# Effect of sodium azide mutagen on balsam plant (*Impatiens balsamina)*

**Bui Minh Sang[1,2], Tran Dinh Gioi[2], Nguyen Thi Pha[1*]**
[1]*Can Tho University*
[2]*Cuu Long Delta Rice Research Institute*

# Ảnh hưởng của chất gây đột biến sodium azide trên cây Phượng Tiên *(Impatiens balsamina)*

**Bùi Minh Sang[1,2], Trần Đình Giới[2], Nguyễn Thị Pha[1*]**
[1]*Đại học Cần Thơ*
[2]*Viện Lúa Đồng bằng sông Cửu Long*
**Corresponding author: ntpha@ctu.edu.vn*

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***ABSTRACT***

*Impatiens balsamina is a popular ornamental plant in Vietnam. This study was conducted to evaluate the effects of different concentrations of sodium azide (SA) on mutagenesis and the creation of unique variants to increase the genetic diversity of this ornamental plant. Seeds were treated with different concentrations of SA (0%, 0.01%, 0.03%, and 0.05%) for three days to determine the rate of seed germination, plant survival, growth, agronomic traits, mutation frequency, mutation effectiveness, and efficiency. The results showed that an increase in SA concentration (0.03% and 0.05% SA) resulted in a delay in germination and a decrease in plant survival. The height of plants treated with 0.05% SA was lower than that of the control. The test concentrations tended to increase leaf length and leaf area compared to the control, but had little effect on leaf width. Nevertheless, higher concentrations (0.05% SA) markedly slowed the initiation of buds and the time it took for flowers to bloom. Some mutant individuals produce flowers that significantly differ from the control variety in size, color, and structure. Within the range of SA treatment applied, 0.03% SA for three days is the most suitable treatment for mutant Balsam inducement, with the highest mutation effectiveness and efficiency of 4.79% and 0.52%, respectively. These results suggest that SA can function as a potent mutagenic agent that causes advantageous variation in Balsam when administered at the appropriate concentrations.*

***TÓM TẮT***

*Cây Phượng Tiên (Impatiens balsamina) là một loài cây cảnh được trồng phổ biến ở Việt Nam. Nghiên cứu được thực hiện nhằm đánh giá ảnh hưởng của các nồng độ natri azide (SA) khác nhau đến hiệu quả gây đột biến và tạo các biến dị độc đáo làm gia tăng sự phong phú nguồn gen của loài cây hoa cảnh này. Hạt giống được xử lý bằng các nồng độ SA (0%, 0,01%, 0,03% và 0,05%) trong ba ngày để xác định tỷ lệ nảy mầm của hạt, sức sống cây, sinh trưởng, đặc điểm nông học, tần số, hiệu quả và hiệu suất đột biến. Kết quả cho thấy, nồng độ SA tăng (0,03% và 0,05% SA) làm chậm quá trình nảy mầm và giảm tỷ lệ sống sót của cây. Chiều cao cây ở nồng độ 0,05% SA thấp hơn so với đối chứng. Các nồng độ thử nghiệm có xu hướng làm tăng chiều dài và diện tích lá so với đối chứng, nhưng ít ảnh hưởng đến chiều rộng lá. Tuy nhiên, nồng độ cao hơn (0,05% SA) làm chậm thời gian hình thành nụ và ra hoa. Một số cá thể đột biến có sự khác biệt đáng kể so với giống đối chứng về kích thước, màu sắc và cấu trúc hoa. Trong phạm vi nồng độ SA được áp dụng, liều lượng 0,03% SA trong 3 ngày  phù hợp để gây đột biến ở cây Phượng Tiên với hiệu quả và hiệu suất đột biến cao nhất lần lượt là 4,79% và 0,52%. Những kết quả này cho thấy SA có thể hoạt động như một tác nhân gây đột biến mạnh, tạo ra sự biến đổi có lợi ở cây Phượng Tiên khi được sử dụng ở nồng độ thích hợp.*

## 1. INTRODUCTION

Balsam (*Impatiens balsamina*) is an annual herbaceous plant belonging to the Balsaminaceae family, primarily distributed in warm climates and commonly known as rose balsam or garden balsam [1, 2]. It is native to southern Asia, especially in India and Myanmar, and is now widely cultivated in subtropical parts of the world [3]. In Vietnam, *Impatiens balsamina* is widely grown for both ornamental and medicinal purposes [4]. However, they were primarily cultivated in home gardens by collecting and re-sowing seeds from trees with desirable flowering traits. This method of breeding leads to minimal introduction of new genetic material, which restricts the variety available for breeding. As a result, the genetic diversity of balsam varieties has been steadily declining, limiting their ability to adapt to changing environmental conditions. Mutation is one of numerous plant breeding techniques to improve some traits or to create variations that do not occur naturally [5]. The nature and effect of mutations are of fundamental importance to the evolutionary process. Applications of mutagens include dissecting the genetic basis of trait variation, inducing desirable traits in crops, and understanding the nature of genetic load [6].

