

**DECIPHERING THE EFFECTIVENESS OF INDUCED
MUTATION FOR SALINITY TOLERANCE ON
STRAWBERRY (*FRAGARIA X ANANASSA* DUCH.) CV.
CHANDLER AND WINTER DAWN**

Thesis Submitted for the Award of the Degree of

DOCTOR OF PHILOSOPHY

in

Fruit Science

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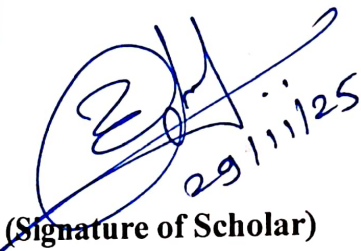


LOVELY PROFESSIONAL UNIVERSITY, PHAGWARA, PUNJAB

2026

DECLARATION

I, hereby declared that the presented work in the thesis entitled “**Deciphering the effectiveness of induced mutation for salinity tolerance on Strawberry (*Fragaria x ananassa* Duch.) cv. Chandler and Winter Dawn**” in fulfilment of degree of **Doctor of Philosophy (Ph.D.)** is outcome of research work carried out by me under the supervision of **Dr. Rajni Rajan**, working as **Assistant Professor**, in the **Department of Horticulture** of Lovely Professional University, Punjab, India. In keeping with general practice of reporting scientific observations, due acknowledgements have been made whenever work described here has been based on findings of other investigator. This work has not been submitted in part or full to any other University or Institute for the award of any degree.



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CERTIFICATE - I

This is to certify that the work reported in the Ph. D. thesis entitled "**Deciphering the effectiveness of induced mutation for salinity tolerance on Strawberry (*Fragaria x ananassa* Duch.) cv. Chandler and Winter Dawn**" submitted in fulfilment of the requirement for the award of degree of **Doctor of Philosophy (Ph.D.)** in the **Department of Horticulture, (Fruit Science)**, School of Agriculture, is a research work carried out by **Mr. Rodge Rahul Ramprasad, 12116704**, is Bonafide record of his original work carried out under my supervision and that no part of thesis has been submitted for any other degree, diploma or equivalent course.

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Place: LPU, Phagwara

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Date:

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LIST OF ABBREVIATIONS

Abbreviated Form	Full Form
MT/ha	Metric tonnes per hectare
mha	Million hectares
Govt	Government
FAO	Food and Agriculture Organization
IAEA	International Atomic Energy Agency
60 Co	Cobalt
Gy	Gray
LD₅₀	Lethal Dose at 50% mortality
%	Per cent
DES	Diethyl Sulphate
Kbp	Kilo basepair
SNP₃	Single Nucleotide Protein
v/v	Volume/volume
mM	Milli molar
Mg/1 or mg/L	Milligram/litre
cm	Centimeter
μM	micromolar
RAPD	Random Amplified Polymorphic DNA
SSR	Single Sequence Repeat
ISSR	Inter Simple Sequence Repeats
REMAP	Retrotransposon-microsatellite amplified polymorphism
w/v	Weight/volume
NaCl	Sodium chloride
Na⁺	Sodium ion
ppm	Parts per million
Gm/1	Gram per liter
EC	Electrical Conductivity
LPU	Lovely professional University
PAU	Punjab Agriculture University
FCRAD	Factorial Complete Randomised Design
Cm	Centimeter
Fig	Figure(s)
G	Gram
et al	et alia and others
°C	Degree Celsius
CV	Coefficient of Variation
C.D.	Critical difference
AFLP	Amplified Fragment Length Polymorphism

SCHOOL OF AGRICULTURE, LOVELY PROFESSIONAL UNIVERSITY, PHAGWARA

Title	: Deciphering the effectiveness of induced mutation for salinity tolerance on Strawberry (<i>Fragaria x ananassa</i> Duch.) cv. Chandler and Winter Dawn
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ABSTRACT

The development of salinity-tolerant strawberry varieties through mutagenesis aims to enhance crop productivity and quality in regions with saline soils or water, where conventional cultivation is challenging. Salinity stress adversely affects plant growth, yield, and fruit quality due to ionic and osmotic imbalances. This experiment concluded that inducing genetic variations using physical mutagen gamma irradiation and chemical mutagen EMS improved traits like Plant height, Chlorophyll content, Leaf area and Fruit weight, Fruit yield, higher proline content, membrane stability, malonaldehyde content, electrolyte leakage and mineral contents imparts tolerance to strawberry against salinity stress. These tolerant varieties not only ensure sustainable strawberry production in affected areas but also support global food security and the agricultural economy under changing environmental conditions.

The present investigation entitled "*Deciphering the effectiveness of induced mutation for salinity tolerance on Strawberry (Fragaria x ananassa Duch.) cv. Chandler and Winter Dawn*" was carried out in the School of Agriculture (Department of Horticulture), LPU, Phagwara during 2022-2024 with the aim to develop salinity tolerant mutant in strawberry cv. Chandler and Winter Dawn using gamma irradiation and chemical mutagens. Two different experiments

were laid out under salinity conditions. Experiments were laid out in four different sections. In the first two sections standardization of gamma irradiation and EMS was conducted to find out the LD₅₀ dose. Secondly, the gamma irradiation experiment was carried out in 4 X 4 factorial complete randomized design consisting of gamma irradiation doses 20 Gy, 30 Gy, 40 Gy and 0 Gy under salinity conditions 0 mM, 50 mM, 75 mM and 100 mM NaCl replicated four times for Winter Dawn and Chandler variety separately. The next trail was conducted using the chemical mutagen EMS in 4 X 4 factorial completely randomized design of EMS doses 1 %, 2 %, 3 % and 0 % exposed with salinity conditions 0 mM, 50 mM, 75 mM and 100 mM NaCl replicated four times for Winter Dawn and Chandler variety separately. The treated runners, along with untreated runners, were planted in plastic pots under a protected polytunnel condition for growth.

The outcome of findings suggested the standardized doses of the physical mutagen gamma irradiation in the Winter Dawn and Chandler varieties which determine the mortality and identify the LD₅₀ dose, which was found to be 40 Gy. Compared to the control, a progressive increase in runners lethality was observed with higher doses of irradiation, with the highest mortality recorded at 40 Gy (0.66%) and the lowest at 20 Gy (0.16%). After standardization, the surviving runners were subjected to salinity stress using NaCl to assess their tolerance. Consequently, the entire experiment was extended to include further observations on physiological, morphological, yield-related, and salinity stress-related parameters up to the 40 Gy dose.

The salinity stress related observations of Winter Dawn variety runners treated with gamma irradiation under salinity stress demonstrated notable outcomes. The result suggested plant height found maximum (28.37 cm) in 30 Gy dose exposed to a 75 mM salinity. While Chlorophyll content was found maximum (58.18) in 30 Gy dose when exposed to 100 mM salinity and fruit weight was observed highest (13.32 gm) in 30 Gy under 50 mM salinity. A significant reduction in malondialdehyde content (2.05 µmol/g) was observed at a 20 Gy dose under 100 mM salinity, indicating its effectiveness in mitigating salinity stress. The lowest electrolyte leakage (24.56%) was recorded at a 30 Gy dose under 50 mM salinity, suggesting improved cell membrane integrity, likely due to tolerance mechanisms induced by the mutagenic dose. Furthermore, the highest proline content (3.82 µmol/g) was recorded in runners treated with a 30 Gy dose under 50 mM salinity, indicating enhanced osmotic adjustment and stress tolerance. In terms of salinity stress-related mineral parameters, the lowest sodium content (24.51 ppm) and the highest potassium ion (27.76 ppm) were recorded

in runners treated with a 30 Gy gamma dose under 50 mM salinity, reflecting improved ionic balance and salinity tolerance.

The gamma irradiation-treated runners of the Chandler variety showed positive effects in morphological observations. The maximum leaf area (24.79 mm) was recorded in runners treated with a 30 Gy dose under 100 mM NaCl stress, while the maximum fruit length (39.33 mm) was observed in runners treated with a 20 Gy dose under 75 mM salinity stress. The highest fruit diameter (32.94 mm) was recorded in plants treated with a 30 Gy dose under 50 mM salinity stress. Additionally, gamma irradiation-treated runners exhibited positive results for physiological, yield-related, and salinity stress-related parameters. Mutagen-treated runners showed malonaldehyde content (1.50 $\mu\text{mol/g}$) in 20 Gy dose against 75 mM NaCl, Proline content (3.29 $\mu\text{mol/g}$) with 30 Gy dose exposed to 50 mM salinity stress, electrolyte leakage in 20 Gy dose exposed to 100 mM salinity level. except for those treated with a 40 Gy dose.

Again, in another experiment the chemical mutagen EMS-treated runners of the Winter Dawn strawberry showed significant results under salinity stress conditions. In terms of morphological observations, the maximum plant height (24.82 cm) was found in runners treated with a 2% EMS dose under 50 mM salinity conditions. In stress related observation best malondialdehyde content (1.87 nmol/g) in 2% EMS dose exposed to 100 mM salinity, proline content (4.22 $\mu\text{mol/g}$) recorded in 1% EMS dose exposed to 50 mM salinity. Leaf scorching was recorded at a minimum of (12.17%) in treated runners with a 2% EMS dose combined with a 75 mM salinity level. Additionally, the highest chlorophyll content (55.74%) was recorded in runners treated with a 2% EMS dose under 75 mM salinity stress. The highest leaf relative water content (80.96%) was observed in runners treated with a 2% EMS dose under 50 mM salinity conditions. Furthermore, the number of buds, flowers, and fruits was highest in runners treated with a 2% EMS dose under induced salinity stress.

However, in the Chandler variety, the EMS mutagen-treated runners exhibited significant changes in salinity stress-related parameters. A significant change in the growth parameter was observed where the maximum plant height (20.96 cm) was found in 1% EMS dose under 75 mM NaCl, chlorophyll content (65.35) was observed in 1% EMS under 100 mM salinity also the maximum fruit weight (14.73 gm) recorded in 1% EMS treated runners under 50 mM salinity level while the yield of fruit was recorded maximum in same mutagen dose induced with 100 mM salinity condition. Sodium levels were lowest in the runners treated with a 2% EMS dose and exposed to 50 mM salinity. This treatment also led to the highest potassium

levels observed in the same mutagen treated runners under the 50 mM salinity condition. Additionally, the minimum malondialdehyde content was recorded in the runners treated with a 1% EMS dose exposed to a 75 mM salinity level (1.53 $\mu\text{mol/g}$) malondialdehyde is a marker of oxidative stress and lipid peroxidation, and its reduction indicates that the EMS treatment effectively mitigated oxidative damage under saline stress. A significant change in proline content (3.94 $\mu\text{mol/g}$) was recorded in 1% EMS dose induced with 75 mM NaCl and electrolyte leakage (17.85 %) was recorded in 2% EMS under 50 mM NaCl. These findings suggest that Gamma doses of 30 Gy in winter Dawn and EMS 1% in chandler has the maximum potential to improve plant traits, ion balance and oxidative stress tolerance, which could be beneficial for improving in strawberry cultivars in salinity affected area.

Keywords: *Physical mutagen; Chemical mutagen; Gamma Irradiation; EMS; Abiotic Stress; Sodium Chloride; Stress; Tolerance; Strawberry*

Chapter - 1

Introduction



The strawberry (*Fragaria* × *ananassa* Duch) is a perennial, herbaceous plant known for its sweet, red, and juicy fruits. It belongs to the Rosaceae family and is cultivated widely across temperate regions. Strawberry plants typically have a low-growing habit, producing trifoliate compound leaves with serrated edges and a green colour. The plant has a shallow root system and spreads through stolons (runners), which form new daughter plants. Strawberry flowers are small, white, and five-petaled, arranged in clusters on short stems, and are typically pollinated by insects (Prohaska *et al.*, 2024). The fruit is technically an aggregate accessory fruit, meaning its fleshy part develops from the flower's receptacle rather than the ovaries. The tiny seeds, or achenes, are embedded on the fruit's surface, contributing to its characteristic appearance and texture. This crop is highly valued for its flavour nutritional and rich vitamin C, flavonoids, Folic Acid, and ellagic acid (Suvalaxmi *et al.*, 2015). Recent data reveals that China is currently the world's leading producer of strawberries, with an output of 3,221,557 metric tons (FAO, 2023). In India, strawberries are cultivated over an area of 1,000 hectares, yielding a total production of 8,000 metric tons (NHB, 2019-20).

