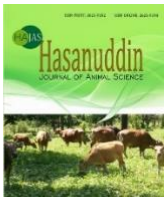

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Morphological Response of *Indigofera zollingeriana* Mutant 2 Plants Irradiated with Gamma Rays in Coastal Areas

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ABSTRACT

Coastal areas are potential land for the cultivation of feed crops to support the availability of livestock forage. *Indigofera zollingeriana* plants have a high tolerance to salinity stress. Gamma irradiation technology, which can change the genetic structure of plants, is expected to increase plant tolerance. This study was conducted to determine the morphological response of *Indigofera zollingeriana* Mutant 2 plants irradiated with gamma rays in coastal areas. This study used a complete randomized design (CRD) method with ten treatments and six replications. The treatments consisted of P0 (control/without irradiation); P1 (*Indigofera* M2 50 Gy); P2 (*Indigofera* M2 100 Gy); P3 (*Indigofera* M2 150 Gy); P4 (*Indigofera* M2 200 Gy); P5 (without irradiation + salinity stress); P6 (*Indigofera* M2 50 Gy + salinity stress); P7 (*Indigofera* M2 100 Gy + salinity stress); P8 (*Indigofera* M2 150 Gy + salinity stress); and P9 (*Indigofera* M2 200 Gy + salinity stress). The analysis of variance showed that different doses of gamma irradiation and salinity stress had a significant effect (P<0.05) on stem diameter and number of flowers of *Indigofera zollingeriana* plants. Irradiation dose of 150 Gy with salinity stress during seedling can grow higher leaves when planted in coastal areas.
Keywords: Salinity Stress, *Indigofera zollingeriana*, Gamma Irradiation

INTRODUCTION

The utilization of coastal areas in the livestock sector is very potential to be empowered, especially forage cultivation. The development of marginal land as forage cultivation can support the availability of animal feed, which is still influenced by the season. Sustainable forage provision requires plants tolerant of marginal environmental conditions such as coastal areas. Soil degradation, salinity, and lack of nutrients in coastal areas can cause inhibition of plant productivity and sustainability [1]. Salinity stress will produce plants that vary according to the ability of the genotype [2]. The plant's growing environment will affect the morphological and anatomical structure, indicating a plant that can survive in an extreme environment.

Genetic diversity of plants that are tolerant to environmental stress can be obtained through plant genetic engineering with several methods such as crossing, gamma in vitro culture technology [3], and physical mutations that are safer from residues, namely light irradiation [4]. Various doses of gamma irradiation can create different plant varieties [5]. Plant diversity caused by genetic factors is more dominant than environmental factors [6]. Seed irradiation with gamma rays has been known to affect plant growth through changes in cytology, genetics, biochemistry, physiology, and morphogenetic modifications in cells and tissues [7].

Quality feed consumed will affect the survival and productivity of livestock, so it is necessary to have forage species whose growth is not influenced by the season. One alternative to developing forage production is to develop *Indigofera* plants. The characteristics of irradiated *Indigofera zollingeriana* plants have tolerance to saline land. Research conducted by Nadir et al. [8] showed that *Indigofera* was able to survive in conditions of NaCl concentration of as much as 50 mM/2.925 g/L of water and NaCl concentration of as much as 100 mM/5.85 g/L of water despite the salt stress effect that inhibits the growth of *Indigofera* plant seedlings. The successful development of irradiated *Indigofera zollingeriana* is expected to maintain the superiority of its genetic traits in different growing environment conditions. This study aims to determine the growth ability of irradiated *Indigofera zollingeriana* plants that have been given salinity stress in coastal areas.