Genetic modification and mutation induction have been used to introduce new and improved traits in ornamental plants, which have high commercial interest for producers and consumers. These traits include the improved floral anatomy and morphology, new floral colors, early flowering, enhanced fragrance or longevity, stress tolerance, and disease resistance [7]. Sodium azide (SA) is a potential mutagen that is used for inducing genetic changes in crop plants worldwide. This compound, after being metabolized by the plant, disrupts the normal activity of the cell by affecting the intracellular calcium levels and subsequently reduces the levels of calmodulin and production of Adenosine Triphosphate (ATP). Low levels of ATP disrupt spindle fiber organization and movement of chromosomes during mitosis. Furthermore, SA interferes with the respiration process within the cells by inhibiting various enzyme activities that include peroxidase, catalase including cytochrome oxidase, proton pump, and cell cycle inhibition [8]. Sodium azide is mainly used to induce gene mutants that regulate essential traits. These mutations may also clear up the gene function of mutant phenotypes [9]. When seeds are treated with SA, it can directly impact the expression and regulation of genetic material within plant cells. This can lead to phenotypic differences at any stage of the plant's growth. However, at higher doses of SA, it can cause significant deleterious effects, including reduced germination rate [10, 11], altered agronomic and growth [12-14], malformations in reproductive organs [15, 16], and even plant death [17, 18]. Growth and agronomic characteristics were commonly used as indirect indicators to evaluate the extent of biological damage caused by mutagenic agents. Monitoring these characteristics is essential for determining the appropriate treatment dose threshold before selecting individuals that exhibit valuable mutations in flower morphology. While several studies have assessed the impact of sodium azide (SA) on germination rates and some morphological traits of ornamental plants, the effects of SA have not been examined in the balsam plant.

The objective of the study was to investigate the effects of sodium azide on germination rate, plant survival, and agronomic characteristics of *Impatiens balsamina*. Additionally, this study was to determine the optimal mutagen concentration based on mutagenic efficacy and efficiency, record induced changes in floral morphology.

## 2. RESEARCH METHODS

### 2.1. Plant material

Seeds of Balsam (*Impatiens balsamina*) with purple petals were collected from a local population at the Cuu Long Delta Rice Research Institute, Can Tho city, Viet Nam. These seeds were carefully maintained over three generations under automated conditions to ensure optimal phenotypic similarity before the induction of mutations. During the selection process, seeds with uniform size and the viability of the seeds were tested by soaking seeds in water, and floating seeds were discarded, while sinking seeds were treated to reduce experimental errors due to seed health [19]. Initial germination tests indicated that the seeds had a germination rate exceeding 98% before treatment. The time and place of the research were January to November 2024 at the greenhouse and laboratory of the Institute of Food and Biotechnology, Can Tho University, Can Tho city, Viet Nam. The experiment was conducted in a greenhouse with an average temperature of 28 ± 2°C, humidity levels ranging from 70% to 80%, and natural light providing approximately 12 hours of daylight per day.

### 2.2. Mutagenesis method

Fabric bags (3 x 3 cm) were used to pack 30 seeds before soaking in warm water [4]. After being soaked in water for 6 hours [3], the seed bags were taken out and allowed to dry naturally at room temperature, which should be around 25 - 30°C [20]. During sodium azide treatment, seeds were placed in small cloth bags, each containing 10 seeds, to facilitate soaking and retrieval after treatment. The seed bags were then soaked in 50 ml of the SA solution ($NaN_3$, ≥ 99.5% purity, Sigma-Aldrich, USA) for 3 days with concentrations of 0.01%, 0.03%, and 0.05% [4, 13, 15, 20]. Fifty milliliters of phosphate buffer pH 3 was added to each of the treatments to maintain the sodium azide pH at 3 (SA solution was replaced every 24 hours). The pH of the SA solution was checked prior to treatment and remained stable throughout the three-day treatment period. For the control treatment, the seeds were soaked in distilled water and buffer (pH 3) [21]. The seeds were then washed with tap water to remove excess chemicals and exudates [18]. After washing, all seeds from different treatments were incubated separately on Petri dishes for germination at room temperature (25 - 30°C), then planted in pots (60 x 40 x 40 cm) in the garden conditions. Substrate ingredients include soil, sand, and organic matter with a ratio of 1:1:1 to obtain get M1 plants [22].

### 2.3. Observation parameters

The total number of seeds was sown, and on the seventh day, the germinated seedlings were counted from each treatment [23]. Germination day (days) was defined as the day when 50% of the seeds in each treatment exhibited radicle emergence. The percentage of survival rate was calculated at 15 days after sowing and at the flowering stage (14 weeks old) [4]. The survival rate was calculated by counting the plants that survive, have green stems and leaves, and continue to grow normally at the time of observation. The time to bud initiation and flowering was documented through daily visual observations. Days to bud initiation (days) was the number of days from seeding to the emergence of the first visible flower bud, and days to flowering (days) was the number of days from sowing to the first flower's full opening. The plant height (cm) was measured at the time of flowering using a digital ruler [23], and the stem diameter (mm) was calculated from its circumference, which was measured using a ruler [19]. Toupview software (version 4.11.19728) was used to measure the length and width of leaves (cm), and leaf area ($cm^2$) [4]. The frequency, effectiveness, and efficiency of mutations were calculated using the standard method for evaluating mutagenic responses [24].

