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Effect of Chemical and Physical Mutagens on Seed Germination of Green gram (*Vigna radiata)* L. Wilczek

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ABSTRACT
Mutation breeding stands out as the most crucial method in crop improvement strategies for the desirable Qualitative and Quantitative characters. Submission of the different concentrations and doses of the chemical and physical mutagens by knowing the LD50 value has immense importance in the mutation breeding experiment. During the kharif season, mungbean holds significance as a crucial pulse crop cultivated in India due to its short life cycle, symbiotic nitrogen-fixing potentiality, and protein and mineral content. In the ongoing trial, green gram (Vigna radiata) L. Wilczek seeds are being used variety Phule Chetak (PM-707-5) were procured from Oilseed Research Station, Jalgaon. The seeds were subjected to both chemical mutagenesis using Ethyl Methane Sulphonate (EMS) at varying concentrations (10mM, 20mM, 30mM, and 40mM) and physical mutagenesis through exposure to Gamma rays emitted by Cobalt-60 at different doses (100Gy, 150Gy, 200Gy, 250Gy, 300Gy, and 350Gy). The M1 Generation was observed for variations in germination percentage in response to various concentrations and doses of the mutagens. It is observed that increased concentration and doses of the mutagen decreased the germination percentage of the treated seeds compared to the control. Both the mutagen causes biological damage suggesting the potentiality for induction of the mutation in the Green gram.
Keywords: *Vigna radiata, EMS, Gamma rays, Mutagen, Seed germination, biological damage*

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INTRODUCTION
Mutation breeding is a traditional technique within plant breeding, demonstrating successful application in enhancing various food crops, ornamentals, and export crops through induced genetic variation [6]. Utilizing physical and chemical mutagens stands as an essential and intricate technique in creating mutations, pivotal for fostering new varieties with superior traits. Geneticists employ mutations as instruments to explore the characteristics and roles of genes, fundamental components crucial for the growth and development of plants, thus furnishing essential resources for enhancing the hereditary traits of valuable crops [1]. Critical factors for bringing change with EMS include awareness, treatment duration, and solution temperature [7]. Mutational breeding offers the advantage of enhancing specific traits without altering the overall genetic makeup [2]. Mutagens, both physical and chemical, were extensively employed in generating mutations in crop plants [3,5]. The Mungbean belongs to the family Fabaceae. Mungbean botanically described as (*Vigna radiata* L. Wilczek)**.** It is referred to as green gram or Mungbean. Mungbean, valued for its high-quality protein and mineral content, serves as a vital grain legume for India's vegetarian population. Cross-breeding is restricted in its ability to boost mungbean production due to limited genetic diversity, despite its significance as a crucial crop in India, Pakistan, Thailand, Vietnam, Myanmar, and China, with a diploid chromosome count of 2n=22.

MATERIAL AND METHODS
Plant Material-
The seeds of mungbean (*Vigna radiata* L. Wilczek) variety Phule Chetak (PM-707-5) was purchased from Oilseeds Research Station Jalgaon and research was conducted in 2023–2024. In order to cause mutations, physical mutagens like gamma rays and chemical mutagens like EMS are used in modern treatments.

Mutagenic treatments

Gamma irradiation treatment

The seed was exposed to several gamma radiation doses at SPPU Pune Department of Chemistry, which used 60Co as the irradiation source. These dosages were given in increments of 50 Gy, ranging from 100 Gy to 350 Gy.

Ethyl Methane Sulphonate treatment

EMS ($CH_3SO_2OC_2H_5$), an alkylating agent was used in the current study. The seeds were pre-soaked in distilled water for six hours to increase their susceptibility to mutagenesis effects. They were then exposed to EMS at different concentrations (10 mM, 20 mM, 30 mM, and 40 mM) for four hours. To get rid of any remaining chemicals, the seeds were carefully rinsed with running tap water after the chemical operation. Healthy, mature seedlings that had not received any treatment made up the control samples.

Germination percentage (%)

In the laboratory, Seeds from every treatment and the control sample were placed on germination paper in the Petri dish, and the number of seeds that sprouted by the 7th day was checked to determine the growth percentage for every treatment.

The germination percentage serves as a measure of the seed population's viability, and it's determined by using a specific equation:

$$GP = (\text{seeds germinated} \,/\, \text{total seeds}) \times 100$$

RESULT AND DISCUSSION

The control group showed a 100% seed germination rate (Table 1). As the level concentrations of EMS and Gamma Rays increased, there remained a corresponding decline in the germination percentage over time. (Figure 1 and 2). The percentage of seed growth remained highest in lower doses of EMS (10mM-86%) than in Gamma rays (100Gy-82%). (Table 1). In the present study, the Mungbean shows maximum germination on the 7th day after sowing on blotting paper in Petri dishes. A decrease in seed germination might result from mutagenic influence on the meristematic cells of the bud [4]. Higher doses/concentrations of the mutagens may hinder seed germination due to disruptions occurring at the cellular scale (resulting also at the physiological level).

