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Research Article

Morphological Changes Through Mutation of Local Chilli in North Sumatra, Indonesia

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Abstract

Background and Objective: The chilli production produced is still less than optimal due to obstacles from biotic and abiotic stress. Genetic improvement of superior chilli varieties in Indonesia are very important considering that problem. The aim of this study was to induce mutations in chilli plants to increase their genetic diversity through changes in plant morphology. **Materials and Methods:** This study method is a descriptive statistical method. Local chilli cultivar seeds was used and evaluated the response on the growth of the first generation (M1) through gamma ray irradiation treatment. This seeds irradiated with doses of 0, 100, 200 and 300 Gy. The study assessed variations in quantitative traits such as plant height, dichotomous height, leaf area and stomata density. Data analysis used the means, standard deviation and paired t-test to compare the means of the irradiation treatment with the means of the control treatment. **Results:** The variation derived from the plant height, dichotomous height, leaf area, stomata density on M1 generation affects plant growth at vegetative phase than the control plants. Higher levels of irradiation at 300 Gy decreased the growth and development of the chilli seedling. The low level of stomata density and supported by broad leaves really supports the better growth in a population of 100 Gy compared to the control population (0 Gy). **Conclusion:** Exposing chilli plants to doses ranging from 0 to 300 Gy through mutation induction alters their growth and morphology diversity in comparison to non-irradiated plants.

Key words: Diversity, gamma ray, *Capsicum annuum* L., irradiation, morphological changes

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Red chilli (*Capsicum annuum* L.), holds significant importance in horticulture, being commercially cultivated for its considerable economic worth. It finds extensive use both in households and the food industry. Red chillies not only add flavor and color to dishes, stimulating appetite, but they also boast a rich vitamin content. Furthermore, they possess medicinal properties and serve as essential ingredients in both medicinal and food ingredients^{1,2}.

The low productivity of red chilli in West Java, Indonesia, indicates that there is a serious problem in the cultivation aspect. In addition, extreme climate change also causes red chilli plants to be damaged. As a result, production decreases so that the price of production increases and farmers' income decreases. Climate changes causes the risk of a resistance and tolerance breakdown caused by biotic and abiotic stresses, affect the durability of pathogens resistance and environment tolerance. Therefore, there is an urgent need to develop new resistant and tolerant cultivars that can be adapted to varied pedoclimatic conditions^{3,4}.

In North Sumatra, especially Batu Bara Regency, many local red chillies are cultivated which have high productivity, good quality, long life and are liked by consumers. The chilli production produced is still less than optimal due to obstacles from plant pest organisms.

Genetically assembling stress-resistant chilli plants is a relatively better and environmentally friendly solution. This can be achieved by plant breeding techniques to produce new varieties that are resistant to stress, including by using mutation techniques.

Mutation breeding plays a crucial role in enhancing plant characteristics when the desired traits are absent in a species' gene pool. This is achieved through the application of mutagens, including the use of gamma ray exposure. The primary goal of inducing mutations in chilli plants is to broaden their genetic variation, as evidenced by morphological changes and environmental adaptability.

Several researchers⁵⁻⁸ have investigated the effects of gamma irradiation, finding it effective in increasing both the genetic diversity and quantitative attributes of chilli plants. The results of these studies were expected to offer valuable insights into enhancing the growth of chilli plants by utilizing gamma ray irradiation.

The objective of this study was to increase the genetic diversity of chilli plants to obtain superior plants, especially because chilli plants are self-pollinated plants with limited genetic variability. Therefore, it is necessary to increase genetic diversity, one of which is by inducing gamma ray mutations to obtain the appropriate irradiation dose.

MATERIALS AND METHODS

Study area: This study method is a descriptive statistical method. The study was carried out from October, 2023 to December, 2023. This study was conducted at the National Atomic Energy Agency (BATAN) for irradiation treatments, with the field study taking place at Universitas Sumatera Utara, Medan. One hundred white local chilli cultivar seeds were treated with gamma irradiation at doses of 0, 100, 200 and 300 Gy using a gamma irradiator chamber.