### 2.4. Data analysis

Data were collected from three replicates, with each replicate consisting of 10 plants within a specific treatment. The recorded data were entered into Microsoft Excel 2013 for initial organization and were then analyzed using Minitab 16. Descriptive statistics and mean comparisons were conducted using Duncan's Multiple Range Test (DMRT) at a significance level of 5%.

## 3. RESULTS AND DISCUSSION

### 3.1. Effect of Sodium azide on germination rate and plant survival

Data on the germination rate were shown in Figure 1A. As the concentration of SA increases, the rate of germination tends to decrease. In the mutant treatments, the highest percentage of germination was recorded at 0.01% SA and the control (100%). The concentration of 0.03% SA reduced the germination rate to 96.7%, and the lowest germination percentage was recorded at 0.05% SA (93.3%). The cause may be due to the concentration of the mutant agent being increased to very high levels, and the application period is prolonged, negative effects may occur [25]. According to the report of Al-Qurainy and Khan, reduction in seed germination as a result of mutagenic treatments has been explained due to delayed or inhibited physiological and biological processes including enzyme activities required for seed germination [26]. In addition, reduced seed viability may be due to inhibition of essential physiological processes such as endocrine imbalance and mitosis inhibition [27].

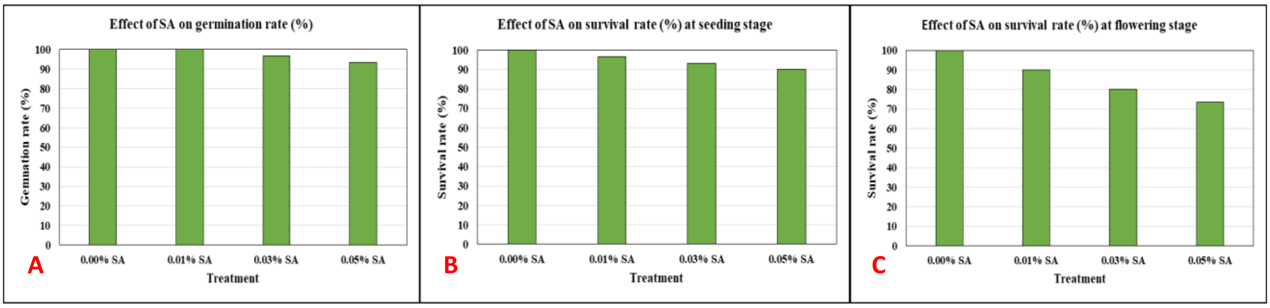

**Figure 1. Effects of sodium azide on germination and survival of *Impatiens balsamina***

*(A) Germination rate at 7 days after sowing, (B) Survival rate at seeding stage (15 days after sowing), (C) Survival rate at flowering stage (14 weeks)*

Observing the survival rate at different growth stages can reveal the impact of the chemical mutagen on plant viability. After 15 days of sowing, the highest number of plants was obtained at 0.01% SA (96.7%), and the lowest growing plants were observed at 0.05% SA (90.0%) (Figure 1B). Survival at the flowering stage due to different mutagenic doses ranged from 73.3% (0.05% SA) to 90.0% (0.01% SA), lower than that of 100% in the control treatment (Figure 1C). The survival rate of *Impatiens balsamina* decreases during the flowering stages as compared to the rate 15 days after sowing, indicating that the effects of chemical mutagens last throughout the plant's growth and development. Sodium azide is a potent chemical mutagen that is used worldwide for inducing genetic changes in crop plants. This compound, after being metabolized by the plant, disrupts the normal activities of the cells by affecting the intracellular calcium levels and subsequently reducing the levels of calmodulin and the production of ATP. Reduced ATP levels impair mitotic processes by disturbing spindle fiber formation and chromosome movement during cell division [8]. This cellular disruption contributes to a reduced plant's survival, whereas an increase in the mutagen concentration resulted in a greater inhibition of plant growth [17]. Furthermore, mutagens have the capacity to accelerate or halt mitotic divisions, contingent upon the location and nature of the mutation [28]. Previous research results indicated that *Abelmoschus esculentus* treated with a combination of radiation and SA either died before reaching the flowering stage or survived to maturity and flowering but was stunted [18].