In India, soils affected by salinity pose a major agricultural challenge, impacting around 6.73 million hectares with salinity and sodality issues. The states most affected include Gujarat, Rajasthan, Uttar Pradesh, Maharashtra, and Haryana. Gujarat has the largest area of salt-affected soil at approximately 2.22 million hectares, making it one of the hardest-hit regions. Uttar Pradesh follows with roughly 1.37 million hectares, primarily sodic soils in the Indo-Gangetic Plains. Andhra Pradesh, Rajasthan 0.38 million hectares, and Maharashtra 0.61 million hectares, West Bengal 0.44 also have significant saline-affected areas (Durairaj and Rajammal, 2023). These conditions lead to decreased crop productivity due to high soil salinity, which restricts plant growth and water absorption. To address this, the Indian government and agricultural research organizations have introduced various strategies, including salt-tolerant crop varieties and soil amendments, aimed at enhancing productivity in these areas (Qadir *et al.*, 2008).

Salinity presents a major obstacle to sustainable food production in many parts of the world. Worsening climate variability, land and water degradation, biodiversity decline, and shifting trade regulations are transforming global conditions, making food and nutritional security a critical concern for researchers and policymakers. Rising competition for fertile land and freshwater, coupled with widespread land use, is pushing agriculture into less ideal areas.

This trend underscores the need to ensure sustainable agricultural productivity while achieving profitable outcomes (Sharma and Singh, 2015).

Salinity stress is a significant abiotic factor affecting crop productivity, particularly in regions dependent on irrigation for agriculture. Elevated salt levels in the soil can significantly hinder plant growth and yield by disrupting water absorption, causing ionic imbalances, and triggering oxidative stress (Munns & Tester, 2008). Strawberry is highly sensitive to salinity, which negatively influences its physiological and biochemical processes, such as growth, photosynthesis, and fruit quality (Saied *et al.*, 2005). In arid and semi-arid areas, salinity stands out as one of the most limiting environmental factors impacting plant growth and productivity (Turhan and Eris, 2004).

The impact of salinity on plants negatively influences growth and fruit yield, mainly by hindering root development and reducing water absorption (Albert *et al.*, 2012). An increase in electrical conductivity (EC) from 1 to 2.5 dS/m can reduce strawberry yield by as much as 50% (John, 1999). Numerous studies have shown that salt stress can lead to around a 70% decline in plant productivity (Mantri *et al.*, 2012).

Mutagenesis, the process of inducing mutations, has become an essential tool for improving crop traits and boosting genetic diversity. Both physical and chemical mutagens are extensively used in plant breeding to develop new varieties with enhanced qualities. Among physical mutagens, gamma irradiation is especially effective for inducing mutations in plants. As a form of ionizing radiation, gamma rays penetrate deeply into plant tissues, causing DNA damage that results in point mutations, chromosomal alterations, and other genetic modifications. These mutations can lead to diverse phenotypic changes, such as variations in plant height, flowering time, and fruit quality (Ahloowalia and Maluszynski, 2001). Gamma irradiation in plant breeding has contributed to the creation of numerous new cultivars with better yield, improved disease resistance, and greater tolerance to abiotic stress (Micke, 1991).

One approach to addressing cultivation challenges is the development of new, superior varieties with desired traits. As an octoploid plant that reproduces vegetatively, strawberry has relatively low genetic diversity, making traditional breeding methods challenging (Nunes *et al.*, 2013). Mutation breeding has emerged as a highly effective strategy to rapidly generate new genetic diversity across various plant species without altering the overall genetic makeup (Khursheed *et al.*, 2019).

Gamma radiation has been successfully utilized to improve various plant traits, including salinity tolerance. It affects plant physiological processes by altering or damaging essential cellular components, leading to changes in plant morphology, biochemistry, physiology, and anatomy, depending on the radiation dose (Dehpour *et al.*, 2011). The ability of gamma radiation to deeply penetrate tissues can enhance plant responses to stress (Moussa, 2011). Research suggests that lower doses of gamma radiation can stimulate enzymatic activity, promote cell growth, induce beneficial mutations, and enhance stress tolerance, helping plants adapt to osmotic stress by lowering osmotic potential or closing stomata (Askari *et al.*, 2021).

Chemical mutagens offer an alternative method for inducing mutations. Ethyl methanesulfonate (EMS) is one of the most widely used chemical mutagens in plant breeding. EMS works by alkylating guanine bases in DNA, causing mispairing during replication and creating point mutations (Konzak *et al.*, 1965). It has been effectively applied across various crops to induce beneficial mutations, such as increased yield, enhanced stress tolerance, and pathogen resistance (Talebi *et al.*, 2012). In strawberry breeding, EMS mutagenesis has been utilized to improve traits such as fruit quality, size, and disease resistance (Mondal *et al.*, 2015).

The strawberry cultivars Chandler and Winter Dawn have been thoroughly investigated for their desirable traits, such as fruit quality and adaptability to various environmental conditions. However, their ability to tolerate salinity stress is still a concern. Inducing mutations through EMS and gamma irradiation treatments can potentially enhance their salinity tolerance and improve their agronomic performance in saline environments (Rahman *et al.*, 2019). Mutation breeding has been utilized in several crops to enhance resilience to abiotic stresses, including salinity, and has emerged as a promising strategy for tackling the increasing challenges posed by climate change and soil salinization (Datta *et al.*, 2018).

Limited research has been conducted to generate variability in key strawberry cultivars grown under salt stress conditions using chemical and physical mutagens. Hence, the current study was designed to induce variability in strawberries through mutation breeding with the following objectives:

Objective

1. Standardization of suitable gamma irradiation doses for mutagenesis and development of mutant line in strawberry cv. Chandler and Winter Dawn
2. Optimization of suitable EMS doses for mutagenesis and development of mutant line in strawberry cv. Chandler and Winter Dawn
3. Evaluation of mutants developed for salinity tolerance under induced salinity stress concentration

Research Gap

1. Lack of germplasm tolerant to salinity in strawberry
2. Major research has been focused on biotic stress but very less work done on abiotic stress
3. Variability through mutation for salinity tolerance was still found unexplored in strawberry

Chapter – 2

Review of Literature



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2.3 Impact of Chemical Mutagen (EMS)

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2.4 Impact of Physical mutagen against salinity stress

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2.5.1 Strawberry

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3. Induce Salinity in Strawberry

4. Induce Salinity in Fruit crops

Mutagenesis is a powerful approach widely utilized to increase variability among crops. Mutation breeding is a traditional method of plant breeding, intersects with multiple areas, including morphology, cytogenetics, biotechnology, and molecular biology.

Mutation serves as a fundamental step in the evolutionary journey of organisms. The earliest documented instance of mutation dates back to 1791, when an English farmer, Seth Wright, noticed a male lamb with unusually short legs, leading to the development of the ancon breed of sheep.

In 1901, Hugo de Vries introduced the term "mutation" to describe sudden, heritable changes while studying Evening primrose (*Oenothera lamarckiana*). However, it was later revealed that these changes resulted from recombination within a specific genetic system rather than true mutations. In his influential paper *Die Mutationstheorie*, de Vries predicted that understanding mutation principles would one day allow for the deliberate induction of mutations, enabling humans to develop new traits in plants and animals. He envisioned that this knowledge would empower the production of improved plant and animal varieties. Later, in 1962, mutations are sudden, heritable changes in an organism's genotype that result in either continuous or discontinuous variation, independent of outcrossing, somatic crossing over, or genetic drift.

2.1 Mutation applied

Genetic variability is essential for crop improvement, making its creation and management a key focus in crop breeding programs. This can be approached in two ways: by utilizing the gene pool found in native crop plants and their wild relatives, or by generating induced variability through the use of mutagenic agents (IAEA, 1972).

The use of artificial mutation to create new variability in cultivated plants and domestic animals began Gager (1908), who exposed plants to radium rays. Muller (1927) further demonstrated the potential of mutation induction through irradiation, marking a significant advancement in both fundamental and applied mutation research.

The discovery of chemical mutagens by Morgan (1914) to induce mutations in *Drosophila* (*Drosophila melanogaster*) marked a significant milestone in the field of induced mutations. Later, Auerbach (1943), Oehlkers (1943), and Rapoport (1946) identified a wide range of chemical mutagens, concluding that they are as effective as physical mutagens in inducing mutations. They also suggested that certain chemical mutagens exhibit specificity in inducing mutations. Freese (1963) and Kawai (1969) further established that the frequency of mutations

induced by different mutagens serves as a benchmark for comparing the mutagenic efficiency of various agents.

2.1.1 Mutagens

In biology, a mutagen (from the Latin term meaning "source of change") is a physical or chemical agent that modifies the genetic material, typically DNA, of an organism, leading to an increased rate of mutations in the natural environment. Mutagens are generally classified into two main categories: physical and chemical mutagens, which differ in their mechanisms of action.

2.1.2 Physical Mutagens

The use of physical mutagens for the artificial induction of mutations in crop plants was first initiated by Stadler (1928) in barley, followed by Goodspeed (1929) in tobacco using x-rays as a mutagen. Various types of irradiations with mutagenic properties are categorized as physical mutagens, including ionizing radiations (x-rays, gamma rays, Q-rays, β -rays, fast neutrons, and thermal neutrons) and non-ionizing radiations (ultraviolet rays). From 1930 to 1939, a range of both ionizing and non-ionizing radiations were employed to induce mutations in plants, animals, and microorganisms. Among ionizing radiations, gamma rays have been found to be the most effective and widely used mutagen for inducing mutations in various crop plants.

2.1.3 Gamma Irradiation

The genetic effects of radiations on DNA include changes in base pairs (such as deamination), disruption of hydrogen bonds between complementary DNA strands (Bacq and Alexander, 1961), single and double-strand breaks, cross-linking of chromosomal proteins, base loss, and chemical alterations of bases (Cassarett, 1968). Base loss or alteration leads to point mutations, while strand breaks result in chromosomal mutations. Cross-linking of DNA strands can cause cell death. Pyrimidine bases are more sensitive to radiation than purines. The indirect effects of gamma radiation are primarily related to the formation of free radicals, which are believed to be the main cause of induced mutations. However, the overall effect on an organism is influenced by internal and external factors that may act as either protective or sensitizing agents (IAEA, 1977).

2.1.4 Chemical Mutagens

Many chemicals have been found to have the ability to induce mutations. Auerbach (1941) demonstrated that chemical mutagens can mimic the effects of radiation in inducing mutations in *Drosophila*. Chemical mutagens are classified into various groups, such as alkylating agents, base analogues, antibiotics, and miscellaneous agents (Auerbach, 1976). These mutagens can be further categorized into four main types: alkylating agents, base analogues, intercalating agents, and others. Among these, alkylating agents are considered the most potent in inducing mutations (Rapoport *et al.*, 1966).

The general chemistry of alkylating agents and their interactions with nucleic acids were extensively reviewed by (Bautz and Freese, 1960) with additional confirmation from (Ross, 1962), (Lawley, 1966), and (Heslot, 1959). Alkylating agents contain one or more functional alkyl groups, which can transfer to molecules at sites with high electron density. These agents interact with DNA, causing the alkylation of phosphate diester bonds, as well as purine and pyrimidine bases. The alkyl groups are typically transferred to the 6 oxygen and 7 nitrogen positions of these bases. Methylation often results in modified nucleotides with altered base-pairing properties, leading to gene or point mutations, including transitions or transversions. Additionally, chromosomal mutations may occur due to chromosomal breaks. Ethyl methanesulfonate (EMS) has been reported as one of the most widely used chemical mutagens, being as potent as NG and NMU (Heslot, 1959), a finding later confirmed by (Swaminathan *et al.*, 1968).

