### STRAIN IMPROVEMENT OF PLEUROTUS SPP.

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**Abstract:** The effect of gamma irradiation, ultraviolet irradiation and nitroso-guanidine treatment of spore suspensions of three mushrooms viz., *Pleurotus sajor-caju*, *P. citrinopileatus*, *P. djamor* on the mycelial growth and yield performance was studied. Ultraviolet irradiation had no effect either on the mycelial growth or yield. Gamma irradiation at dose rate of 2000, 2500 and 3000 rad showed significantly increased mycelial growth and yield of *Pleurotus* spp. Treatment with nitroso-guanidine also resulted in increased mycelial growth, significantly higher yield and earliness in the appearance of sporophores.

Key words: Gamma irradiation, nitroso-guanidine, P. citrinopileatus, P. djamor, P. sajor-caju.

#### INTRODUCTION

The improvement of strains is essential to increase the productivity of mushrooms. The low yield of mushrooms is due to a number of factors among which genetic potential of the strain is the most important one (Dhar and Kaul, 1987). The genetic make up of mushrooms can he manipulated by mutation. Strain improvement by induction was reported by various workers (Dhaliwal *et al.*, 1992).

## **MATERIALS AND METHODS**

In the present study, both physical agents viz., ultraviolet (UV) and gamma irradiations and chemical viz., N-methyl-N-nitro-N-nitro-guanidine (NTG) were used for inducing mutation. The mushrooms viz., *P. sajor-caju*, *P. citrino-pileatus* and *P. djamor* were used for the study. The spore suspension (10<sup>4</sup> ml<sup>-4</sup>) of mushrooms was exposed to gamma irradiations at 1500, 2000, 2500 and 3000 rad. The irradiation was done by using Gamma Shine 1000 at dose rate of 64.3 kg h<sup>-1</sup>. For UV irradiation, the spore suspension was exposed to UV light at a distance of 10 cm at definite time intervals, viz., 10, 15, 20 and 25 min.

Mutation by using NTG was done following the standard procedure (Miyamoto *et al.*, 1983). The spores of mushrooms were collected and finely dispensed in 0.1 *M* phosphate buffer containing the mutagen (1000 ppm). After 1h of agitation, the activity of mutagen was

arrested by adding sterile phosphate buffer. From this, 1 ml of the solution was mixed with 15 ml of potato dextrose agar medium and incubated for the development of colonies. Each fungal propagule obtained was separately transferred and observed for growth. These colonies were transferred and maintained on potato dextrose agar slants. Sorghum grain spawn was prepared using these cultures following the standard procedure (Sinden, 1934); beds were laid out following the 'polybag' method of cultivation (Bhaskaran et al., 1978) and the common yield parameters were recorded.

### RESULTS AND DISCUSSION

The cultures (mycelia) of *Pleurotus* spp. developed from the spores exposed to UV irradiation for 20 and 25 mm and incubated for 5 days had less growth as compared to the untreated cultures (Table 1). Though the radial growth of cultures irradiated at 10 and 15 min was higher than the control, the growth was very thin and feeble compared to the thick growth in the untreated control plates.

Gamma irradiation resulted in the faster growth of *Pleurotus* spp. (Table 2). **In** the case of *P. sajor-caju*, irradiation at 2000 rad resulted in significantly increased mycelial growth (7.80 cm) followed by irradiation at **2500** and 1500 rad (7.63 and 7.43 cm), whereas in the case of *P. citrinopileatus* and *P.* 

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Table 1. Effect of UV irradiation on the mycelial growth of *Pleurotus* spp.