### 3.2. Influence of Sodium azide on some growth characteristics

The results presented in Figure 2A illustrate the impact of SA on the germination time of Balsam seeds. The average germination time for seeds treated with SA ranged from 8.1 to 8.5 days, compared to the control average of 8.8 days. This finding aligns with previous reports indicating that Balsam seeds typically germinate within 7 to 14 days [2]. Several studies on other crops have demonstrated that a decrease in the seed germination rate is often accompanied by an increased number of days to germination. This phenomenon is attributed to changes in physiological, biochemical, and biological processes as SA concentrations rise

[29]. Specifically, higher doses of SA can hinder ATP biosynthesis, resulting in a decreased availability of ATP molecules; as a result, the germination rate may slow down [20]. In this study, treatment with SA at concentrations ranging from 0.01% to 0.05% did not significantly influence the time it took for *Impatiens balsamina* seeds to initiate germination. However, at higher concentrations of SA, the data distribution was broader, indicating that some seeds germinated more slowly. This suggests that higher levels of SA may slightly inhibit germination initiation in certain seeds.

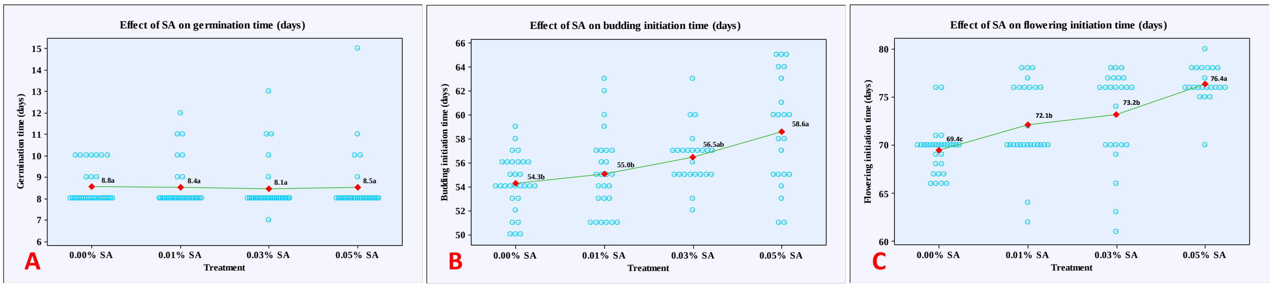

**Figure 2. Effects of sodium azide on some growth characteristics of *Impatiens balsamina***

*(A) germination time (days), (B) Budding initiation time (days), (C) Flowering initiation time (days)*

The data presented in Figures 2B and 2C illustrate the time to bud initiation and flowering initiation after treatment with SA. The 0.01% SA treatment (55.0 days) and the 0.03% SA treatment (56.5 days) did not significantly differ, whereas the 0.05% SA treatment (58.6 days) resulted in a significantly delayed bud initiation compared to the control (54.3 days). Regarding flowering initiation time, two individuals from the 0.01% SA treatment and two from the 0.03% SA treatment showed early flowering (≥ 5 days earlier than the control) (Figure 2C). However, the general trend of the mean values indicates that all SA treatments tended to delay flowering in *Impatiens balsamina*, with flowering initiation times ranging from 72.1 to 76.4 days, compared to the control (69.4 days). The transition from the vegetative phase to the reproductive phase is primarily governed by genetic factors, and flowering is influenced by changes in several physiological aspects. During this transition, appropriate changes take place in the meristem [30]. The induction of sodium azide has been shown to facilitate the identification of changes in various quantitative traits. This is evidenced by frequency distributions as well as changes in the mean and variance of the treated populations. The results of this study indicate that sodium azide significantly increased the variation in the number of days to flowering by 50% and in the days to maturity in *Triticum aestivum* [29]. The delay in flowering may be attributed to disruptions in the biochemical pathways that are crucial for floral initiation [3].

### 3.3. Influence of Sodium azide on some agronomic characteristics

Effects of mutation treatment and its related data were presented in Table 1. The results showed that the plant height of balsam plants in different treatments ranged from 71.8 to 78.4 cm and differed significantly ($p < 0.01$). The concentration of 0.05% SA gave the lowest plant height (71.8 cm), which was significantly different from the remaining treatments. The treatments of 0.01% and 0.03% SA had plant heights that were not significantly different from the control without SA treatment. Reduction in plant height may be considered a desirable characteristic by aggressive breeding programs that seek to produce compact and aesthetically balanced plants. Sodium azide was reported to reduce plant height at maturity in both tomato and *Capsicum annuum* [12, 13]. Similarly, the dwarf mutation phenomenon was reported in Japonica rice [31] and semi dwarf was reported in Cowpea [10]. The decrease in plant height caused by sodium azide at high concentrations could be attributed to damage to meristematic tissues, metabolic disturbances, and physiological variations [32]. An increased growth inhibitor concentration, decreased auxin concentration, or inhibition of auxin synthesis, is also the cause of reduced plant height [15].