2.1.5 Ethyl Methane Sulfonate (EMS)

Ethyl methane sulfonate ($\text{CH}_3\text{-SO}_2\text{-O-C}_2\text{H}_5$) is a mono-functional alkylating agent, with its mutagenic properties first identified by (Loveless, 1958) in the T2 bacteriophage. Heslot *et al.* (1959) further documented its mutagenic effects. Research by Bautz and Freese (1960) and Freese (1963) demonstrated that EMS interacts with three of the four DNA bases, excluding thymine, and primarily induces gene mutations rather than chromosomal breaks. Kreig (1963) later confirmed that EMS acts on DNA bases in the order: guanine > adenine > cytosine > thymine. EMS's methyl group reacts with the N-7 position of guanine, forming 7-ethyl-guanine, which leads to both point mutations and chromosomal breaks. Additionally, (Bautz and Freese, 1960) observed the release of ethylated purine from DNA, revealing that 7-ethyl-guanine pairs with thymine instead of cytosine, resulting in a GC-AT transition mutation. A secondary effect of N-7-ethyl-guanine is the weakening of the bond between the base and sugar, which results in the gradual loss of guanine from the alkylated DNA. The "apurine gap"

created by this loss, if filled incorrectly, can lead to transition and transversion mutations, or if left unfilled, may result in frameshift mutations (Lawley and Brooke, 1963).

2.2 Impact of Physical Mutagen (Gamma Irradiation)

2.2.1 Strawberry

Weimin *et al.* (2009) revealed that three doses of ^{60}Co -gamma radiation were given to the runners plants of five different strawberry varieties. Five strawberry cultivars that had been exposed to ^{60}Co -gamma rays within the range of 30–80 Gy had dead runners plants. The death rate ranged from 22% to 88.7% and increased as the dose of ^{60}Co -gamma rays increased; however, the rates varied depending on the type of varieties. When compared to control, all treatment cases showed abnormal leaves. With irradiation dosage and strawberry variety, the change in leaf length and width, plant height, and size differed. The most sensitive variety was ‘Akihime’, followed by ‘Hokowase’.

Saha *et al.* (2010) reported that gamma irradiation has shown potential in improving strawberry yield and fruit quality, particularly in terms of fruit size, colour, and sugar content. Low to moderate doses of gamma radiation have been found to enhance fruit characteristics by inducing favourable genetic mutations. For example, a study conducted by Su *et al.* (2018) demonstrated that strawberry plants treated with 30 Gy of gamma radiation produced larger fruits with higher sugar content compared to controls. This improvement in fruit quality is attributed to the induction of beneficial mutations in genes responsible for sugar metabolism and fruit development.

Jan *et al.* (2012) found that low doses of gamma radiation (10 Gy to 30 Gy) in strawberry enhanced growth parameters such as plant height and leaf area, while higher doses (above 50 Gy) resulted in growth inhibition. These growth alterations are largely attributed to the disruption of cellular processes like mitosis, leading to reduced cell division and elongation at higher doses.

Ashraf *et al.* (2014) reported that gamma irradiation has been shown to influence the growth and development of strawberry plants by inducing both beneficial and deleterious mutations. For instance, studies have shown that exposure to gamma radiation at moderate doses can stimulate vegetative growth and increase yield in strawberry cultivars. However, higher doses tend to reduce plant height, number of leaves, and other growth parameters due to the detrimental effects of radiation on cellular structures.

Anwar *et al.* (2015) demonstrated that gamma irradiation can significantly alter the growth parameters of strawberry plants. For instance, exposing strawberry plants to various doses of gamma radiation affects their morphological traits, including plant height, number of leaves, and leaf area.

Hegazi *et al.* (2019) provided molecular evidence using techniques such as Random Amplified Polymorphic DNA (RAPD), confirming the genetic variations induced by gamma irradiation in strawberry plants. Gamma-irradiated strawberry plants showed significant polymorphism in RAPD markers, indicating a broad spectrum of genetic variation. This variation is essential for selecting mutants with superior traits in breeding programs.

In a study by Bhattarai *et al.* (2020) the application of gamma irradiation at doses ranging from 20 to 40 Gy on the strawberry cultivar 'Camarosa' led to a noticeable reduction in plant height and chlorophyll content, while simultaneously inducing beneficial traits like early flowering and increased fruit size at optimal doses. These findings suggest that gamma radiation can effectively induce variability in morphological traits, which can be harnessed in strawberry breeding programs.

2.2.2 Other Fruit Crops

Dziadczyk *et al.* (2003) the use of molecular markers, such as RAPD and SSR, is also becoming common to screen and validate the induced mutations at the genetic level, ensuring the desired traits are preserved in future generations. Mutation breeding through gamma irradiation thus remains a promising tool for enhancing papaya crops, particularly in response to changing environmental conditions.

Lopez *et al.* (2007) gamma irradiation has been used to induce mutations in banana plantlets during different growth stages, particularly in embryogenic cell suspensions (ECS). Mutagenesis using a cobalt⁶⁰ gamma source at doses ranging from 0 to 100 Gy has shown effective results in inducing mutations. Studies have shown that exposing ECS to gamma radiation leads to somatic embryo formation, followed by plant regeneration. The optimal gamma dose for inducing beneficial mutations in bananas without causing excessive damage to the plant is often determined using the LD₅₀ value, which is the dose at which 50% of the plantlets survive. In bananas, this value typically falls around 30–40 Gy, depending on the variety and experimental conditions.

Ollitrault *et al.* (2008) citrus crops have been one of the most extensively studied fruit crops subjected to gamma irradiation. Studies have shown that doses ranging from 20 to 50 Gy can induce mutations that improve fruit quality, enhance disease resistance, and shorten the juvenile phase. For example, gamma irradiation has been used successfully to improve fruit size and sweetness in seedless mandarin cultivars, which are highly valued in the market.

Jain (2010) Banana and plantain are important staple crops in tropical regions. Gamma irradiation has played a crucial role in improving their genetic diversity, as these crops are mostly propagated vegetatively, limiting natural variation. Mutation breeding has led to the development of improved cultivars with enhanced resistance to diseases such as Fusarium wilt and black Sigatoka, as well as better post-harvest characteristics.

Vietnam *et al.* (2010) successfully used radiation-induced mutation to enhance papaya varieties, showing improvements in fruit characteristics.

Shu *et al.* (2012) found that moderate doses of gamma radiation (around 30–40 Gy) in apple scion cuttings resulted in genetic variability, with some plants showing improved resistance to apple scab and better fruit quality. In apple breeding programs, gamma irradiation has been applied to induce mutations for improved fruit size, colour, and resistance to pests and diseases.

Sales *et al.* (2013) recent advancements in molecular techniques, such as low-coverage sequencing, have allowed for the identification of mutations at the genomic level in gamma-irradiated banana plants. These studies highlight the significance of using irradiation doses between 20 Gy and 40 Gy for inducing genetic variation while preserving essential plant traits. Lower doses may lead to insufficient mutation rates, while higher doses can cause excessive damage and sterility in banana plants. The detection of these mutations is crucial for selecting desirable traits like disease resistance, enhanced fruit quality, or stress tolerance.

Figueiredo *et al.* (2014) the application of irradiation at 0.8 kGy as a preservation method does not compromise the nutritional potential or the ability to meet daily nutritional requirements for total carotenoids and vitamin C in papaya fruits, despite any induced changes in their levels.

Kanchana *et al.* (2015) Papaya is another fruit crop that has seen significant improvements due to gamma irradiation. Studies have shown that irradiation doses between 20–50 Gy can enhance fruit yield, improve quality, and increase resistance to papaya ringspot virus (PRSV), which is a major threat to papaya production worldwide. The application of gamma irradiation has led to the release of new papaya cultivars with improved agronomic traits.

Husselman *et al.* (2016) similarly, research in South Africa has demonstrated that mutation breeding can effectively improve papaya's resistance to diseases and adaptability to local conditions. In papaya (*Carica papaya*), gamma irradiation has been explored to induce mutations for improved traits, such as seedlessness and better fruit quality.

Morya *et al.* (2018) Date palm, a key fruit crop in arid regions, has been improved through gamma irradiation techniques. Radiation-induced mutations have led to varieties with superior fruit quality, enhanced drought tolerance, and better resistance to pests and diseases. The development of these improved cultivars has contributed to the sustainability of date palm cultivation in regions facing extreme environmental stress.

Reddy *et al.* (2019) reported that moderate doses (30–50 Gy) of gamma radiation in mango induced mutations that improved fruit color, size, and post-harvest life. These mutations have provided breeders with new genetic material to develop cultivars suited to export markets.

Sinha and Khurana (2020) Pomegranate, a high-value crop, has benefited from mutation breeding using gamma irradiation to enhance traits such as fruit size, sweetness, and resistance to pests. They found that irradiated pomegranate seeds exhibited a wide range of genetic variability, with some plants producing larger, sweeter fruits and others showing improved resistance to fungal diseases.

Arthur (2021) Mango cuttings were exposed to varying doses of gamma radiation 0 (control), 2.5, 5.0, 7.5, and 10.0 Gy at a dose rate of 0.323 kGy/h using a cobalt-60 source. Among these, the 2.5 Gy dose enhanced plant growth, whereas doses exceeding 2.5 Gy had an inhibitory effect on growth.

Kondo and Niimi (2021) gamma irradiation has been used in grape breeding to induce mutations that improve both fruit quality and stress tolerance. Gamma irradiation has resulted in the development of new grape varieties with enhanced berry size, sweetness, and disease resistance. Furthermore, irradiated grapevines exhibit greater tolerance to abiotic stresses, making them more resilient in different environmental conditions.

Jankowicz-Cieslak *et al.* (2022) gamma irradiation has been shown to affect various physiological traits, including plant height, leaf length, and bunch weight. For instance, irradiated banana plants exhibit shorter stature and reduced leaf size at higher doses (above 40 Gy), whereas moderate doses (20–30 Gy) can enhance desirable traits such as higher fruit yield and improved resistance to environmental stress. Field trials have demonstrated that gamma-

irradiated banana plants can exhibit significant variation in traits like pseudostem circumference, flowering date, and number of hands per bunch.

Eun *et al.* (2024) studies on Citrus unshiu, for example, have demonstrated the effectiveness of gamma irradiation in creating new mutants with altered fruit shapes and sizes, such as the Gwonje-early variety. This mutant showed significant morphological changes compared to the wild type, including larger fruit size and unique shapes, suggesting the potential for gamma irradiation to enhance the commercial viability of citrus crops by diversifying their characteristics.

2.3 Effect of Chemical Mutagen (EMS)

2.3.1 Strawberry

Abbas *et al.* (2018) investigated the impact of EMS on plant height in strawberry runners. They found that EMS treatment could induce significant variability in plant height, with optimal results observed at specific doses. For instance, a 2% EMS dose resulted in increased plant height compared to lower doses.

Adak and Kaynak (2018) examined EMS effects on bud and flower initiation. Their findings indicated that EMS treatment could accelerate bud initiation and increase the number of flowers, although higher doses sometimes resulted in delayed flowering.

Moneim *et al.* (2021) Conducted a study on EMS using three concentrations (0.0, 0.1, and 0.3%) applied for durations of 30, 60, and 90 minutes to runners tips of the strawberry variety "Fortuna cv." EMS treatment significantly enhanced vegetative growth and yield traits under salt stress conditions compared to the control, including improvements in plant height, leaf count, chlorophyll content, fruit number per plot, and fruit weight per plot. Additionally, EMS-treated plants exhibited superior fruit quality parameters compared to control plants.