MATERIALS AND METHODS

The materials used in this study were second-generation seedlings of *Indigofera zollingeriana* from the first generation of gamma irradiation with different doses cultivated at the *Indigofera zollingeriana* Nursery, Faculty of Animal Husbandry, Hasanuddin University. *Indigofera* M2 seedlings were given salinity stress using technical NaCl at a dose of 50 mM, which is equivalent to 2.925 g/litre of water. The design was completely randomized (CRD), consisting of 10 treatments and six replications. The treatments were P0 (Control/Without irradiation); P1 (*Indigofera* M2 50 Gy); P2 (*Indigofera* M2 100 Gy); P3 (*Indigofera* M2 150 Gy); P4 (*Indigofera* M2 200 Gy); P5 (Without irradiation + salinity stress); P6 (*Indigofera* M2 50 Gy + salinity stress); P7 (*Indigofera* M2 100 Gy + salinity stress); P8 (*Indigofera* M2 150 Gy + salinity stress); and P9 (*Indigofera* M2 200 Gy + salinity stress).

Research Procedure

Gamma-irradiated *Indigofera zollingeriana* M2 seedlings were planted in Untia Fishery Harbour for 45 days at a distance of 1.5 meters from the seaside. Plant cultivation was carried out by paying attention to water availability, applying organic fertilizer, and controlling weeds that grew around the plants.

Observed Parameters

The parameters observed in this study were stem diameter, number of flowers, number of leaves, and plant height. Data were collected 45 days after planting. The diameter of the stem was measured using a digital vernier 10 cm above the ground, the number of flowers and compound leaves was counted collectively, and the height of the plant was measured using a measuring tape starting from the base of the plant to the tip of the leaves.

Data Analysis

Data were analyzed using analysis of variance based on a completely randomized design, with each treatment repeated six times, resulting in 60 treatment combinations. Statistical analysis was carried out with SPSS Software (Version 16.0). If the effect of treatment was significantly different in the analysis of variance, then further test was continued with Duncan's test.

RESULTS AND DISCUSSIONS

The treatment of salinity stress and gamma irradiation with different doses showed a significant effect on stem diameter and number of flowers but not significantly different on the number of leaves and flowers, as seen in Table 1.

Table 1. Stem Diameter, Number of Flowers, Number of Leaves, and Plant Height of *Indigofera zollingeriana* Irradiated and Given with Salinity Stress

Treatment	Stem diameter (cm)	Number of Flower	Number of leaves	Plant height (cm)
P0	13.83 ± 2.31^{a}	85 ± 6.43^{ab}	217	114.67
P1	14.80 ± 1.40^{abc}	89 ± 16.20^{ab}	216	130.33
P2	14.27 ± 0.57^{ab}	104 ± 64.08^{b}	256	124.33
P3	15.37 ± 1.11^{abcd}	58 ± 16.62^{abc}	187	118.00
P4	17.33 ± 1.89^{cd}	51 ± 3.21^{bc}	255	116.67
P5	15.27 ± 1.47^{abc}	69 ± 27.06^{abc}	244	122.33
P6	15.20 ± 0.89^{abc}	32 ± 21.28^{c}	334	109.67
P7	17.23 ± 2.03^{bcd}	62 ± 19.50^{abc}	415	129.00
P8	18.27 ± 2.25^{d}	41 ± 6.66^{abc}	232	121.67
P9	15.87 ± 0.35^{abcd}	47 ± 12.77^{bc}	378	138.33

Source: Primary Data, 2023

Description: P0 (Control/without irradiation); P1 (50 Gy); P2 (100 Gy); P3 (150 Gy); P4 (200 Gy); P5 (Without irradiation and given salinity stress); P6 (50 Gy and was given salinity stress); P7 (100 Gy and was given salinity stress); P8 (150 Gy and was given salinity stress); and P9 (200 Gy and was given salinity stress). Different superscripts in the same line show significantly different (P<0.05)

Based on the results of the analysis of variance on the observation of the diameter of the stem of the second mutant *Indigofera zollingeriana* plants, it is known that the treatment of gamma irradiation dose of 200 Gy (P4) showed a significant effect (P<0.05) on the control treatment (P0) and *Indigofera* M2 100 Gy (P2). The existence of a significant effect on *Indigofera* M2 plants indicates that as the dose of irradiation increases, it can accelerate the enlargement of plant stem diameter in coastal areas as a form of environmental adaptation. Genetic modification by the gamma irradiation can produce better plant characteristics than the original species [9]. Table 1 shows that the diameter of *Indigofera* M2 dose of 150 Gy and salinity stress (P8) on early vegetative growth of *Indigofera zollingeriana* plants has a significant effect (P<0.05) on *Indigofera* without irradiation (P5) and *Indigofera* M2 50 Gy and has been given salinity stress (P6) in the seedling phase. However, it showed no significant difference in *Indigofera* M2 with 150 Gy and 200 Gy doses.