**Table 1. Effects of sodium azide on some agronomic characteristics of *Impatiens balsamina***

| Treatment | Plant height (cm) | Stem diameter (mm) | Leaf length (cm) | Leaf width (cm) | Leaf area ($cm^2$) |
|---|---|---|---|---|---|
| Control | $75.3^a$ | 14.4 | $8.9^b$ | 1.9 | $9.3^b$ |
| 0.01% SA | $78.3^a$ | 14.2 | $9.7^b$ | 2.0 | $11.7^a$ |
| 0.03% SA | $78.4^a$ | 14.2 | $10.4^a$ | 2.0 | $12.0^a$ |
| 0.05% SA | $71.8^b$ | 14.6 | $9.8^{ab}$ | 2.1 | $11.9^a$ |
| *P* | *0.000* | *0.891* | *0.001* | *0.379* | *0.055* |
| *CV (%)* | *7.44* | *13.81* | *13.41* | *12.75* | *22.63* |

*Note: CV (%): coefficient of variation, P: probability value, and values of any traits assigned the same superscript letter in a column are not significantly different at the 5% significance level.*

The stem diameter measured after SA treatment at the flowering stage varied from 14.2 to 14.6 mm (Table 1), and there was no significant difference in stem diameter between the treatments and the control (14.4 mm). This indicates that sodium azide, at the tested concentrations, which are relatively low in toxicity, did not result in any measurable phytotoxic effects on the stem diameter of *Impatiens balsamina* during this growth stage. This resilience may suggest that the plant is less affected by mutagenic stress. This finding differs from earlier reports in other species. For instance, in *Capsicum annuum*, the lowest stem diameter values were recorded 12 weeks after planting in both the "Shombo" and "Tatase" pepper cultivars when treated with a 0.03% SA [13]. The ability of stem tissue to maintain a stable diameter under mutagenic stress may reflect an inherent tolerance or compensatory growth mechanism.

Determining the threshold effect of sodium azide on morphological characteristics such as leaf length, width, and area is necessary to comprehend the physiological response and photosynthetic capacity of *I. balsamina*, as sodium azide can either improve energy synthesis [8]. The findings in Table 1 demonstrate that while SA concentrations between 0.01% and 0.03% had no discernible effect on leaf width, they did alter the length and area of the leaves of *Impatiens balsamina*. In this study, the width of the balsam leaves ranged between 1.9 cm and 2.1 cm; however, the differences between the treatments were not statistically significant. The mean leaf length from 0.03% SA treatment (10.4 cm) was significantly longer than the control (8.9 cm) and 0.01% SA treatment (9.7 cm), but there was no significant difference when compared to 0.05% SA treatment (9.8 cm; Table 1). The average leaf area of *I. balsamina* increased in all treatments with SA (11.7 to 12.0 $cm^2$) compared to the control (9.3 $cm^2$). This result is consistent with earlier research showing that mutagenesis induced by sodium azide can enhance vegetative growth characteristics in a variety of crop species. The ability of sodium azide to cause genetic variation and modify physiological processes associated with growth regulation has been connected to improvements in plant growth, including leaf development and biomass accumulation [12, 26, 29, 32]. The positive response could be due to improved metabolic activity or the stimulation of specific growth-regulating pathways that promote leaf development. This result is consistent with a previous study; sodium azide-induced mutants showed significant improvement in growth traits, including leaf size [14, 32].

### 3.4. Influence of Sodium azide on mutagenic effectiveness and efficiency

Morphological variations must be observed and classified in order to properly evaluate the effects of sodium azide. These morphological variations directly reflect the genetic changes caused by the mutagen, and this information is the basis for calculating the frequency and efficacy of mutations linked to each concentration of treatment. There were noticeable differences between the control plants and the flower-related morphological mutations, according to monitoring. Mutants showed changes in the structure, color, and size of their flowers (Figure 3).