Moneim *et al.* (2021) explored the influence of EMS on leaf area and fruit diameter. Their study revealed that EMS treatment could enhance leaf area and fruit size, particularly at 2% EMS, which resulted in superior fruit diameter and leaf growth.

Ali *et al.* (2022) investigated the impact of EMS on strawberry plants, focusing on growth parameters such as plant height, leaf area, and number of leaves. They found that EMS treatment led to significant variations in these traits, with a 2% EMS dose producing the most favourable results for plant height and leaf area in the variety 'Chandler'.

2.3.2 Other fruit crops

Mahadevamma *et al.* (1997) treated seeds of two commercial papaya cultivars, Coorg Honey Dew and Sunrise Solo, with ethyl methane sulphonate at concentrations of 0.1%, 0.2%, and 0.3%, along with a control, for 8 and 12 hours. Seeds treated with 0.3% EMS for 12 hours showed the lowest rates of germination and survival. The LD₅₀ dose for germination was found to be greater than 0.3% for both durations and cultivars. As the EMS concentration and treatment duration increased, there was a decline in the number of leaves per plant and seedling height, observed at both 15 and 30 days post-germination.

In comparison to the control group and other treatments, Kumar *et al.* (2016) discovered that EMS 0.50% had the strongest stimulating impact. The germination percentage, growth rate, and yield characteristics of papaya were all optimized with a modest dose of ethyl methane sulphonate (EMS). The stimulatory effect at lower doses is a result of mutagens at lower concentrations stimulating the growth- and yield-promoting enzyme and growth hormone, whereas the inhibitory effect at higher doses is a result of the faster rate of biological damage. Induced mutagenesis by ethyl methane sulphonate is one possible method for producing genetically modified plants with an increased fruit yield in papaya.

Kumar *et al.* (2017) revealed the effect of EMS on fruit quality of papaya (*Carica papaya* L.) cv Pusa Dwarf. EMS (0.50%) was most effective in improving the physical quality of papaya. Maximum fruit length (23.54 cm), fruit girth (47.32 cm), fruit weight (1514.26 gm) and pulp thickness (3.22 cm) was recorded with 0.50% EMS seed treatment. However, higher doses of EMS (1.00%) had minimum fruit length (19.99 cm), fruit girth (42.02 cm), fruit weight (1306.79 gm) and pulp thickness (2.71 cm).

Kumar *et al.* (2017) In his investigation, EMS at 0.50% demonstrated superior efficacy and had a stimulatory impact relative to the control and other treatments. The stimulatory effect of EMS at lower doses arises from the ability of mutagens at reduced concentrations to enhance the activity of enzymes and growth hormones associated with growth, yield, and fruit quality, whereas greater concentrations of mutagens exert an inhibitory influence on fruit quality.

Abd El-Latif *et al.* (2018) EMS inhibited cell division, unlike the addition of Colchicine. This effect may be attributed to the disruption of DNA replication during mitosis, resulting from increased damage caused by BA and EMS.

Kumar *et al.* (2021) demonstrated that chemical mutagens significantly impact the physiology of Kinnow plants. The reduction in chlorophyll concentrations confirmed the detrimental effects of EMS on plant pigment levels. The higher levels of total proteins and phenolic compounds in all mutants indicate their involvement in the plant's defense system. Mutagenesis also triggered the activation of the enzymatic antioxidant defense system, enhancing its ability to scavenge reactive oxygen species (ROS). Therefore, antioxidant enzyme activities can serve as indicators of plant sensitivity to EMS and the development of new genotypes.

Kumar *et al.* (2021) found that the chemical mutagen EMS generated significant variability in the Kinnow population. Long-term evaluation and characterization of mutants led to the identification of mutant E-16, which exhibited dual traits of dwarfism and fruits with fewer seeds, and mutant E-19, which had a reduced number of seeds per fruit. These mutants can be utilized in future breeding programs. Additionally, EMS can be applied to other citrus species to generate variability and develop mutants with desirable traits.

Rawat *et al.* (2023) conducted an investigation on pomegranate cv. Bhagwa, which demonstrated that EMS treatment affected leaf physiology. Lower EMS concentrations had a minimal impact on leaf traits, while higher mutagenic doses resulted in more significant changes in cutting survival, leaf area, and chlorophyll content. This suggests that at higher mutagenic doses, the degradation of cell organelles occurs, disrupting cellular functions due to the accumulation of free radicals in plant tissues. Moreover, the broad range of variations induced by EMS has expanded the potential for a detailed examination of the physiological traits of desirable mutants obtained during the vegetative phase. Therefore, it can be concluded that EMS can generate mutants in pomegranate, which could be used as commercial cultivars after undergoing appropriate agronomic and quality assessments.

2.4 Impact of Physical mutagen against salinity stress

2.4.1 Strawberry

Alhamza *et al.* (2016) used gamma radiation doses of 20, 50, and 100 Gy to induce variations *in vitro*. The results showed that a 20 Gy dose was most effective for improving strawberry traits, resulting in the highest number of shoots, as well as increased fresh and dry weight, and enhanced activity of peroxidase and catalase. Higher salinity levels also led to increased proline and peroxidase activity.

Kepenek (2016) developing somaclonal variants offers a potential solution for generating salt-tolerant genotypes. In a study focusing on the Arnavutköy strawberry cultivar, axillary shoots were irradiated with gamma rays (20 and 40 Gy from ^{60}Co) and subjected to different NaCl concentrations (0, 250, 500, and 750 ppm) to evaluate their effects on salinity tolerance. The combination of increased gamma irradiation doses and NaCl concentrations had a notable impact on survival rates, axillary shoot proliferation, shoot length, fresh weight, leaf deformation, and necrotic areas. Among the treatments, a gamma irradiation dose of 40 Gy combined with 500 ppm NaCl was found to be the most effective, producing somaclonal variants with minimal lethal effects and leaf deformities. This methodology, if applied successfully, could facilitate the rapid identification of new NaCl-resistant strawberry cultivars.

Pimonrat and Yi (2017) exposed *in vitro* banana shoots to gamma radiation at doses of 0, 10, 20, 30, 40, and 50 Gy, and regenerated them on media with NaCl concentrations of 0, 6, 8, and 10 g/L. The results demonstrated that gamma irradiation reduced the severity of salinity, with irradiated shoots showing a higher survival rate on 8 g/L NaCl compared to non-irradiated shoots. This suggests that gamma irradiation could be a viable method to enhance salt tolerance in commercial banana varieties.

Geng *et al.* (2019) the results indicated that pretreating seeds with varying doses of gamma radiation improved the tolerance of sweet osmanthus seedlings to salinity stress, and the beneficial effect of gamma radiation became more significant as the radiation dose increased (50–150 Gy).

2.4.2 Other fruit crops

Ashraf (2009) in the context of bananas, *in vitro* propagation combined with gamma irradiation has proven effective in improving salt tolerance banana shoots exposed to gamma rays (doses of 10-50 Gy) and cultured in saline conditions showed enhanced survival rates compared to non-irradiated shoots. This suggests that gamma irradiation induces physiological and genetic modifications that allow the plant to better manage saline stress.

Miri *et al.* (2019) explored the use of gamma irradiation to improve salinity tolerance in banana (Musa AAA cv. ‘Dwarf Cavendish’). Shoot tips were exposed to gamma radiation at doses of 25, 35, and 45 Gy, followed by two cycles of salinity stress using different NaCl concentrations (0, 100, 120, 140, and 160 mM). The results showed that higher NaCl concentrations reduced the growth rate during the first salinity stress cycle, but gamma-irradiated explants exhibited

better survival rates, more shoots, and higher fresh weights than non-irradiated ones. During the second salinity stress cycle, only the irradiated explants exposed to 160 mM NaCl experienced a decline in shoot number. When *in vitro*-regenerated plants were transferred to greenhouse conditions (third salinity stress), irradiated plants demonstrated a higher survival rate and leaf count than non-irradiated plants. Furthermore, non-irradiated plants exhibited more severe leaf burn and yellowing as salt concentrations increased. Overall, gamma-irradiated shoot tips displayed improved growth under both *in vitro* and greenhouse salinity stress, suggesting that gamma irradiation can enhance salinity tolerance in bananas. Further research is needed to evaluate these mutants in field conditions under saline stress.

Rajan *et al.* (2021) Investigated the effect of gamma irradiation at various doses (0 Gy, 10 Gy, 20 Gy, 30 Gy, and 40 Gy) on the salinity tolerance of the Grand Naine variety of banana, under induced NaCl stress at concentrations of 50 mM, 100 mM, and 150 mM. The study found that the gamma irradiation doses of 10 Gy and 20 Gy were the most effective in generating salinity-tolerant mutant lines.

2.5 Impact of Chemical mutagen against salinity stress

2.5.1 Strawberry

Saied *et al.* (2005) moreover, EMS treatment reduced the impact of salinity stress on fruit yield, with EMS-treated plants showing a smaller reduction in yield compared to untreated plants. Untreated 'Korona' saw a 27% yield reduction, while 'Elsanta' had a 64% reduction, but EMS-treated plants exhibited less severe declines. Fruit quality also improved in EMS-treated plants, as indicated by consumer-type panels that assessed taste, aroma, and texture. In contrast to the 24% reduction in quality in untreated 'Elsanta', EMS-treated plants retained higher quality, with fewer reductions in total soluble solids (Brix) and the Brix/TA ratio. EMS-treated 'Korona' and 'Elsanta' plants experienced smaller decreases in Brix (20% in 'Korona' and 35% in 'Elsanta'), indicating that EMS played a significant role in enhancing strawberry resilience under salinity stress.

Munns and Tester. (2008) salinity is a major environmental stress that adversely affects plant growth, development, and productivity. Strawberry, a highly valued crop, is sensitive to salinity, which impairs its yield by disrupting water uptake, nutrient balance, and physiological processes. The severity of salinity stress is determined by factors such as the concentration of salts, the duration of exposure, and the plant's inherent tolerance. Salinity stress typically

induces osmotic stress, ion toxicity, and oxidative damage, leading to reduced photosynthesis, growth inhibition, and increased leaf scorching in plants.

Abbas *et al.* (2018) plants regenerated from EMS-treated leaf explants demonstrated enhanced tolerance to NaCl stress compared to *in vivo* plants. These *in vitro* regenerated plants exhibited better growth and performance in terms of the assessed morphological parameters under high NaCl concentrations. Genetic analysis using RAPD-PCR revealed greater genetic variation in EMS-treated plants, with 15 unique bands detected, while 3 unique bands were identified in response to NaCl stress. The study concludes that EMS mutagenesis is a promising approach to enhance NaCl tolerance in strawberry plants, providing an effective strategy for improving salt stress resilience in sensitive cultivars.

Al-Salihy *et al.* (2018) Under varying NaCl concentrations (0.0, 10, 25, 35, and 45 mM). The results demonstrated that increasing levels of NaCl significantly reduced shoot regeneration and rooting, as well as decreased chlorophyll content plants regenerated *in vitro* from EMS-treated explants exhibited greater resilience compared to *in vivo* plants, showing improved chlorophyll retention and better rooting at higher NaCl concentrations. This indicates that EMS mutagenesis mitigated the negative effects of salinity stress. Genetic variation analysis using RAPD-PCR revealed seven unique bands in EMS-treated plants, suggesting successful induction of genetic diversity. These findings highlight the potential of EMS mutagenesis to enhance salt tolerance in strawberries.

Bates *et al.* (2021) salinity stress adversely affects chlorophyll content and photosynthetic efficiency in strawberry plants, leading to reduced growth and yield. Studies have demonstrated that EMS can mitigate the negative effects of salinity on chlorophyll content by inducing mutations that enhance chlorophyll stability and photosynthetic activity under salt stress. For example, strawberry plants treated with 2% EMS under salinity stress showed higher chlorophyll content compared to untreated controls, indicating improved photosynthetic performance.