Different appearances and genetic changes depend on how much the growing environment changes that occur in plant genotypes [10]. The stem diameter of *Indigofera zollingeriana* M2 plants with a dose of 200 Gy (P4) and a dose of 150 Gy that had been given salinity stress (P8) were 17.33 cm and 18.72 cm, respectively, which did not have a significant effect (P>0.05). This indicates that cultivating *Indigofera* M2 plants on coastal land requires plant seeds that have been adapted in the nursery phase. A large stem diameter suggests the strength of the stem in supporting the plant [11]. However, research conducted by El-Khateeb *et al*. [12] found that a higher irradiation dose can reduce the stem's diameter. The ability of plants to absorb water will encourage cell walls and membranes to increase in size [13]

During the growth and development stage, morphological changes will occur, followed by physiological changes. According to Yan *et al*. [14], physiological and metabolic adjustments of plants are carried out by changing their morphology in response to environmental stress. The main characteristic of plants in the juvenile phase is their inability to form flowers and fruits, even though ecological conditions allow flowering. Flowers are reproductive organs that are important in plants [15]. The analysis of variance showed that the number of flowers of *Indigofera* M2 plants with a dose of 100 Gy (P2) had a significant effect (P < 0.05) on *Indigofera* M2 with a dose of 200 Gy. However, there is no significant effect on *Indigofera* M2 that has been given salinity stress in the nursery phase.

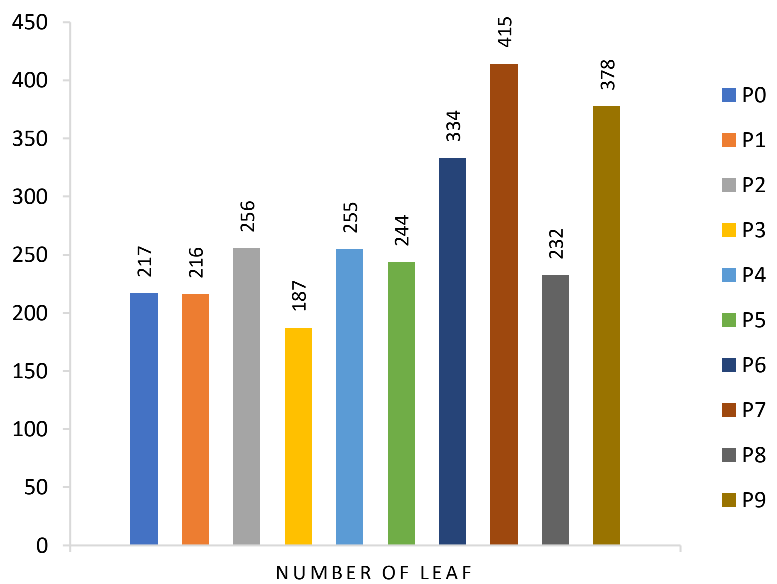

Figure 1. Graph of doses of irradiation and salinity on the number of leaves

The initial vegetative growth of the *Indigofera zollingeriana* second mutant subjected to salinity stress was inhibited. Plant growth is initially affected due to a decrease in soil water potential, leading to plant death due to a rapid increase in salt concentration in the cytoplasm [16]. According to Elizar *et al*. [17], the higher the dose of gamma irradiation given to kenikir plants can affect the length of the flowering process in plants due to the physiological damage caused. Plants that experience heat stress also increase the biosynthesis of several hormones to regulate their stress response [2]. The number of flowers of the second *Indigofera* mutant grown on coastal land varied [18]

Leaves are the main vegetative organ for photosynthesis and transpiration, which shows one of the effects of salinity stress. So, leaf growth is considered an important parameter to evaluate plant tolerance. It can be seen that there is no natural effect on each treatment, but the treatment of P6, P7, and P9 have several leaves that are above average. The vegetative growth response of plants to salinity will be reduced due to a decrease in soil water potential due to the effects of water stress. It can be regulated by inhibitory signals from the roots. When excessive amounts of salt enter the transpiration stream, it will eventually damage transpiring cells and reduce further growth [19]. Low turgor pressure will inhibit metabolism, impacting the growth rate seen in the narrowing of the leaf area [20].

