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Effect of Gamma Irradiation on Growth, Yield and Composition of Oil in Patchouli (*Pogostemon patchouli* Pellet.)

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Abstract

Effect of different doses of gamma rays was investigated to develop high yielding mutant in patchouli. The *in vitro* developed callus were exposed with 3, 5, 6, 8, 9 and 10 kR dose of gamma rays. The irradiated callus were cultured on regeneration media. The regenerated plants serve screened in the field for quantitative and qualitative traits. The study resulted in isolation of several economically useful mutant showing variation in agronomic as well as essential oil characteristic. The vegetative growth shows inverse relationship with dose state, as dose increases, herb yield decreases. However, high frequency of alteration in oil content and oil yield was noticed in several treatments, especially in callus exposed with 6 kR which exhibited pronounced effect in enhancing oil content (2.4 to 3.42%) and oil yield (68.34 to 76.26 t/ha per year).

Key words : Gamma irradiation, Patchouli, Growth, Yield, Oil composition.

Pogostemon patchouli Hook. f., commonly known as patchouli is believed to be a native of Philippines. *Pogostemon* is also reported to occur in wild form in the humid tropical parts of eastern, western and southern India. It is highly valued for its essential oil "patchouli oil" which is a perfume by itself. Due to lack of any synthetic substitute for patchouli oil, it is currently indispensable in cosmetics, soaps, incense and flavor. In addition the oil also possesses bacteriostatic and antifungal properties. India spends a huge sum of money (about Rupees 25 million per annum) for importing patchouli oil mainly from Indonesia to meet the growing needs of perfumery/flavor industry (1). Patchouli being a vegetatively propagated crop, virtually there is no scope for generating the variability as the available genetic variation in this crop is narrow. Traditional breeding methods tried for breaking yield barriers were not successful as the plant rarely flowers and does not set seed. Possibility of obtaining high yielding clone/genotype through mutation breeding in patchouli was therefore considered on alternative to conventional breeding technique. Therefore, present study was designed for induction of mutation using gamma irradiation for genetic variability so as to improve the quantitative and qualitative traits.

Methods

In vitro developed callus were irradiated with different doses of gamma rays (3, 5, 6, 8, 9 and 10 kR) using 60_{CO} source, at Rashmi irradiation center, Kidwai, Bangalore. immediately after irradiation, the callus were transferred to the culture room. Dose response curve was first established and LD_{50} was determined. The irradiated callus were subcultured for 5 cycles at 12 days interval and cultivated in regeneration media. Healthy plantlets were hardened and later, the irradiated plants along with control plants were transplanted in the field at 60 × 60 cm spacing under uniform field condition. Cultural operations are carried out at timely interval and after five months of planting, plant were ready for harvesting. Observation on five agronomic traits viz., plant height, number of branches, leaf area, herb yield, oil content and yield were recorded at the time of harvesting.

Oil estimation of dry herb was done by hydro distillation using clevanger's apparatus. The percentage of oil was calculated on moisture free basis, quality was assessed with the help of gas chromatographs. The mean value for all the economic traits were subjected to statistical analysis (randomized complete block design) for different characters pairs at differ-

Table 1. Plant height, number of branches and leaf area as influenced by gamma rays in the plants obtained from the callus of patchouli (*Pogostemon patchouli* Pellet.).

Treatments	Plant height (cm)	Number of branches/ plant	Leaf area/plant (cm)
T_1 Control	82.50	35.75	4555.75
T_2 3 kR	81.50	31.25	4243.50
T_3 5 kR	75.00	26.75	3929.50
T_4 6 kR	67.00	23.25	3365.00
T_5 8 kR	60.75	19.75	2792.50
Mean	73.35	27.35	3777.25
F-test	*	*	*
SE ±	1.25	1.00	18.49
CD 5%	3.86	3.09	57.00

Table 2. Herb yield, oil content and oil yield as influenced by gamma rays in the plants obtained from the callus of patchouli (*Pogostemon patchouli* Pellet.).

Treatments	Herb yield/ harvest/ ha (t)	Herb yield/ ha/yr (t)	Oil content (%)	Oil yield/ harvest/ ha (1)	Oil yield/ ha/yr (1)
T_1 Control	0.90	2.70	2.40	22.75	68.34
T_2 3 kR	0.88	2.64	2.37	21.12	63.36
T_3 5 kR	0.86	2.58	2.62	22.87	68.61
T_4 6 kR	0.73	2.19	3.42	25.42	76.26
T_5 8 kR	0.74	2.22	2.77	20.68	62.04
Mean	0.82	–	2.72	22.57	–
F-test	*	–	*	*	–
SE ±	0.01	–	0.06	0.66	–
CD 5%	0.05	–	0.19	2.05	–

ent doses.