1 2		Colony o	Colony cliameter after five days*, cm							
	UV irradiation in min r	P. sajor-caju P. citrinopileatus P. djamor		7.44 7.41 6.71 6.54 7.16						
1	10	7.13	7.47	8.07	7.44					
2	15	7.00	7.57	7.73	7.41					
3	20	6.38	6.87	6.97	6.71					
4	25	6.13	6.47	7.03	6.54					
5	Control	6.33	7.07	7.67	7.16					
	Mean	6.58	7.09	7.49	2#2					

CD (0.051 for treatment, fungi and treatment x fungi are 0.14, 0.11 and 0.24 respectively; \* Mean of three replications

Table 2. Effect of gamma irradiation on the mycelial growth of *Pleurotus* spp.

SI. No.	_	Colony	*, cm		
	Treatment	P. sajor-caju	P. citrinopileatus	P. djamor	7.56 7.92 7.89
1	Gamma 1500 r	7.43	7.57	8.00	7.56
2	Gamma 2000 r	7.80	7.60	8.37	7.92
3	Gamma 2500 r	7.63	7.67	8.37	7.89
4	Gamma 3000 r	7.33	7.83	8.40	7.86
5	Control	6.38	7.17	7.67	7.16
	Mean	7.30	7.57	8.16	-

CD (0.05) for treatment, fungi and treatment x fungi are 0.21, 0.16 and 0.36 respectively; \*Mean of three replications

Table 3. Effect of mutation by NTG on the mycelial growth of Pleurotus spp.

SI. No.	_	Colony	*, cm		
	Treatment	P. sajor-caju	P. citrinopileatus	P. djamor	Mean
1	NTG-1	7.63	8.00	8.30	7.98
2	NTG-2	7.73	8.07	8.50	8.10
3	NTG-3	7.87	7.90	8.47	8.08
4	NTG-4	7.43	8.10	8.33	7.95
5	Control	6.30	7.13	7.80	7.08
	Mean	7.39	7.84	8.28	(4)

CD (0.05) for treatments, fungi and treatment x fungi are 0.14, 0.11 and 0.24 respectively; \*Mean of three replications

djamor the maximum growth (7.83 and 8.40 cm) was recorded by the cultures irradiated at

a dose rate of 3000 rad, followed by those irradiated with 2500 and 2000 rad (Table 2).

Table 4	Effect of gamm	a irradiation on	the enorophore	production	(no.) of Pleurotus s	nn
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SI.		P. sajor-caju					P. citrin	opileatu	S	P. djamor			
No.	Treatment	DFC	No. of crops	Sporo- phores		DFC		Sporo- phores			No. of crops	Sporo- phores	Fresh wt., g
1	Gamma rays 1500 r	21.7	4.00	: 52.7	343	18.0	3.67	56.00	380	15.7	3.67	49.1	390
2	Gamma rays 2000 r	• 21.0	3.67	50.7	353	17.3	3.67	61.33	381	15.3	3.67	51.0	397
3	Gamma rays 2500 r	21.0	4.33	49.3	375	17.6	4.33	59.00	384	15.3	4.00	53.0	411
4	Gamma rays 3000 r	20.3	4.33	52.0	373	17.3	4.00	69.00	418	15.0	4.00	50.3	428
5	Control	21.3	4.00	52.3	360	18.0	4.00	68.33	402	15.3	3.67	55.3	402

DFC = Days for cropping; CD (0.05) for days for cropping, no. of crops, no. of sporophores, fresh weight of mushrooms are 0.61, 0.43, 2.01 and 5.63 respectively; \* Mean of three replications

Table 5. Effect of mutation by NTG on the sporophore production (no.) of *Pleurotus* spp.