Moneim *et al.* (2021) evaluated strawberry plants under saline stress conditions with NaCl concentrations of 500 and 1000 ppm. The results showed that at 1000 ppm NaCl, the plants had the lowest number of leaves, plant height, and root length per plant (5.00 leaves, 9.50 cm, and 4.67 cm, respectively), while at 500 ppm NaCl, the plants showed better growth, with 11.56 leaves, 12.28 cm plant height, and 9.28 cm root length per plant.

2.5.2 Other fruit crops

Deng *et al.* (1989), in their study on selecting salt-tolerant mutants by treating calli of citrus rootstock and scion cultivars, showed that the lethal concentration of EMS plays a crucial role in mutagenesis effects, and it is determined by both the concentration and the duration of treatment.

Munns *et al.* (2009) comparison of EMS with Other Stress Alleviation Methods While EMS-induced mutagenesis is effective in improving salinity tolerance, other methods such as genetic engineering, conventional breeding, and exogenous application of osmoprotectants like glycine betaine and proline have also been employed to mitigate salinity stress. However, EMS is considered more advantageous due to its ability to induce a wide range of mutations without the regulatory complexities of genetically modified organisms (GMOs). EMS-induced mutants also have the potential to exhibit long-term heritable salinity tolerance.

2.6 Induce Salinity in Strawberry

Husaini and Abdin (2008) indicated that strawberry plants genetically modified to excessively generate osmotin may withstand salt levels nearing an EC of 20 dS m⁻¹. Prior salinity tolerance research performed in greenhouse settings using *Fragaria ananassa* cultivars Douglas and Toro indicated that salt and sulfate did not induce ion toxicity in strawberries. Chloride-based fluids (NaCl and KCl) caused leaf blistering after 26 days, or six irrigations, when the water had a concentration of 18 mmol L⁻¹ (corresponding to a salinity of EC_{iw} = 1.8 dS m⁻¹) that leaf Cl⁻ concentrations below 1% and irrigation water electrical conductivity (EC_{iw}) levels under 2.0 dS m⁻¹ did not induce toxicity symptoms in the examined cultivars. Salinities over 5 dS m⁻¹, despite modest leaf Cl⁻ concentrations, may result in leaf damage.

Sun *et al.* (2015) found that strawberry cultivar "Chandler" exhibited a significant reduction in shoot dry weight (DW) as the salinity of the irrigation water increased, with salt solutions having electrical conductivities (ECs) of 2.2, 3.3, or 4.4 dS·m⁻¹. At an EC of 4.4 dS·m⁻¹, the salt treatment did not significantly reduce the yield of "Chandler," but plants at this EC had visual quality scores lower than 3.0.

Abbas *et al.* (2018) conducted a study where pots were irrigated with distilled water supplemented with different concentrations of NaCl (0, 10, 25, 35, or 45 mM). The results showed that the NaCl-treated plants exhibited better tolerance compared to the untreated plants, as evidenced by improved morphological parameters such as the number of leaves, plant

height, vegetative area, and fresh and dry weight of shoots and roots, in strawberry cultivars when compared to the untreated *in vivo* plants.

In their irrigation nutrient solution, Avestan *et al.* (2019) dissolved NaCl to 0, 25, and 50 mM concentrations to stress strawberry plants. The plants had salt stress two weeks after planting. Salt concentrations were gradually increased over the first two weeks, followed by saline solution every four days to reduce salt stress shock. The containers also got 600 mL of water biweekly to drain excess salt during salinity treatment. The 50 mM NaCl treatment decreased growth metrics more than the 25 mM.

Faghih *et al.* (2019) studied strawberry (*Fragaria × ananassa* cv. ‘Camarosa’) plants planted in plastic pots with a 2.5 kg mixture of soil, peat, and perlite 1:1:1 (v/v/v). This soil mixture had a silt-loam texture, pH of 7.3, and electrical conductivity of 2.28 dS m⁻¹. The pots were in a greenhouse and irrigated every three days with 0, 30, and 60 mM NaCl salty water. Each irrigation let 1/3 of the water drain through the pots to prevent media salt buildup. After two irrigation cycles, the media was rinsed with distilled water before the third. After 14 days of salt stress, plant damage symptoms appeared.

Singh *et al.* (2020) investigated the effects of different salt concentrations (NaCl: Na₂SO₄: CaCl₂ in a 7:1:2 ratio) on three strawberry cultivars: Capri, Winter Star, and Camarosa. The salt solution was prepared by mixing NaCl, Na₂SO₄, and CaCl₂ in the specified ratio, with three treatment levels: (0.0 EC - Control), (1.5 dS/m), and (3.0 dS/m). The results indicated that the Camarosa cultivar exhibited the highest salt tolerance, showing better growth in terms of number of leaves, shoots, leaf length, and chlorophyll content, suggesting its resilience to salinity. In contrast, the Capri cultivar displayed the lowest salt tolerance, performing poorly in these growth parameters and being more susceptible to salinity.

Abd El-Moneim *et al.* (2021) conducted a study on strawberry plants evaluated under saline stress conditions with NaCl concentrations of 500 and 1000 ppm. The results showed that at 1000 ppm NaCl, the plants had the lowest number of leaves, plant height, and root length per plant (5.00 leaves, 9.50 cm, and 4.67 cm, respectively). In contrast, at 500 ppm NaCl, the plants exhibited better growth, with 11.56 leaves, 12.28 cm plant height, and 9.28 cm root length per plant.

2.7 Induce Salinity in Fruit crops

Morsy (2003) examined the growth performance of mango cultivars irrigated under various salinity conditions. The study found that salinity significantly hindered plant growth in all mango cultivars, particularly at the highest concentration (45 mM). Water salinity led to a considerable reduction in stem thickness, number of leaves per plant, and average leaf area. Root and shoot dry weight decreased as salinity increased in all cultivars, while the root: shoot ratio remained unaffected. Negative effects of salinity on the growth of all the mango cultivars were observed at a concentration of 30 mM or higher.

Atmane *et al.* (2005) investigated the impact of salinity (NaCl at 0, 35, or 70 mM for 30 or 60 days) on the growth of citrus rootstocks. They concluded that the reduction in growth of the rootstocks under saline conditions could be due to an imbalance of essential nutrients.

Pandey *et al.* (2014) studied seven mango (*Mangifera indica* L.) rootstocks Moovandan, Bappakai, Nekkare, Kurukkan, Olour, Terpentine, and Chandrakaran which were irrigated with water containing 0, 50, 100, and 150 mM NaCl to 70% of field capacity for 90 days. They suggested that lower levels of Cl⁻ and Na⁺ accumulation could serve as useful indicators for screening mango rootstocks for resistance to NaCl stress. The rootstocks Olour, Terpentine, and Nekkare were able to exclude Cl⁻ ions, while Kurukkan, Bappakai, and Moovandan appeared to exclude Na⁺ ions at lower salt concentrations. Based on their overall performance and leaf scorching, the study concluded that salinity tolerance increased in certain rootstocks.

Chapter – 3

Materials and Methods



The present study entitled “**Deciphering the effectiveness of induced mutation for salinity tolerance on Strawberry (*Fragaria x ananassa* Duch.) cv. Chandler and Winter Dawn**” were conducted during the two seasons 2022-2023 and 2023-2024. Evaluating the salinity tolerance of Chandler and Winter Dawn strawberry cultivars by applying physical and chemical mutagens and determining optimal mutagen doses for enhanced salinity resilience.

- 3.1 Experimental site and location**
- 3.2 Collection of planting material**
- 3.3 Experiment details**
- 3.4 Selection of variety for experiment**
- 3.5 Cultivar**
- 3.6 Material required**
- 3.7 Physical mutagen treatment**
 - 3.7.1 Gamma irradiation Cobalt-60 (^{60}Co)**
- 3.8 Chemical mutagen treatments**
 - 3.8.1 Ethyl methanesulfonate**
- 3.9 Creation of salinity level**
- 3.10 Observation to be recorded**

3.1 Experimental Site and Location

This study was carried out at the Agriculture Farm of Lovely Professional University, Phagwara Punjab, situated at a latitude of 30°56'11.90" N and a longitude of 76°18'13.18" E, with an average altitude of 279 meters above sea level.

3.2 Collection of planting material

First-generation planting material of the Winter Dawn and Chandler strawberry variety was obtained from the IARI Regional Station located in Shimla, Himachal Pradesh. This material was gathered for the initial trial in mid-October 2022 and for a second trial in October 2023.

3.3. Table 3.1. Experiment Details

Treatments and Replications	Details
Total Number of Treatments (Factor 1)	4 (F1: Gamma Irradiation/EMS)
Total Number of Treatments (Factor 2)	4 (F2: Salinity Doses)
Total Number of Replications (Factor 1)	4
Total Number of Replications (Factor 2)	4
Total Number of Plants per Replication	3
Total Number of Treated Plants of (V1)	150 (V1: Winter Dawn)
Total Number of Treated Plants of (V2)	150 (V2: Chandler)
Total Number of Control Plants	(V1 and V2): 100
Experiment Design	FCRD (Factorial Completely Randomized Design) and CRD

3.4 Selection of cultivar for experiment

The Winter Dawn and Chandler strawberry varieties were selected for the salinity experiment due to their commercial importance, growth adaptability and distinct responses to environmental stress.

3.5 Cultivar

3.5.1. Winter Dawn

(Cultivar 1)

The Winter Dawn strawberry variety was developed by the University of Florida is known for its early fruiting, making it ideal for winter production. It produces large, firm, and glossy red berries with excellent flavour and high sugar content. The variety is heat-tolerant and performs well in warmer climates, displaying good resistance to common diseases. It has a vigorous plant structure with high yield potential, making it popular among growers for both commercial and fresh market production Bhamini *et al.*, 2017.

3.5.2. Chandler

(Cultivar 2)

The Chandler strawberry variety is often preferred for its overall productivity and superior quality, as it shows greater salt tolerance, though only up to a certain level (Sun *et al.*, 2015). Developed by the University of California and introduced in 1983, Chandler strawberries are a hybrid, resulting from a cross between Douglas strawberries and an unnamed cultivar from the University of California.

3.6 Material required

3.6.1 Pot preparation

The experiment set up was taken up in a 25 cm plastic pots by adding soil, vermicompost and cocopeat in 1:1:1 ratio. 400 pots were filled up five days prior to transplanting. During transplanting, the plants were trimmed to eliminate runners, dead leaves, and flowers before potting, the soil was loosened with a rotavator, and weeds and stubble were thoroughly removed.

3.7 Physical mutagens treatment

The gamma radiation treatment was given for strawberry runners at Punjab Agricultural University, Ludhiana, Punjab

3.7.1 Gamma irradiation cobalt-60 (^{60}Co)

Factor 1

Table 3.2 Dose determination of physical mutagen gamma irradiation

Treatments	Dose Rate	Time of radioactive decay.
T ₁	20 Gy	10 min 14 sec
T ₂	30 Gy	15 min 22 sec
T ₃	40 Gy	20 min 29 sec
T ₄	Control	

3.7.2 Application of the treatments

Strawberry runners were treated with ^{60}Co -gamma rays at a dosage rate of 1.953 Gy/min. Each treatment was replicated five times, with three plants taken per replication. After that, they were transplanted immediately.

Layout of different dosages of Gamma irradiation for V1.

T₁ V₁	R₁ 	R₂ 	R₃ 	R₄ 
T₂ V₁	R₁ 	R₂ 	R₃ 	R₄ 
T₃ V₁	R₁ 	R₂ 	R₃ 	R₄ 

Layout of different dosages of Gamma irradiation for V2.

T₁ V₂	R₁ 	R₂ 	R₃ 	R₄ 
T₂ V₂	R₁ 	R₂ 	R₃ 	R₄ 
T₃ V₂	R₁ 	R₂ 	R₃ 	R₄ 
T₄ V₂	R₁ 	R₂ 	R₃ 	R₄ 

3.8 Chemical mutagens EMS treatments

The ethyl methane sulfonate treatment was given for strawberry runners at Lovely Professional University, Phagwara, Punjab.