Study design: The first generation (M1) seeds that have been irradiated are planted in a seeding medium mixed with top soil, rice husks and compost in a ratio of 1:1:1. Chilli seeds according to the treatment dose, are sown in small polybags (8×9 cm in size). Transplanting is carried out when the chilli seedlings are 3 weeks old after sowing and have 3-4 leaves. The chilli seedlings are moved by cutting the bottom of the plastic, then the seeds are carefully removed and planted in a 20×20 cm polybag. The media used in polybags is the same as the media composition in nurseries. Pest and disease control is carried out according to conditions in the field. Maintenance is carried out in accordance with optimal cultivation.

The number of plant samples is 10% from each irradiated population. The study assessed variations in quantitative traits such as plant height, dichotomous height, leaf area and stomata density.

Statistical analysis: Data analysis used the means, standard deviation and paired t-test to compare the means of the irradiation treatment with the means of the control treatment.

RESULTS AND DISCUSSION

Increasing the gamma-ray dose from 100 to 300 Gy impacted the development of chilli seeds. Higher levels of irradiation affected the growth of the seedlings. The effect of the irradiation treatment on the plants could be visually assessed, including observing the plants' growth response.

The decrease in the percentage of plant growth with increasing doses of gamma ray irradiation is thought to be closely related to plant physiological disorders. The percentage of growth and the amount of seed damage depend on the viability of the seeds and the amount of gamma irradiation dose (Table 1).

Díaz-López *et al.*⁹ reported a negative correlation between germination percentage and radiation, indicating that as the radiation dose increases, germination tends to decline.

Gamma ray irradiation treatment causing some chilli seeds do not grow with increasing irradiation dose. The viability of the seeds could have been adversely affected by an imbalance caused by gamma radiation, leading to reduced seed germination and impaired development of the plants.

Moghaddam *et al.*¹⁰ reported that high doses of irradiation disrupt the hormonal balance and enzyme activities within plant cells. Similarly, Pavadai¹¹ attributed the reduction in plant survival rates to physiological disruptions or chromosomal damage inflicted on plant cells by mutagens.

The research showed that exposure to gamma irradiation ranging from 100 to 300 Gy affected the expression of morphological traits in each irradiated M1 generation population. Differences in the means values of these morphological traits are visible when compared with populations that were not exposed to irradiation, as shown in Table 2.

Pavadai¹¹ noted the challenges in determining the ideal mutagenic dose. This is because low and high doses of mutagens have their respective advantages and disadvantages. Typically, higher mutagenic doses result in a greater number of potential mutants.

van Harten¹² and Piri *et al.*¹³ explained that the M1 generation experienced physiological and biochemical changes, along with inhibition of growth and germination, due to irradiation. Gamma rays produce free electron radicals that damage cell structures. The dose of gamma ray irradiation required varies for each type of plant, causing changes in quantitative characteristics. Anticipated changes aim at beneficial developments and not abnormalities.

The young plants of chilli have leaves that vary with each irradiated population. Treatment of dose irradiation causes the appearance of leaf area and size different from control plant Fig. 1(a-d). The higher the irradiation dose, the lower the growth of plant height, dichotomous height and leaf area (decreased growth was found in the 200 and 300 Gy populations compared to the control population). The 100 Gy population tended to have better growth in the observed characters, especially in the leaf area character compared to the control population (0 Gy) (Table 2, Fig. 1).

Increased doses of gamma ray irradiation affects stomata density of chilli leaf after treated with gamma irradiation. Dose of irradiation affects less stomata density than the control plants Table 2, Fig. 2(a-d). The stomata density in the 100 and



Fig. 1(a-d). Plant phenotype and leaf area, (a) 0 Gy, (b) 100 Gy, (c) 200 Gy and (d) 300 Gy