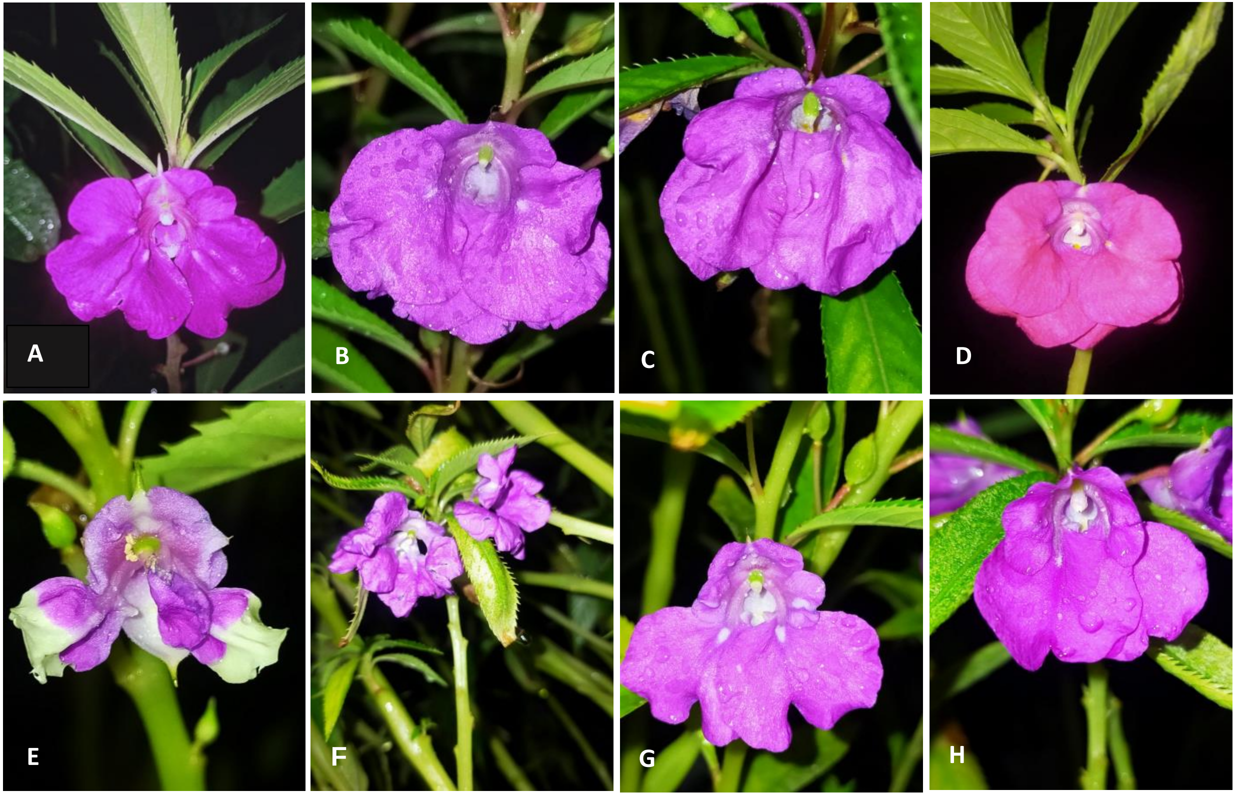

**Figure 3. Representative morphological flower mutations observed in the M1 generation of *Impatiens balsamina* after sodium azide treatment**

*(A) Control plant, (B - C) Mutants showing variation in flower size, (D - E) Mutants with altered flower color, (F - 3) Mutant with modified floral structure*

Various physical and chemical mutagens are used to increase the frequency of mutations and facilitate the selection processes [9]. Mutation rates varied substantially across treatment concentrations (Figure 4A). The highest mutation frequency of 10.34% was observed with 0.03% SA. The mutation frequency for 0.05% SA was recorded at 7.14%, while 0.01% SA showed the lowest frequency of 3.33%. Augmentation of mutation frequency and the alteration of mutation spectrum in a predictable manner remain all-time important aspects of mutation research [33]. The highest mutagenic effect is usually not found at the lowest dose, but at the intermediate dose level, where the agent is potent enough to induce genetic variation but does not exceed the threshold of causing serious cell damage [14, 17]. This trend is consistent with previous results, where mutation frequency can increase with increasing mutation variability within the allowable range [10, 15].

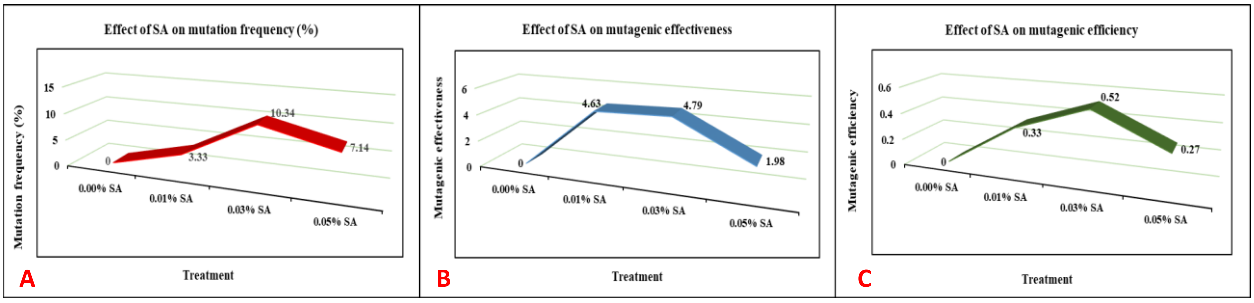

**Figure 4. Effects of sodium azide concentrations on mutation parameters in *Impatiens balsamina***

*(A) Mutation frequency (%), (B) Mutagenic effectiveness, (C) mutagenic efficiency*

The outcome of mutation breeding depends on the mutagenic effectiveness, efficiency, plant material, mutagen dose, and duration [33]. Mutagenic effectiveness and efficiency are the most crucial factors determining the success of mutation breeding, a valuable tool for rapidly enhancing genetic diversity in crops [10]. The data regarding the effectiveness and efficiency of different mutagens are recorded in Figures 4B and 4C. The mutation effectiveness varied between 1.98% and 4.79%, while the mutation efficiency ranged from 0.27 to 0.52. Generally, increasing the SA concentration from 0.01% to 0.03% resulted in higher mutagenesis effectiveness and efficiency. Among the tested concentrations, the 0.03% sodium azide treatment tended to show higher mutagenic effectiveness and efficiency. However, when the SA concentration reached 0.05%, all these parameters decreased. The efficiency of mutagenesis was higher at lower concentrations compared to higher concentrations. This is because higher concentrations significantly increase the likelihood of cell death and injury, which outweighs the rate of mutation [24]. In contrast, treatments with lower concentrations of sodium azide as a mutagen resulted in less biological damage, making them more suitable for inducing desirable mutations in *Helianthus annuus* [15].