Table 1: Effect of mutagens on Seed Germination Percentage in M_1 Generation of *Vigna radiata* (L.) Wilczek

Treatment	Concentration/Dose	No.of Seeds Sowed	No.of Seeds Germinated On the 7th day	Germination (%)
Control	----	50	50	100
EMS	10mM	50	43	86
	20mM	50	41	82
	30mM	50	38	76
	40mM	50	34	68
Gamma Rays	100Gy	50	41	82
	150Gy	50	40	80
	200Gy	50	43	78
	250Gy	50	39	76
	300Gy	50	34	68
	350Gy	50	41	64

Fig 1: Seeds for counting germination percentage. A and B). EMS Treated seeds. C). Gamma rays treated seeds.

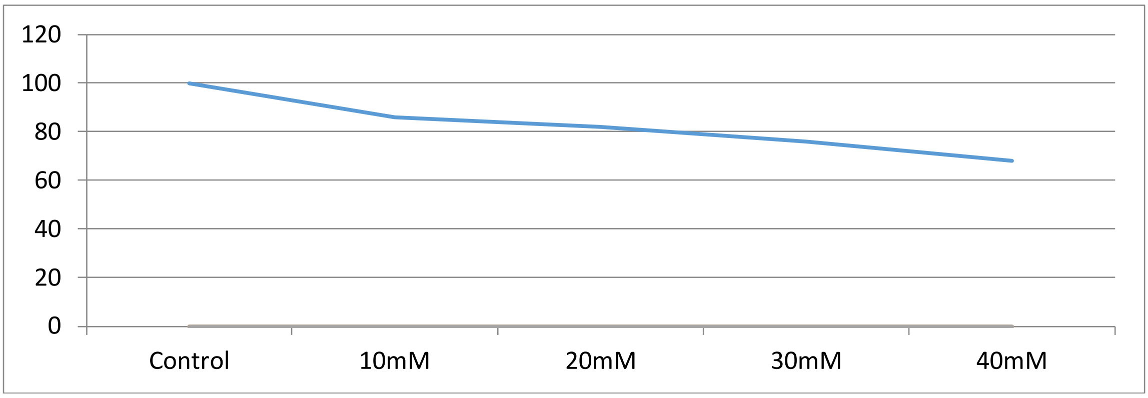

Fig. 2. The outcome of EMS on seed growth percentage (%) of green gram.

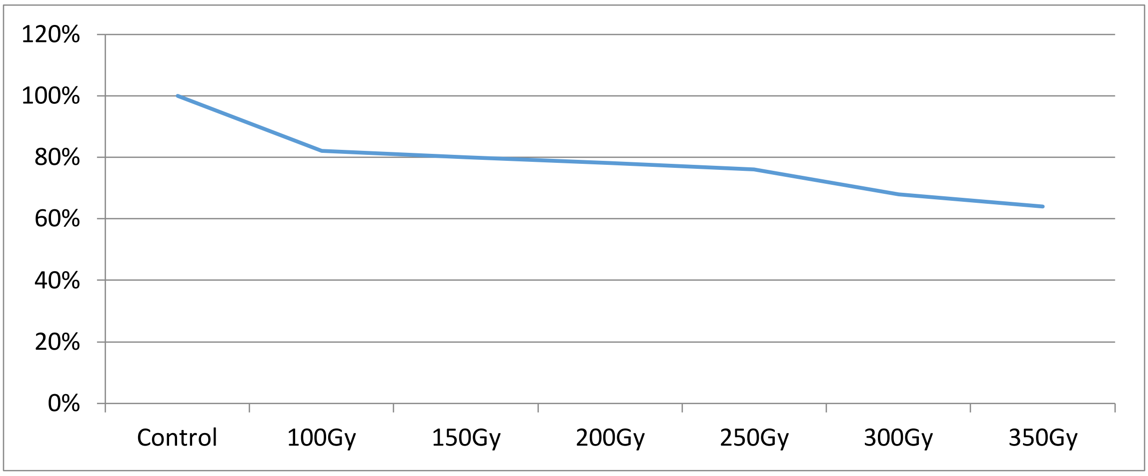

Fig. 3. The outcome of Gamma Rays on seed growth percentage (%) of Green gram.

CONCLUSION

It concluded that how gamma rays and ethylene methane sulphonate (EMS) affect the rates of seed germination. Physical therapies ranged from 100 Gy to 350 Gy, while chemical treatments used dosages between 10 mM and 40 mM. The results indicated that the highest germination percentage (86 %) was obtained with lower doses of EMS (10 mM), whereas the lowest germination percentage (68 %) was obtained with higher doses (40 mM). Likewise, gamma radiation showed the lowest germination at 350 Gy (64 %) and the highest at 100Gy (82 %). Overall, in lab settings, seed germination rates were inversely connected with rising EMS concentrations and gamma radiation dosages.

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