3.8.1 Ethyl methane sulfonate

Table 3.3 Dose determination of chemical mutagen EMS

Treatments	Dose Rate
T ₅	1 %
T ₆	2 %
T ₇	3 %
T ₈	Control



Plate 1. Visual of Physical Mutagen Gamma Irradiation dose chamber

3.8.2 Material required

3.8.2.1 Preparation of EMS solution

To prepare EMS solutions of 1%, 2%, and 3%, 20 ml of DMSO was used as a carrier agent for each treatment. The 1% EMS solution was prepared by mixing 20 ml of DMSO, 979 ml of distilled water, and 1 ml of EMS. Similarly, the 2% EMS solution was prepared by adding 20 ml of DMSO, 978 ml of distilled water, and 2 ml of EMS, while the 3% EMS solution was prepared using 20 ml of DMSO, 977 ml of distilled water, and 3 ml of EMS.

3.8.2.2 Application of the EMS treatments

Strawberry runners will be soaked in three EMS doses 1%, 2%, and 3% for 1 hour, after which they will be immediately transplanted. Each treatment will be replicated three times, with three plants used per replication for each treatment.

Layout of different dosages of EMS for V1.

T4 V₁	R₁ 	R₂ 	R₃ 	R₄ 
T5 V₁	R₁ 	R₂ 	R₃ 	R₄ 
T6 V₁	R₁ 	R₂ 	R₃ 	R₄ 
T7 V₁	R₁ 	R₂ 	R₃ 	R₄ 
T8 V₁	R₁ 	R₂ 	R₃ 	R₄ 

Layout of different dosages of EMS for V2.

T4 V ₁	R ₁ 	R ₂ 	R ₃ 	R ₄ 
T ₅ V ₁	R ₁ 	R ₂ 	R ₃ 	R ₄ 
T ₆ V ₁	R ₁ 	R ₂ 	R ₃ 	R ₄ 
T ₇ V ₁	R ₁ 	R ₂ 	R ₃ 	R ₄ 



Plate 2. Visual of Chemical mutagen dose treatment (EMS) for strawberry runners

3.9 Creation of salinity level

3.9.1 Treatment details

Factor 2

The experiment employed a factorial completely randomized design, where physical and chemical mutagens served as the primary factor, and salt stress treatments were considered the secondary factor, with five replications for each treatment. Each replication included three plants per line.

3.9.2 Imposition of salt stress

Stress treatments were administered through the addition of NaCl. The plants received specific NaCl concentrations (0 mM, 50 mM, 75 mM, and 100 mM) after determining the electrical conductivity (EC) of the potting mixture. To evaluate the impact of salt stress, a control group of plants was kept next to each treatment, with these control plants receiving water without any added NaCl (Table 3).

Table 3.4 Salinity doses for treatments of V1 and V2

Treatments	Salinity levels
S ₁	0 mM NaCl (Control)
S ₂	50 mM NaCl
S ₃	75 mM NaCl
S ₄	100 mM NaCl

Similar treatments will be imposed in V2 as well chemical mutagen treated runners for both V1 and V2.

Table 3.5 Treatment combination for (V1)

Treatment	Treatment	Treatment	Treatment
T ₁ S ₁	T ₁ S ₂	T ₁ S ₃	T ₁ S ₄
T ₂ S ₁	T ₂ S ₂	T ₂ S ₃	T ₂ S ₄
T ₃ S ₁	T ₃ S ₂	T ₃ S ₃	T ₃ S ₄
T ₄ S ₁	T ₀ S ₂	T ₀ S ₃	T ₀ S ₄
T ₅ S ₁	T ₄ S ₂	T ₄ S ₃	T ₄ S ₄
T ₆ S ₁	T ₅ S ₂	T ₅ S ₃	T ₅ S ₄

T ₇ S ₁	T ₆ S ₂	T ₆ S ₃	T ₆ S ₄
T ₈ S ₁	T ₀ S ₂	T ₀ S ₃	T ₀ S ₄

Table 3.6 Treatment combination for (V2)

Treatment	Treatment	Treatment	Treatment
T ₁ S ₁	T ₁ S ₂	T ₁ S ₃	T ₁ S ₄
T ₂ S ₁	T ₂ S ₂	T ₂ S ₃	T ₂ S ₄
T ₃ S ₁	T ₃ S ₂	T ₃ S ₃	T ₃ S ₄
T ₄ S ₁	T ₀ S ₂	T ₀ S ₃	T ₀ S ₄
T ₅ S ₁	T ₄ S ₂	T ₄ S ₃	T ₄ S ₄
T ₆ S ₁	T ₅ S ₂	T ₅ S ₃	T ₅ S ₄
T ₇ S ₁	T ₆ S ₂	T ₆ S ₃	T ₆ S ₄
T ₈ S ₁	T ₀ S ₂	T ₀ S ₃	T ₀ S ₄



Plate.3 Visual of potting mixture before planting

4.0 Observation recorded

LD₅₀

The LD₅₀ doses of irradiation were determined using the probit analysis method, based on the percentage of mortality observed 15 days after treatment. This calculation also incorporated data on sprouting percentage and survival rate, as outlined by Sharma (1998).

Per Cent Survival (Days after irradiation)

The number of shoots exhibiting the growth with non-browning shoots will be taken at 15, 20 and 30 days of treatment. It will be calculated using formula

$$\text{Per Cent Survival} = \frac{\text{Number of explants survived} \times 100}{\text{Number of runners treated}}$$

Morphological Parameters

Plant height (cm)

The height of each plant in each pot was measured in centimeters using a meter scale, and the average height was subsequently determined.

Leaf area (mm)

Twenty fully expanded mature leaves were randomly selected from three plants in each replication of every treatment. The leaf area was measured using a leaf area meter (Licor-Model 3100) and the average leaf area for each treatment was calculated and recorded in square centimetres.

Shoot and root fresh weights (gm)

At the end of the experiment, the random plants treated and control were gently uprooted and washed with tap water to remove any residual foreign particles. Afterward, the fresh weight of the shoots (SFW) and the fresh weight of the roots (RFW) were measured.

Shoot dry weight (g plant⁻¹)

After recording the fresh shoot weight of the same plant, the dry weight was determined by placing the cleaned samples in an electric oven. Initially, the temperature was set to 110 °C for one hour to halt metabolic activities, followed by a constant temperature of 70 °C for 72 hours. Regular measurements were taken using a digital electronic balance until a constant dry weight

was achieved. The final dry weight was then divided by two to calculate the shoot dry weight per plant.

Root dry weight (g plant⁻¹)

The roots of two plants were thoroughly washed, and their dry weight was recorded after being placed in an electric oven. Initially, the temperature was set to 110 °C for one hour to stop metabolic activities, followed by a consistent temperature of 70 °C for 72 hours. Regular measurements were taken on a digital electronic balance until a stable dry weight was achieved. The resulting dry weight was then divided by two to calculate the respective root dry weight per plant at 15 and 30 days after sowing.

Fruit length and diameter measurement (mm)

Fruit length and diameter were measured in millimeters (mm) using a Digital Caliper -515 (DC-515). The average was calculated for each treatment.

Fruit weight (gm)

Fruit weight was measured in grams using an electric balance. The total fruit weight for each pot was determined by summing the weights of all the fruits, and the average fruit weight was calculated by dividing the total fruit weight by the total number of fruits.

Yield per plant (gm)

The total weight of all fruits harvested from each plant will be recorded for each cultivar, and the results will be expressed as the yield per plant.

Chlorophyll content index

A portable chlorophyll meter (SPAD-502, Konica Minolta Sensing, Inc., Japan) will be used to evaluate the leaf greenness of the plants. The SPAD-502 meter provides an accurate, non-destructive estimate of the total chlorophyll content in the leaves of different species (Neufeld *et al.*, 2006). For each plant, measurements will be taken at four points on each fully expanded leaf, two on each side of the midrib, and the results will be averaged (Khan *et al.*, 2003).

Developmental parameters

Number of leaves, flower buds, flowers and fruits per plant

The number of leaves, flower buds, flowers, and fruits per plant was determined by counting all the leaves, flower buds, flowers, and fruits on each plant in every pot, and then calculating the average.

Days to flower bud initiation, flowering, fruit setting and harvesting

The number of days until inflorescence initiation (based on visual observation), flowering, fruiting, and harvesting was recorded from the date of transplanting the strawberry plants.

Salinity stress-specific parameters

Mineral analysis (sodium and potassium)

Digestion of leaf samples:

Two common methods used to completely eliminate the organic compounds in plants are:

A. Dry ashing

B. Wet digestion

A) Dry ashing: It refers to ignition of plant sample and the procedure of ashing varies with temperature. Temperature of 500°C and 2-4 hours time is sufficient to digest the plant samples completely. Boron can be determined by dry ashing only since boron is volatilized during wet digestion.

B) Wet digestion: Wet digestion is a process in which organic matter is broken down through oxidation in a liquid medium. This oxidation is performed using a mixture of three acids HNO_3 , HClO_4 , and H_2SO_4 or a dual acid mixture of HNO_3 and HClO_4 in a 4:1 ratio.

Reagents

Triple acid mixture: Mix concentrated; HNO_3 , HClO_4 and H_2SO_4 of analytical grade in the ratio of 9:3:1.

Procedure

- A. Weigh 1 g of the plant sample and place it into a 250 ml Erlenmeyer flask.
- B. Add 15-20 ml of the triple acid mixture and swirl the flask to mix the contents.
- C. Position a funnel at the neck of the flask and leave it overnight.

- D. Next, heat the flask gently on a hot plate. A vigorous reaction will start right away, emitting brown fumes of nitrous oxide. After the HNO_3 evaporates, white fumes of HClO_4 will form, and the reaction will finish within 1-2 minutes.
- E. Then, raise the temperature to its highest point (around 400°C) to allow the H_2SO_4 to reflux at the bottom of the flask's neck.
- F. Maintain this temperature until the contents of the flask take on a yellowish-green color, leaving about 1-2 ml of digest behind.
- G. Let the mixture cool down, then dilute it with about 25 ml of distilled water.
- H. Pour the solution into a 100 ml volumetric flask, ensuring to rinse the Erlenmeyer flask to include all the contents.
- I. Filter the solution using Whatman No. 1 filter paper, and use the resulting filtrate to assess phosphorus and potassium concentrations.

Leaf burning %

Leaf burn observations will be conducted visually.

Physiological parameters

Malondialdehyde content

It is measured to evaluate membrane lipid peroxidation in plants and is commonly used as an indicator of oxidative lipid damage caused by environmental stress. Lipid peroxidation levels were determined by assessing the malondialdehyde content, following the method developed by Heath and Packer in 1968.

Reagents

A 0.1% (w/v) solution of trichloroacetic acid (TCA) was prepared by dissolving 0.1 g of TCA in water and adjusting the total volume to 100 mL. To prepare the thiobarbituric acid (TBA) reagent, a 0.5% solution was made by first preparing a 20% (w/v) TCA solution (dissolving 20 g of TCA in 100 mL of distilled water). Then, 0.5 g of TBA was dissolved in a small amount of the 20% TCA solution, and the final volume was brought up to 100 mL by adding more of the 20% TCA solution.

Procedure

A 100 mg sample of plant material was homogenized in 0.5 mL of a 0.1% (w/v) trichloroacetic acid solution. The homogenate was then centrifuged at 15,000 x g for 10 minutes at 40 °C, and 0.5 mL of the supernatant was mixed with 1.5 mL of 0.5% (w/v) thiobarbituric acid in 20% TCA. The mixture was heated at 95°C for 20 minutes and rapidly cooled in an ice bath. After cooling, the samples were centrifuged again at 15,000 x g for 5 minutes. Absorbance was measured at 532 nm, with any non-specific absorption at 600 nm subtracted. The concentration of the malondialdehyde-TBA complex (the red pigment) was calculated using an extinction coefficient of 155 mM⁻¹ cm⁻¹. This method is commonly used to evaluate oxidative stress levels in plants exposed to various stress conditions, including salinity.

