

HIGH ALKALOID YIELDING MUTANT IN *RAUVOLFIA SERPENTINA* BENTH

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ABSTRACT

X-rays apart from inducing changes in morphology, have brought significant changes in the alkaloid content of *Rauvolfia serpentina*. Treatment with 51 kR proved to be the most suitable dose for the same.

INTRODUCTION

MUTAGENIC effects of X-rays by Muller (1928) in *Drosophilla*, Goodspeed and Stadler (1929) in plants opened a new field in the experimental modification of genotype and phenotype in the biological world. Subsequently, several economic plants were exposed to X-rays by Gustafsson (1947), Kumar and Joshi (1949), Jacob (1945), Rai (1958 *a, b*), Ammal Kumar (1961), Swaminathan and Vaurghese (1966), Banerjee and Swaminathan (1966). Furthermore, with a view to raise newer types having higher yield and grain weight, early maturity, superior strength of straw and other biochemical characteristics like better grain quality both in protein and oil by Rai *et al.* (1957), Nayar (1961), Banerjee and Swaminathan (1966) and Sundahl (1961).

Although considerable variation in alkaloid content has been achieved through selection and colchipoity in *R. Serpentina* (Parimoo, 1968), by and large mutagenic agents have not been used for this purpose so far. Induction of variation in the alkaloid content by X-rays was taken up as a next tool in the improvement of *R. serpentina*.

MATERIAL AND METHODS

Because plenty of material was available of the Dehra Dun race, 50 seed lots were taken for each treatment. As the roots are the source of alkaloid

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content in *R. serpentina* fresh seeds (50 each treatment) were irradiated with various doses of X-rays ranging from 50,000 R to 55,000 R at Bhabha Atomic Research Centre, Trombay (Bombay). This included six treatments in all, (50,000 R, 51,000 R, 52,000 R, 53,000 R, 54,000 R and 55,000 R). Philips X-ray machine was used. Estimation of alkaloid content was done by Horammer and Rao's method (1954). The roots were washed thoroughly dried at 60° C for 24 hours, powdered and passed through 60 mesh. The powder was extracted with absolute ethanol by the above method.

RESULTS AND DISCUSSION

Seeds treated with dosages more than 51,000 R either failed to germinate or germinated but did not survive. Maximum germination percentage and survival was seen in 51,000 R treated plants. Morphological changes in the M_1 and M_2 are summarised in Table I. Two phenotypes are distinguished. One of these (A) had an overall resemblance to the control plants but was shorter and at some nodes there were 5 leaves instead of the usual 3 or 4 (Fig. 1). The other type (B) was much different from the control in being

TABLE I
Summary of morphological characters
Dosage 51,000 R

Characters	Control	A Type	B Type
Plant height (cm)	.. 75±8	51.4±2.41	32.5±6.17
Leaf size :			
Length (cm)	.. 11.9±1.08	11.5±0.05	10.5±0.60
Breadth (cm)	.. 4.5±0.8	4.3±0.20	5.0±0.35
Petiole length (cm)	.. 2.1±0.622	2.4±0.966	1.8±0.763
Peduncle length (cm)	.. 8.2±2.4	7.1±0.466	6.1±0.793
Flowers peduncle	.. 195—320	202—303	56—120
Flower colour	.. Whitish pink	Whitish pink	Dark pink
Corolla tube length (cm)	.. 2.1±0.14	1.8±0.17	2.5±0.5
Pollen fertility(%)	.. 93	53.4	38.3

dwarf and had dark pink flowers with shorter corolla tube and thicker dark green leaves with shapes varying from lanceolate to ovate (Fig. 2). While pollen morphology did not change, the pollen fertility was lower in treated types (being 38% in B type) in comparison to the control plants.

M₂ generation was raised from morphologically aberrant M₁ plants through seeds. In addition to the two types of plants observed in M₁ generation there were chimeral plants in which some branches possessed characteristics of A while others had those of B type (Fig. 3).

Variation in the alkaloid content was ascertained from three year old roots of treated as well as control plants. The data is summarised in Table II.

TABLE II
Variation in the total alkaloid content

Material	M ₁ generation		M ₂ generation	
	Mean alkaloid percentage	Percentage control	Mean alkaloid percentage	Percentage control
Control	1.80	100.00	1.64	100.00
Treated (50,000 R)	1.60	88.89	1.20	73.18
	1.70	97.23	1.80	109.10
	2.40	133.00	2.23	137.20
	1.20	66.67	1.40	85.40
	1.30	72.23	1.65	100.80
	2.00	110.00	2.00	121.95
	2.95	164.80	2.85	171.34
	3.50	173.30	3.10	189.02

It can be inferred from Table II that the mean alkaloid percentage in the control was 1.80, whereas in the treated plants the range was from .20-3.10 in M₂. Two plants possessed significantly higher alkaloid content., Variance in the alkaloid content in the control population was 0.05 to 0.07%

whereas in the treated plants it was 0.06 to 1.50% in M_1 . In the M_2 variance in the control population was 0.03 to 0.04%; but 0.061 to 1.40% in the progeny raised from M_1 . The percentage of alkaloid content expressed as the percentage of control, varied from 88.89 to 173.30 in M_1 , while 73.18 to 189.02 in M_2 generation.

Morphological changes are common following exposure to ionizing radiations. Normal expression of leaves, roots, stems and flowers are frequently altered by radiations. Such changes have been observed in wheat (Vaurghese and Swaminathan, 1966; Banerjee and Swaminathan, 1966), rice (Basu, 1961; Shama Rao, 1970; Navahari, 1970), *Cajanus* (Chopde, 1970), *Brassica juncea* (Narain *et al.*, 1970) and *Sesamum orientale* (Nayar, 1961). The changes in the flower colour and thickness seen in the treated plants of *R. serpentina* have also been observed in *Antirrhinum* (Stubbe, 1960), Carnation (Sagwa and Mehlquist, 1954) and *Chrysanthemum* (Dowrick, 1960).

The chimeral plants found in M_2 progeny in *R. serpentina* have also been reported in some cereals (Mackey, 1954) and dicots (Blixt *et al.*, 1958). Such changes can be attributed to gene mutations rather than any physiological changes brought about in the seeds by irradiation.

As already mentioned significant variation in the alkaloid content has been obtained in *R. serpentina* by X-rays. Similar variation was obtained in the oil and protein content of *Sesamum orientale*, mustard, wheat and barley (Nayar, 1961; Rai and Jacob, 1958; Despande and Jeswani, 1966; Banerjee and Swaminathan, 1966; Sundahl, 1961). The increase in alkaloid content in X-rayed *R. serpentina* can be explained by mutations of major genes or critical gene in the series of steps responsible for ultimate phenotype, in this case the alkaloid content. Thus, the scope of improving the alkaloid content by mutations does exist.

Earlier, similar increase in the active principle has been reported in the autopolyploids of *Teraxecum koksaghyz* (Warmke, 1939), *Labelia inflata*, *Datura stramonium*, *Datura tatula*, *Atropa*, *Hyocymus* (Rowson, 1945) and *Nicotiana* (Eigsti, 1957).

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FIGS. 1-3. Figs. 1-2. Type A, B among X-rays treated plants. Fig. 3. Chimeral type of plants among X-rays treated plants.