## 4. CONCLUSION

When the concentration of SA increases, the rate of germination tends to decrease. The survival rate of Balsam (*Impatiens balsamina*) decreases during the flowering stages as compared to the rate 15 days after sowing. The treatment of 0.05% sodium azide resulted in reduced height, the time to bud initiation, and flowering initiation of Balsam plants, while 0.03% led to an increase in leaf length. No significant effects on stem diameter and leaf width were observed across the treatments. However, the leaf area at every concentration of SA treatment was larger than that of the control treatment. Additionally, the study discovered a number of mutations linked to modifications in the size, color, and structure of flowers. Among the tested concentrations, treatment with 0.03% sodium azide for 3 days is the most suitable treatment for mutant inducement, with the highest mutation effectiveness and efficiency of 4.79% and 0.52%, respectively.

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## REFERENCES

[1]. Reshmi Rajan, KM Shana, VV Vishnupriya, PP Fahmeeda, PP Prajna & E Tamil Jothi (2022). Phytochemical and pharmacological potential of *Impatiens balsamina*. World Journal of Biology Pharmacy and Health Sciences. 12(02): 54-60.

[2]. Sanjar Umarov, Xushbek Ayasov & Fozilbek Mardonov (2025). Balsam impatiens (*Impatiens balsamina* L.) plant's distribution and bioecological and medicinal properties. Theoretical aspects in the formation of pedagogical sciences. 4(8): 200-202.

[3]. Sumit Pal, Anil K Singh & Anjana Sisodia (2023). Effect of physical and chemical mutagens on flowering and seed attributes of balsam (*Impatiens balsamina*). Journal of Pharmaceutical Innovation. 12: 4318-4326.

[4]. Nguyen Thi Pha, Bui Minh Sang, Mai Thanh Thao & Tran Dinh Gioi (2023). Effect of ethyl methane sulfonate on the mutants inducement in balsam plants (*Impatiens Balsamina* Linn.). Asian Journal of Plant Sciences. 22: 628-636.

[5]. Udage AC (2021). Introduction to plant mutation breeding: Different approaches and mutagenic agents. Journal of Agricultural Sciences – Sri Lanka. 16(03): 466-483.

[6]. Chaochih Liu, Giulia Frascarelli, Adrian O Stec, Shane Heinen, Li Lei, Skylar R Wyant, Erik Legg, Monika Spiller, Gary J Muehlbauer & Kevin P Smith (2025). Sodium azide mutagenesis induces a unique pattern of mutations. PLoS genetics. 21(6): e1011634.

[7]. Anne-Laure Boutigny, Nicolas Dohin, David Pornin & Mathieu Rolland (2020). Overview and detectability of the genetic modifications in ornamental plants. Horticulture Research. 7.

[8]. Sajad Hussain Mir & Insha Jan (2023). Sodium Azide as a Mutagen. Biotechnologies and Genetics in Plant Mutation Breeding. Apple Academic Press. 89-121.

[9]. Aras Türkoğlu, Metin Tosun & Kamil Haliloğlu (2022). Mutagenic effects of sodium azide on in vitro mutagenesis, polymorphism and genomic instability in wheat (*Triticum aestivum* L.). Molecular Biology Reports.

49(11): 10165-10174.

[10]. Aamir Raina & Samiullah Khan (2020). Mutagenic effectiveness and efficiency of gamma rays and sodium azide in M2 generation of Cowpea (*Vigna unguiculata* (L.) Walp.). BioRxiv.
DOI: 10.1101/2020.03.09.983486

[11]. Damanpreet Kaur & Amarjeet Kaur (2023). Effect of chemical mutagens on seed germination and seedling traits of jamun. Biological Forum – An International Journal. 15(2): 454-460.

[12]. Adeosun CA, Elem KA & Eze CD (2020). Mutagenic effects of sodium azide on the survival and morphological characters of tomato varieties. Nigerian Journal of Biotechnology. 37(1): 55-62.

[13]. Omeke JO, Ojua EO, Eze NM & Abu NE (2021). Effect of sodium azide induction on morphological traits of shombo and tatase (*Capsicum annuum* L.). Nigerian Journal of Biotechnology. 52(1): 111-117.

[14]. Owoeye HO, Ogundipe VO, Azeez AA & Olawuyi OJ (2025). Sodium azide-induced genetic variation of morphological traits in Capsicum accessions. Scientia Africana. 24(1): 170-190.