Leaf relative water content

To determine the Leaf Relative Water Content (RWC) in strawberry leaves, start by collecting fully expanded, healthy leaves in the early morning to minimize water loss from transpiration. Immediately weigh the fresh weight (FW) of the leaves after collection. Then, immerse the leaves in distilled water and let them soak for 4 hours at room temperature to achieve full turgidity. After soaking, gently blot the leaves to remove any excess water and weigh them again to obtain the turgid weight (TW). Finally, dry the leaves in an oven at 70°C for 24 hours and weigh them to find the dry weight (DW). Calculate the RWC using the formula: This method is commonly used in plant physiology research to evaluate the water status of plants under various environmental conditions (Barrs & Weatherley, 1962).

$$\text{LRWC (\%)} = \frac{(\text{FW} - \text{DW})}{(\text{TW} - \text{DW})} \times 100$$



Figure 4: Determination of leaf relative water content

Electrolyte leakage

To assess electrolyte leakage (EL) from strawberry leaves, begin by gathering fresh, fully expanded leaves and rinsing them with distilled water to remove any surface impurities. Cut the leaves into uniform discs of 1 cm in diameter and place them in a test tube containing 10 mL of deionized water. Incubate the samples at room temperature (25°C) for 24 hours, ensuring that the discs are fully submerged. After the incubation, measure the initial electrical conductivity (EC1) using a conductivity meter. Following this, autoclave the samples at 121°C for 20 minutes to release all electrolytes from the leaf tissues. Allow the samples to cool to room temperature before measuring the final electrical conductivity (EC2). Calculate the percentage of electrolyte leakage using the formula. This technique is indicative of membrane integrity, with higher EL values reflecting greater membrane damage due to stress (Bajji *et al.*, 2002).

$$EL (\%) = \frac{(EC1)}{(EC2)} \times 100$$



Figure 5: Determination of electrolyte leakage

Proline content (mg g⁻¹)

Samples for assessing total proline content were collected on the 15th and 30th days after sowing, with three replications analysed for each treatment.

Estimation of proline content:

The free proline content in the leaves was determined using the method outlined by Bates *et al.* (2021).

Reagents used for Proline estimation:

A. Orthophosphoric acid

B. Acid Ninhydrin: This reagent was prepared by dissolving 1.25 g of ninhydrin in 30 mL of glacial acetic acid and 20 mL of 6.0 M orthophosphoric acid, with continuous stirring, by gently warming until completely dissolved. The solution was then stored at 4°C and used within 24 hours.

C. Aqueous Sulfosalicylic Acid (3%): This was prepared by dissolving 3.0 g of sulfosalicylic acid in 100 mL of distilled water.

D. Glacial Acetic Acid (99.7%)

E. Toluene (99.5%)

Procedure

A 200 mg leaf sample was weighed and homogenized with 5 mL of 3% sulfosalicylic acid using a mortar and pestle. The resulting homogenate was filtered through Whatman No. 1 filter paper and centrifuged at 6000 rpm for 10 minutes to collect the supernatant. The residue was then re-homogenized with 2 mL of sulfosalicylic acid and centrifuged again at the same speed for another 10 minutes. This process was repeated 4-5 times, pooling all the supernatants and adjusting the final volume to 10 mL with 3% sulfosalicylic acid. Next, 2.0 mL of the extract was transferred to a test tube along with 2 mL of glacial acetic acid and 2 mL of ninhydrin reagent. The reaction mixture was heated in a water bath at 100°C

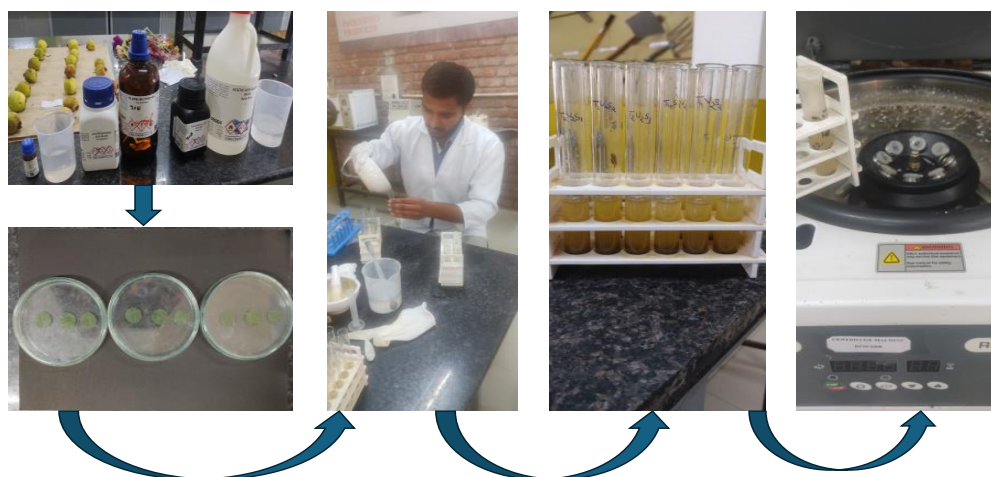


Figure 6: Determination of leaf proline content

for 30 minutes until a brick-red color developed. After cooling, 5 mL of toluene was added, and the mixture was transferred to a separating funnel. The absorbance was measured at 520 nm using a spectrophotometer, with toluene serving as the blank.

Membrane stability index (MSI)

To evaluate the Membrane Stability Index (MSI) in strawberry leaves, fresh leaf samples were collected and rinsed with distilled water. Uniform leaf discs weighing approximately 100 mg were cut and placed in a test tube containing 10 mL of deionized water. The test tubes were then placed in a water bath at 40°C for 30 minutes, after which the initial electrical conductivity (C1) was measured using a conductivity meter. The samples were boiled at 100°C for 10 minutes, allowed to cool to room temperature, and the final conductivity (C2) was recorded. This method, based on the work of Sairam *et al.* (1997), is widely used in plant stress physiology studies to assess cell membrane stability under various stress conditions (Sairam *et al.*, 1997).

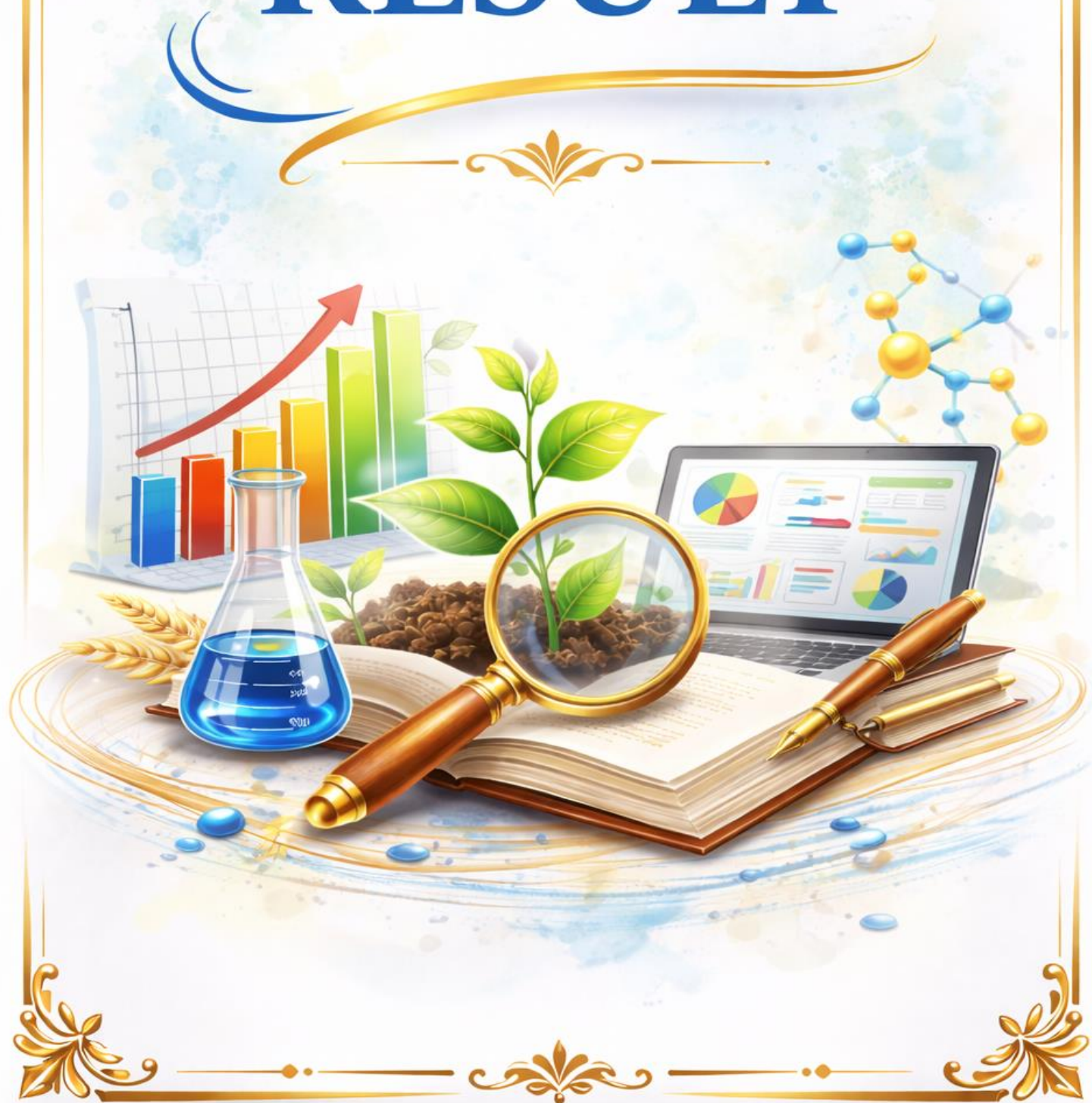
$$MSI = \frac{(C1 - C2)}{(C1)}$$



Figure 7. Determination of membrane stability index

Chapter - 4

RESULT



4.1 LD₅₀ Dosage for gamma irradiation

In the current study, probit analysis was conducted for various doses of gamma rays on runners of the strawberry varieties Winter Dawn and Chandler, as detailed in the (Table 4.1.1) and illustrated in the Fig.4.1 The LD₅₀ value, determined based on lethality following exposure to different doses of gamma irradiation (20 Gy, 30 Gy, 40 Gy, and 0 Gy), was found to be 40 Gy. Compared to the control, a progressive increase in the lethality of runners was observed with higher doses of irradiation, with the highest mortality recorded at 40 Gy (66%) and the lowest at 20 Gy (16%). Consequently, the entire experiment was extended to include further observations on physiological, morphological, yield-related, and salinity stress-related parameters up to the 40 Gy dose.

4.2 LD₅₀ Dosage for ethyl methanesulphonate

In the present investigation, probit analysis for various doses of ethyl methanesulphonate on runners of strawberry varieties Winter Dawn and Chandler was conducted, with the results presented in the (Table 4.2.1) and depicted in the Fig.4.2 The LD₅₀ value, determined based on lethality following treatment with different doses of ethyl methanesulphonate (1% EMS, 2% EMS, 3% EMS, and 0), was observed at the 3% EMS dose. Compared to the control, a steady increase in the lethality of runners was noted as the dose of the chemical mutagen increased, with the highest mortality recorded at the 3% EMS dose (75%) and the lowest at 1% EMS (16%). Therefore, the entire experiment was carried out for further physiological, morphological, yield-related, and salinity stress-related observations in chemical mutagen-treated runners up to the 3% EMS dose.