Results and Discussion

The treatments of plant cells and tissue *in vitro* with physical mutagens enhance the spectrum and frequency of variations. In the present investigation, the callus exposed with different doses of gamma-rays showed wide variability in both quantitative and qualitative traits.

Growth Parameters

The vagetative growth showed signficant differences along with different dose of gamma rays. The callus exposed with 8 kR dose of gamma rays recorded minimum plant height (60.75 cm), number of branches (19.75/plant) and leaf area (2792.50 cm^2), it is the maximum dose rate where we observed 50% of survivability (LD_{50}), whereas the control treatments i.e., unexposed callus recorded maximum plant height (82.50 cm), number of branches (35.75/plant and leaf area (4,555.75 cm^2) (Table 1). By the results we observed that the growth traits showed inverse relationship with dose rate i.e., as the dose rate increases, the plant height, number of branches and leaf area get decreases. This might be due to the fact that, the increase dose of gamma rays may effect the vital mutation in the genotype of patchouli (2). Similar results were observed by Malpathak and David (3) in garlic and Nayak et al. (4) in lemongrass, where they reported that, enzyme activity associated with biosynthesis of primary metabolic traits like plant height, number of branches, leaf area was recorded with increased dose rate of gamma rays.

Yield Parameters

Gamma irradiation was found to be influence on herbage yield (Table 1). The maximum dry herb yield (2.7 t/ha per year) was recorded in the unexposed callus and minimum (2.19 t/ha per year) was recorded in callus exposed to 6 kR dose of gamma rays. From the result it was observed that as the dose rate increase, the dry herb yield decreases. This is because in increased dose rate, the vegetative growth decrease so as the herbage yield. Regarding oil content, the callus exposed with 6 kR dose of gamma rays recorded maximum oil content (3.42%) whereas, minimum oil content (2.37%) was recorded in plant treated with 3 kR dose of gamma rays which was on par with unexposed callus (2.4%). However, any increase in the dose rate behind the 6 kR, the oil content get decreases. This is because, the synthesis of secondary metabolite like, essential oil, alkaloids, fallows a definite biosynthetic pathway catalyzed by specific enzyme involved in each step. In this sequential enzymatic activation, it is possible to hit the specific enzyme at specific step or activate them all by gamma irradiation by changing or enhancing the rate of biosynthesis. The 6 Kr dose of gamma rays might favorably activate the enzyme which associated with biosynthesis of essential oil in callus of patchouli could cause the enhancing the rate of biosynthesis leading to higher oil content. This study is in agreement with

earlier observation of Nayak et al. (4) in lemongrass, Shylaraj and Thomas (5) in palmarosa, Sharma et al. (6) in Henbane. In oil yield the callus exposed with 6 kR dose of gamma rays recorded maximum oil yield (76.26 l/ha per year) whereas minimum oil yield (62/04 l/ha per year) was recorded in the callus exposed was 8 kR dose of gamma rays. Here also as that of oil content, the dose rate increases the oil yield increase up to 6 kR dose behind, this oil yield get decreases. This is because, oil yield mainly depend upon herb yield and oil content though 6 kR dose recorded maximum oil content so as more oil yield.

Regarding composition / quality of oil, gamma irradiation had greater effect in callus of patchouli. The callus exposed with 6 kR dose of gamma rays, recorded the maximum patchouli alcohol (52.90) which is major economic significant in the patchouli oil, as against the control/unexposed callus which recorded 32.02% of patchouli alcohol.

In conclusion considerable progress has been made in the application of induced mutation in vegetative propagated plants. In seeds or vegetative propagated aromatic plants. Ioining radiations have been successfully used to enhance their industrial utility. In present study, plant exposed with 6 kR dose of gamma rays may be selected as mutant clone which has higher oil content and oil yield. Further evaluation of these clones are needed to known their consistency. Thus mutation breeding offers considerable scope for altering both quantitative and qualitative characters in patchouli in a desirable direction. Only through careful screening and selection, the range of variability induced by gamma rays can be widened and which can further exploited by recombinant breeding.

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