds were then cultured in MS (Murashige and Skoog, 1962) medium with 1.5 mg/l 2,4-D, 0.25 mg/l BAP, 30 g/l sucrose and 7% agar to initiate the callus. Calli were developed after 2 to 3 weeks of inoculation. 21 days old embryogenic calli were subjected to 7 different doses (0,1,2,3,4,5 and 6 Gy) of gamma rays (^{60}Co source used at BINA, Mymensingh). About 30-40 calli were exposed to each radiation level. The post irradiated material was transferred to MS medium supplemented with 0.5 mg/l NAA and 10 mg/l Kn. Repeated subculturing were done at an interval of 15 days for callus proliferation and organogenesis. Three weeks old irradiated calli were continued to proliferate and differentiate into shoots. When these shoots grew to about 3-4 cm in height , they were separated aseptically from each other and transferred to freshly prepared rooting medium (Half strength of MS + 0.5 mg/l IBA). The conical flasks containing plantlets were incubated in a controlled growth room at 25 ± 2 ^{O}C and data were recorded to note the response.

Results and Discussion

Twentyone days after irradiation it was clearly observed that an extent of survivability of calli varied on different levels of radiation (Table 1). In all cases, the growth and regeneration frequency of calli were found to be higher in control than in the mutated calli. The range of regeneration percentage was observed from 15 to 47 in Binnatoa, 0 to 40 in Pokkali, 0 to 34 in BR-16 and 0 to 23 in BR-26 (Table 2). The number of shoots per callus varied at the range of 1.8 ± 1.09 to 5.6 ± 0.54 in Binnatoa, 0 to 5.0 ± 1.0 in Pokkali, 0 to 5.8 ± 1.096 in BR-16 and 0 to 5.2 ± 1.48 in BR-26 (Table-3). Increasing level of radiation doses decreased the growth and regeneration capacity. The calli could be viable in Binnatoa by 50% up to 5.0 Gy of radiation, whereas in Pokkali, BR-16 and BR-26, growth was reduced to 50% at 4.0 Gy (Table 1).

Table 1: Survival rate of irradiated calli developed from *Indica rice*.

Varieties	Survival (%)						
	0 Gy	1 Gy	2 Gy	3 Gy	4 Gy	5 Gy	6 Gy
Binnatoa	82	70	72	65	54	40	39
Pokkali	77	71	60	46	37	26	18
BR-16	68	52	45	33	30	19	8
BR-26	54	40	36	30	27	12	6

Table 2: Plant regeneration from irradiated calli of *Indica rice*.

Varieties	Regeneration (%)						
	0 Gy	1 Gy	2 Gy	3 Gy	4 Gy	5 Gy	6 Gy
Binnatoa	47	36	30	25	20	23	15
Pokkali	40	34	20	20	6	-	-
BR-16	34	23	18	18	-	-	-
BR-26	23	18	12	12	-	-	-

The 50% inhibition dose (I_{50}) for plant regeneration, 5 Gy of gamma radiation would be upper limit in the variety Binnatoa, whereas 2.0 Gy would be for remaining varieties under study (Table 2). The reason may be due to high sensitivity of Calli

Table 3: Regenerated shoots developed from irradiated calli of *Indica* rice.

Varieties	Number of shoots /callus (Mean ± SD)						
	0 Gy	1 Gy	2 Gy	3 Gy	4 Gy	5 Gy	6 Gy
Binnatoa	5.6±0.54	3.8±0.83	4.0±0.70	5.4±0.65	2.8±0.83	2.4±1.14	1.8±1.09
Pokkali	5.0±1.00	3.8±0.83	2.5±1.30	1.9±0.49	1.0±0.47	-	-
BR-16	5.8±1.09	4.0±1.22	2.8±0.83	1.8±1.09	-	-	-
BR-26	5.2±1.48	3.4±0.84	2.6±0.54	1.0±0.70	-	-	-