Plant cultivation in the nursery phase by applying salinity can suppress vegetative growth, such as abnormalities in leaves, namely changes in leaf color and necrosis at the tip of the leaf [21]. The number of leaves does not differ due to the accumulation of Na + and Cl- in old leaves, so the leaves will quickly age and fall, even dying before harvest [22]. Salt reduces the ability of plants to absorb water and causes slow growth. The content of potassium nutrients in the soil affects the development of leaves so that the leaves do not fall easily [23]. Cowpea plants affected by salinity do not experience a different number of leaves because salinity suppresses plant growth and changes slowly [24]. Soil that contains salt content inhibits plant growth, one of which is the multiplication of the number of leaves [25].

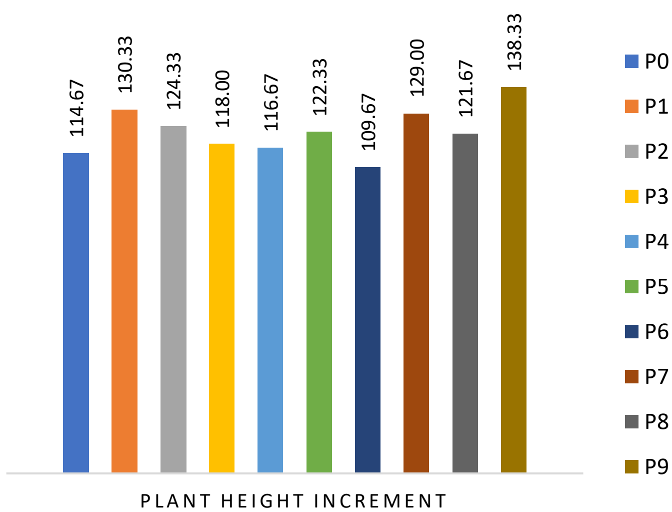

Figure 2. Graph of doses of irradiation and salinity on plant height

The provision of salinity does not have a real effect on the average height of the second mutant *Indigofera zollingeriana* plants. The increase in the dose of gamma irradiation showed no significant increase in morphological growth in each treatment. This is due to the response of plants experiencing hyperosmotic to inhibit the growth of *Indigofera zollingeriana* second mutant plants. According to Pangesti and Ratnawati [26], the higher the dose of gamma-ray treatment, the less optimal plant height is obtained. It can be seen in Table 1 that the plants that were not given the highest saline stress were 130.33 cm at a dose of 50 Gy while after being given saline stress, 138.33 cm at a dose of 200 Gy. The absence of significant differences has been reported by Due *et al*. [27], who found that the higher the irradiation dose, the more plant height parameters will be reduced. Increased physiological growth of tomato plants occurs due to gamma-ray irradiation [28].

Morphological changes are caused by external pressures such as salt stress. Nutrient imbalance and ion toxicity in soil-soluble salts can increase osmotic pressure, reducing water absorption by roots and decreasing plant growth [29]. The growth rate of plant height, number of tillers, and dry weight will decrease with increasing salt content in the growing medium [30]. This is supported by Dalimunthe *et al*. [31], who states that plant growth and height are suppressed as the dose of irradiation increases. Changes in plant height are caused by cell division and elongation [32].

CONCLUSIONS

Based on the data obtained, it can be concluded that the treatment of gamma irradiation dose and salinity stress during the seedling process significantly affects stem diameter and number of flowers. Gamma irradiation dose of 150 Gy and salinity stress can increase stem diameter, plant height, and number of leaves and inhibit flower growth.

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