[15]. Gehan G Mostafa (2011). Effect of Sodium Azide on the Growth and Variability Induction in *Helianthus Annuus*. International Journal of Plant Breeding and Genetics. 5: 76-85.

[16]. Zhengjing Wu, Sujuan Liu, Bingjie An, Hao Zhang, Jingjing Wu, Chenfang Li & Yuan Long (2024). Mutagenesis and flowering-promoting by sodium azide in vitro culture of *Cymbidium faberi* Rolfe. Preprints. 2024061945.

[17]. Zeinullina Aiym, Rysbekova Aiman, Dyussibayeva Elmira, Zhirnova Irina, Zhanbyrshina Nursaule, Zhunusbayeva Zhazira & Svetla Yancheva (2023). Application of sodium azide for chemical induced mutagenesis of proso millet culture (*Panicum miliaceum* L.). Bulgarian Journal of Agricultural Science. 29(4): 623-631.

[18]. Ati Hassana Maryam & Adamu Abu Kasimu (2016). Effect of combined doses of gamma ray and sodium azide (mutagenic agents) on the morphological traits of some varieties of okra (*Abelmoschus esculentus*). African Journal of Agricultural Research. 11(32): 2968-2973.

[19]. Wiendra Ni Made Sastriyani & Pharmawati Made (2019). Morphological and anatomical changes by colchicine in seedling of *Impatiens balsamina* L. Advances in Tropical Biodiversity and Environmental Sciences. 3: 33-36.

[20]. Anas Hamisu, Bhupendra Koul, Ananta Prasad Arukha, Saleh Al Nadhari & Muhammad Fazle Rabbee (2024). Evaluation of the impact of chemical mutagens on the phenological and biochemical characteristics of two varieties of Soybean (*Glycine max* L.). Life. 14(7): 909.

[21]. Eze JJ & Dambo A (2015). Mutagenic effects of sodium azide on the quality of maize seeds. Journal of Advanced Laboratory Research in Biology. 6(3): 76-82.

[22]. Kumar S, Hariprabha S, Kamalakannan S, Sudhagar R & Sanjeevkumar K (2020). Effect panchagavya on germination and seedling growth of Balsam (*Impatiens balsamina*). Plant Archives. 20(1): 3735-3737.

[23]. Muhammad Aslam Baloch, Tanveer Fatima Miano, Niaz Ahmed Wahocho, Naheed Akhtar Talpur & Abdul Qadir Gola (2018). Germination, vegetative and flowering behavior of Balsam (*Impatiens balsamina* L.) in response to natural photoperiods. International Journal of Environment, Agriculture and Biotechnology. 3(4): 1421-1431.

[24]. Konzak CF, Nilan RA, Wagner J & Foster RJ (1965). Efficient chemical mutagenesis. Radiation Botany. 5: 49-70.

[25]. Dylan K Kosma & Matthew A Jenks (2007). Eco-physiological and molecular-genetic determinants of plant cuticle function in drought and salt stress tolerance. Advances in molecular breeding toward drought and salt tolerant crops. Springer. 91-120.

[26]. Fahd Al-Qurainy & S Khan (2009). Mutagenic effects of sodium azide and its application in crop improvement. World Applied Sciences Journal. 6: 1589-1601.

[27]. Ananthaswamy HN, Vakil UK & Sreenivasan A (1971). Biochemical and physiological changes in gamma-irradiated wheat during germination. Radiation Botany. 11(1): 1-12.

[28]. Rafiul Amin Laskar, Bhaskar Dowarah, Dilip Tamang, Sangeeta Das, Protiva Borah & Aamir Raina (2024). Improving French bean yield potential through induced mutagenesis using EMS and SA. Frontiers in Horticulture. 2: 1288720.

[29]. Srivastava P, Marker S, Pandey P & Tiwari DK (2011). Mutagenic effects of sodium azide on the growth and yield characteristics in wheat (*Triticum aestivum* L. em. Thell.). Asian Journal of Plant Sciences. 10(3): 190-201.

[30]. Jana MK (1962). X-ray induced mutants of *Phaseolus mungo* L. II. Sterility and vital mutants. Genetica Iberica. 14: 71-104.

[31]. Hasegawa H & Inoue M (1980). Effects of sodium azide on seedling injury and chlorophyll mutation in rice. Japanese Journal of Breeding. 30(4): 301-308.

[32]. Neha Naaz, Sana Choudhary, Nazarul Hasan, Nidhi Sharma, Khadiga Alharbi & Diaa Abd El Moneim (2024). Enhancing genetic variability in Trigonella species through sodium azide induction: morpho-physiological and chromosomal amelioration. Frontiers in Genetics. 15: 1378368.

[33]. Goyal S, Wani MR & Khan S (2019). Frequency and spectrum of chlorophyll mutations induced by single and combination treatments of gamma rays and EMS in urdbean. Asian Journal of Biological Sciences. 12(2): 156-163.