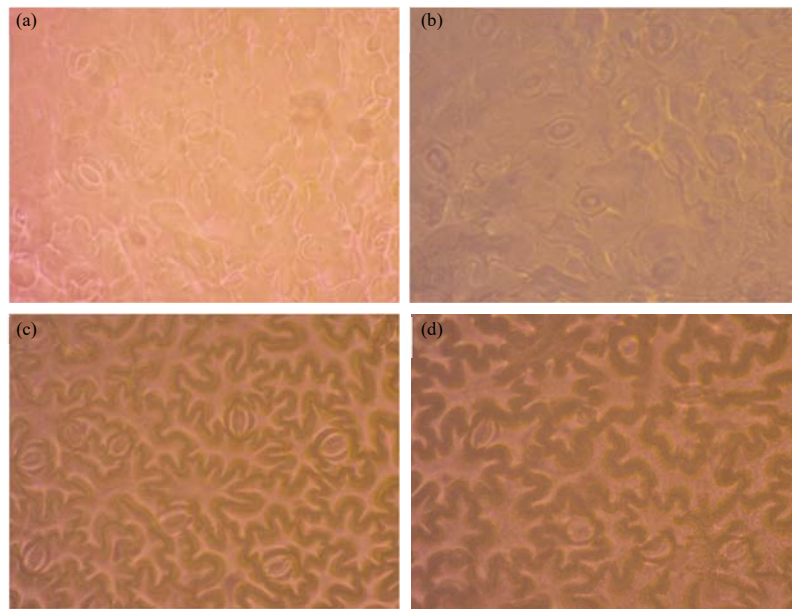


Fig. 2(a-d): Stomata density of chilli leaf, (a) 0 Gy, (b) 100 Gy, (c) 200 Gy and (d) 300 Gy

Table 1: Percentage of growth of chilli seedlings 3 weeks after planting

Irradiation dose (Gy)	Growth (%)
0	85.71
100	71.43
200	57.14
300	57.14

Table 2: Effect of gamma irradiation on morphological characters white local chilli cultivar

Characters	Population			
	0 Gy	100 Gy	200 Gy	300 Gy
Plant height (cm)	44.35±1.48	48.35±0.21	25.60±2.12*	17.90±0.82**
Dichotomous height (cm)	29.90±1.41	34.30±2.69	22.75±2.62	15.20±1.28*
Leaf area (cm ²)	16.20±1.86	25.99±2.48*	7.28±0.9**	5.79±1.13**
Stomata density (mm ²)	204±31.70	83.34±28.87**	133.33±16.67	88.89±9.62*

300 Gy populations had a lower level of stomata density compared to the control plant population (0 Gy). The low level of stomata density and supported by broad leaves really supports the processes of photosynthesis and transpiration in a population of 100 Gy.

The genetic manipulation of stomatal density could be one of the most promising strategies for breeders to improve stress tolerance in crops. Water use efficiency could be improved by decreasing stomatal transpiration without causing a reduction in CO₂ uptake under osmotic stress conditions¹⁴.

In various studies¹⁵⁻¹⁷, significant alterations in traits have been observed, for instance in sesame plants (*Sesamum indicum*), where a dose of 300-400 Gy is capable of

generating mutant plants exhibiting the desired traits. Similarly, low-dose gamma ray irradiation at 50 and 100 Gy has been found to enhance and improve quantitative traits in wheat (Durum wheat). Research on soybean mutant lines obtained the mutant lines that are candidates for resistance lines against stem rot disease *Agroathelia rolfsii*.

Gamma ray exposure affects the variety of characteristics seen in the first generation (M1) of plants via their physical traits. In mutation breeding, especially when working with sizable populations, evaluating the mutagenic impact and enhancing genetic variability aids breeders in promptly identifying populations that have been effectively treated¹⁸. This facilitates the acquisition of desired traits and broadens the range of options for selection.

CONCLUSION

Mutation induction with dose range of 0 to 300 Gy affects the growth and morphology diversity of chilli plants compared to plants without irradiation. Further progress in this study allows the use of mutation techniques to modify and improve the characteristics of chilli plants so as to produce superior plants that have high production and adapt to sub-optimal environments.

SIGNIFICANCE STATEMENT

The purpose of this study was to find out information about changes in morphological characters in the early vegetative period of chilli plants caused by physical mutation treatment. The objective of this study was to enhance the genetic diversity of chilli plants, particularly since they are self-pollinated crops with limited genetic variability. Future study aims to develop chilli plants to obtain desired traits, thereby increasing crop yields and increasing resistance to biotic and abiotic stress.

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