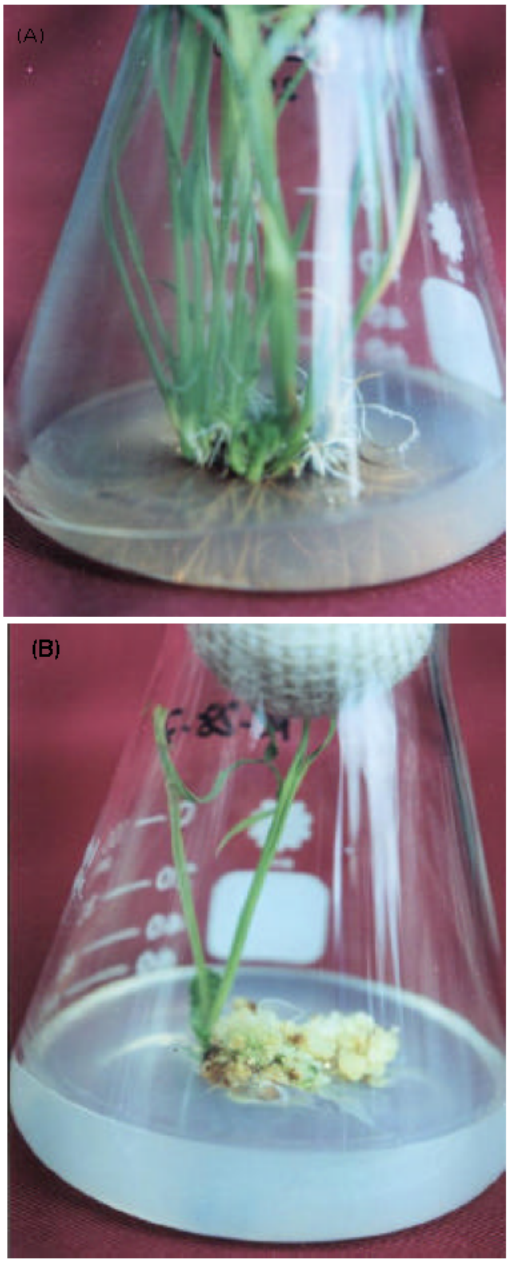

Fig. (A) Multiple shoot regeneration from embryo derived calli of Binnatoa without radiation. (B) Multiple shoot regeneration from embryo derived calli of Binnatoa treated with 5.0 Gy of gamma radiation.

to gamma rays of these varieties. A few number of shoots were produced from the calli of Binnatoa irradiated with 5 Gy as compared to control (Figs. A &B). The 50% inhibition dose for the number of shoots per callus, 4.0 Gy would be the upper limit for Binnatoa while 2.0 Gy would be for the remaining varieties (Table-3).

It was noted that an increasing level of gamma rays negatively affected the callus growth and plant regeneration. This may be attributed to the fact that the extreme doses of ionized radiation induced drastic structural damage in chromosomes which could be harmful to the genotype for its desired combination of genes.

References

Gao, M.W., Q.H. Cai and Z.Q. Liang, 1992. *In vitro* culture of hybrid *indica* rice combined with mutagenesis. Plant Breed, 108: 104-110.

Khanduja, S.D. and V.L. Goel, 1986. Pattern of variability in some fuel wood trees grown on sodic soils. The Indian Forester, pp: 118-123.

Lu, M., C.L. Wang and M. Shen, 1997. Effect of gamma irradiation on the formation of calli and regeneration of green plants in rice anther culture. Acta Agriculturae Zhejiangenesis, 9: 123-126.

Min, S., Z. Xiang, X. Qi and C. Zhao, 1991. Effects of gamma-ray radiation treatment on somatic cell cultura in rice (*Oryza sativa* L.). Cer. Res. Comm., 19: 201-208

Murashige, T. and F. Skoog, 1962. A revised medium for rapid growth and bioassays with tobacco tissue cultures. Physiol. Plant. 15: 473-497.

Pongtongkam, P., P. Ratisoontorn, S. Suputitada, S. Peyachoknagul, L. Ngernsiri, B. Silayoi, P. Rodrangboon, K. Klakhaeng and R. Senghaphan, 1994.Induced mutation in rice by tissue culture. Kasetsat J. Nat. sci., 28 : 381-389.

Sakila, M., S.M. Ibrahim and A. Kalamani, 1999. *In vitro* mutagenesis in scented rice cultivars. Rice Biotecnology Quarterly, 38 : 30-34.

Zapata F.J., R.R. Aldemita, L.B. Torrizo, A.U. Novero, S.K. Rina and R.R. Rola, 1986. Anther culture of Basmati 370 at IRRI. A Gamma-ray induced green plant regeneration. IRRN., 2: 22-23.

Zhang, J.J., K.H. Fan and Z.H. Zhang, 1994. Study on variation in rice (*Oryza sativa* L.). Somaclones from in vitro irradiated somatocytes. Acta Agric. Shanghai,10 : 6-11.