C1		P. sajor-caju				P. citrine	opileatus	3	P. djamor				
Sl. No.	Treatment	DFC	No. of crops	Sporo- phores	Fresh wt., g i	DFC	No. of crops	Sporo- phores	Fresh wt., g	DFC	No. of crops	Sporo- phores	Fresh wt., g
1	NTG-1	18.7	3.67	69.0	417	17.0	4.00	68.7	425	14.3	3.67	71.7	498
2	NTG-2	18.3	3.67	66.0	419	16.7	3.67	64.3	438	14.7	4.00	69.7	503
3	NTG-3	18.3	4.00	68.7	413	17.0	3.33	59.3	435	14.0	3.33	70.7	515
4	NTG-4	18.3	3.67	71.0	428	16.3	3.33	64.3	457	14.3	3.67	73.0	: 512
5	Control ;	21.4	4.00	65.4	410	18.7	3.33	65.0	418	15.0	3.67	79.7	; 488

CD (0.05) for days for cropping, no. of crops, no. of sporophores, fresh weight of mushrooms are 0.55, 0.37, 1.89 and 5.89 respectively; \* Mean of three replications

Among the five isolates of P. sajor-caju obtained by NTG treatment, NTG-3 and NTG-2 showed maximum mycelial growth followed by NTG-1.All the mutants recorded significantly higher mycelial growth as compared to control. In the case of P. citrinopileatus the mutants NTG-4 and NTG-2 recorded the maximum mycelial growth followed by NTG-1 (Table 3). The yield performance of the irradiated cultures was studied. The results indicated that the total yield of P. sajor-caju and P. djamor irradiated at 2500 and 3000 rad was significantly more than that of the control. La the case of *P. citrinopileatus*, the cultures irradiated at 3000 rad recorded maximum yield. The average weight of the mushroom was more in the case of cultures irradiated at higher doses (Table 4).

The cultures of *Pleurotus* spp. treated with NTG showed significantly higher yield and **earliness** in the appearance of first crop by two days (Table 5). The isolate NTG-4 of *P. sajor-caju* recorded higher yield (428.33 g) followed by NTG-2 (418.33 g). In the case of *P. citrinopileatus*, NTG-4 recorded the maximum yield (456.67 g), compared to the control which produced an average yield of 418.33 g. The isolates NTG-3 of *P. djamor* gave significantly higher yield (515.00 g), followed by NTG-4 (511.67 g) compared to the control which gave 488.33 g. The average weight of the sporophores was found to be higher in the case of mutants as compared to control.

The physical as well as chemical mutagens have been frequently used in mushroom to

obtain desirable traits. The NTG mutants recorded higher yield besides earliness in the appearance of sporophore, in the present study. Early fruiting and improved yields by laccase mutants of P. florida was obtained by exposing the germinating basidiospore suspension (1 x 10<sup>4</sup> spores / ml) to NTG (Dhaliwal et al., 1992). According to Bahukhandi and Munjal (1988) an exposure of 30 s to gamma irradiation of spores had practically no effect on the number of colonies and a progressive increase in exposure (15 min) resulted in a decrease in the number of colonies and two isolates developed in their study recorded higher yield. But according to the present study gamma irradiation at higher doses showed significantly higher mycelial growth and yield.

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#### REFERENCES

- Bahukhandi, D. K. and Munjal, R. L. 1988. Effect of gamma irradiation on colony appearance and crop yield of *Pleurotus sajor-caju* (Fr). Singer. *Indian J. Mush.* 14: 41-43
- Bhaskaran, T. L., Sivaprakasam, K. and Kandaswamy, T. K. 1978. Compact hag method a new method of increasing the yield of *Pleurotus sajor-caju*. *Indian J. Mush* 4(2): 10-12
- Dhaliwal, R. P. S., Garcha, H. S. and Phutela R. P. 1992.

  Early fruiting and improved yields by laccase mutants of *Pleurotusflorida*. Mush Sci. 1:73-78
- Dhar, A. K. and Kaul, T. N. 1987. Genetics and improvement of mushroom crops. *Indian Mush. Sci.* 2: 339-347
- Miyamoto, T., Reddy, N. S. and Nakae, T. 1983. Induction of mutation in *Lactobacillus easel* subsp. *alactosus* by nitroso-guanidine. *Agric. Biol. Chem.* 47: 2755-2759
- Sinden, J. W. 1934. Mushroom spawn and methods of making the same. U. S. Patent 2: 844-861