4.3 Interaction effect of gamma irradiation doses under varying levels of salinity on the strawberry cv. Winter Dawn

4.3.1 Plant height

The data pertaining to the interaction effect of Gamma Irradiation doses and varying levels of salinity on plant height of winter Dawn strawberry is presented in the Table 4.3.1. Plant height which was found to be significantly different. During 2023 the maximum plant height was found (29.52 cm) in runners treated with 30 Gy dose imposed with 75 mM NaCl and minimum was recorded (19.58 cm) in 100 mM NaCl. However, in 2024 The maximum plant height was

Table 4.1.1 LD₅₀ for physical mutagen gamma irradiation (doses ranging from 20 Gy to 40 Gy) of strawberry runners

Conc.	Tot No.	No. Kills	% Mort.	Log (Dose)	Exp. Prop	Emp Probit	Exp Probit	Work. probit	Weight	wx	wy	wx2	wy2	wxy	Y'
20 Gy	12	2	16	1.301	0.000	0.000	-3.465	-3.734	0.044	0.057	-0.164	0.075	0.614	-0.214	-3.465
30 Gy	12	3	25	1.477	0.099	-1.282	-1.289	-1.282	4.078	6.024	-5.226	8.898	6.697	-7.719	-1.289
40 Gy	12	8	66	1.602	0.601	0.253	0.256	0.253	7.460	11.951	1.890	19.147	0.479	3.028	0.256
Control	12	3	27	1.573	0.086	0.372	0.437	0.231	2.321	6.043	-5.133	8.267	6.431	-6.876	-1.134

Component	Mean X	Mean Y	Intercept	Beta	LC ₅₀	LL	UL	Log (LC ₅₀)	Log LL	Log UL	Chi-Square ML	Sign (Chi-Square)
Estimate	36.052	-0.302	-19.544	12.359	38.140	19.023	76.467	1.581	1.279	1.883	0.003	0.953

Parameter	Estimate	Std. Error	t- Cal	p-value
Intercept	-19.544	7.424	-2.633	0.119
Beta	12.359	4.765	2.594	0.122

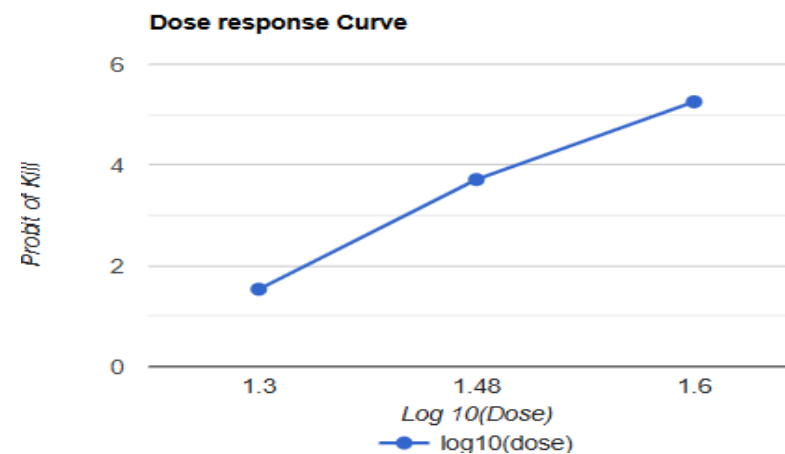


Fig 4.1 Probit curve for gamma irradiation

Table 4.2.1 LD₅₀ for Chemical mutagen ethyl methane sulfonate (doses range from 1% EMS to 3% EMS) of strawberry runners

Conc.	Tot No.	No. Kills	% Mort.	Log (Dose)	Exp. Prop	Emp Probit	Exp Probit	Work. probit	Weight	wx	wy	wx2	wy2	wxy	Y'
1% EMS	12	2	16	0	0	0	-4.37	-4.588	0.002	0	-0.007	0	0.033	0	-4.37
2% EMS	12	3	25	0.301	0.1	-1.282	-1.282	-1.282	4.106	1.236	-5.262	0.372	6.743	-1.584	-1.282
3% EMS	12	9	75	0.477	0.7	0.524	0.524	0.524	6.908	3.296	3.622	1.573	1.9	1.728	0.524
Control	12	4	34	0.413	0.3	0.467	0.276	0.343	3.276	2.364	4.763	0.267	5.432	2.346	1.677

Component	Mean X	Mean Y	Intercept	Beta	LC ₅₀	LL	UL	Log (LC ₅₀)	Log LL	Log UL	Chi-Square ML	Sign (Chi-Square)
Estimate	2.579	2.579	2.579	2.579	2.579	2.579	2.579	2.579	2.579	2.579	0	0.993

Parameter	Estimate	Std. Error	t- Cal	p-value
Intercept	-19.544	7.424	-2.633	0.119
Beta	12.359	4.765	2.594	0.122

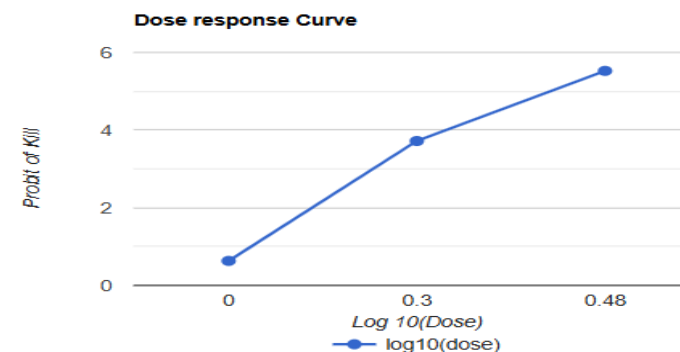


Fig 4.2. Probit curve for Ethyl methanesulfonate

observed (28.37 cm) in 20 Gy dose treated runners when exposed imposed with 100 mM NaCl and minimum height (20.84 cm) was found in 100 mM NaCl. The pooled data of two factor show maximum plant height was observed in 30 Gy dose treated runners when exposed to 75 mM NaCl salinity level (28.37 cm). Which was statistically at par with 30 Gy X Control, 30 Gy X 50 mM NaCl and 20 Gy X 100. However, the minimum plant height was recorded in runners was exposed to 100 mM salinity condition (18.93 cm). Hence, it can be concluded that 30 Gy showed the best result in the significant increase of plant height in salinity exposed conditions instead of treated runners with 40 Gy dose.

4.3.2 Chlorophyll content index

The (Table 4.3.1) presents the data on the combined effect of different physical mutagen doses and varying salinity levels on the chlorophyll content index of Winter Dawn strawberry. A similar pattern was observed in 2023 and 2024 with the pooled data for chlorophyll content index, showing significant differences. The maximum chlorophyll content index reading was observed in runners treated with a 20 Gy dose when exposed to 50 mM NaCl (58.18), which was statistically at par with 30 Gy X 50 mM NaCl, 40 Gy X 50 mM NaCl and 20 Gy 75 mM NaCl. On the other hand, the minimum chlorophyll content index was recorded in the control treated with 75 mM NaCl is (51.73). In conclusion, 20 Gy gave the best results by significantly increasing chlorophyll content index under salinity stress, while 40 Gy was found to be less effective.

4.3.3 Number of buds

The analysis of the data reveals that gamma irradiation increased the number of buds in mutagen-treated runners instead 40 Gy dose. In the Winter Dawn variety, the effects of gamma irradiation treatment and varying levels of salinity are evident from the pooled data (Table 4.3.2). The maximum number of buds was recorded in runners treated with a 30 Gy gamma irradiation dose and exposed to 75 mM NaCl (28.16). Which was statistically at par with 40 Gy X 50 mM NaCl and 20 Gy X 50 mM NaCl. While the minimum number of buds was observed in runners exposed to 100 mM NaCl (22.86). A similar trend was observed during 2023, where the treated runners showed the maximum number of buds under plants induced with 75 mM NaCl salinity condition (27.16). However, in 2024, the maximum number of buds was recorded in runners treated with a 20 Gy dose and exposed to 50 mM NaCl (29.21), while the minimum was observed in plants exposed to 75 mM NaCl (21.33).

Table 4.3.1 Interaction effect of gamma irradiation doses under different levels of salinity on plant height and chlorophyll of strawberry cv. Winter Dawn

Treatment	Plant height 2023	Plant height 2024	Plant height pooled data	Chlorophyll 2023	Chlorophyll 2024	Chlorophyll pooled data	Treatment	Plant height 2023	Plant height 2024	Plant height pooled data	Chlorophyll 2023	Chlorophyll 2024	Chlorophyll pooled data	
20 Gy X Control	27.76	24.39	26.17	58.43	52.30	55.36	Control	20.62	22.10	24.36	59.52	47.86	53.13	
20 Gy X 50 mM NaCl	27.53	24.64	26.10	60.23	55.93	58.18	Control X 50 mM NaCl	21.88	22.18	26.53	60.85	46.86	53.86	
20 Gy X 75 mM NaCl	26.10	23.72	24.91	59.36	51.40	55.38	Control X 75 mM NaCl	20.45	21.33	20.89	61.13	42.40	51.73	
20 Gy X 100 mM NaCl	24.73	28.37	26.55	54.11	50.66	52.36	Control X 100 mM NaCl	19.58	20.84	18.93	58.74	45.30	52.02	
30 Gy X Control	28.70	26.57	27.63	58.32	51.13	54.68	C.D. for Years	0.328	0.526					
30 Gy X 50 mM NaCl	28.63	26.39	27.51	62.55	52.26	57.40	C.D. for Treatment	2.818	7.655					
30 Gy X 75 mM NaCl	29.52	27.23	28.37	60.18	49.53	54.85	C.D. for Years X Treatments	2.225	3.128					
30 Gy X 100 mM NaCl	25.32	23.55	24.43	56.68	50.10	53.39								
40 Gy X Control	27.68	24.61	26.14	58.82	49.60	54.21								
40 Gy X 50 mM NaCl	24.88	21.59	23.23	61.47	49.60	55.53								
40 Gy X 75 mM NaCl	25.67	22.53	24.10	61.16	47.40	54.28								
40 Gy X 100 mM NaCl	25.27	23.30	24.28	56.39	49.70	53.11								

4.3.4 Number of flowers

Evident from the (Table 4.3.2.) that the number of flowers was significantly affected in runners in winter Dawn strawberry variety. The pooled results of gamma irradiation-treated runners with varying levels of salinity treatment show a maximum number of flowers when treated with 30 Gy dose treated runners exposed to 75 mM salinity level (31.83), which was statistically at par with 30 Gy X 50 mM NaCl, 30 Gy X Control and 20 Gy X 50 mM NaCl. However, a minimum number of flowers recorded in runners exposed to 100 mM NaCl (24.23). In 2023 a similar trend was observed, with the treated runners exhibiting the highest number of flowers under the 75 mM salinity condition (32.33). However, in 2024, the maximum number of flowers was recorded in runners treated with a 20 Gy dose exposed to 50 mM NaCl (29.33), while the minimum number of flowers was observed in plant exposed to 75 mM salinity condition (22.46).

4.3.5 Number of fruits

The data presented in (Table 4.3.3) indicates that gamma irradiation treated runners of the winter Dawn variety showed significantly increased the number of fruits per plant in salinity exposed plants, except at the 40 Gy dose. The number of fruits ranged from (24.25) to (33.33). The pooled data reveals that the highest number of fruits per plant was obtained from runners treated with a 30 Gy dose under 75 mM salinity level (33.33), which was statistically at par with 30 Gy X 50 mM NaCl, 40 Gy X Control and 40 Gy X 75 mM NaCl while the minimum was recorded in runners exposed to 100 mM NaCl (24.25). A similar trend was observed in 2024 treated runners showed the maximum number of fruits was recorded under 75 mM NaCl (33) and the minimum number of fruits (23.19) under 100 mM NaCl salinity condition. However, in 2023, the maximum number of fruits was recorded in 20 Gy treated runners exposed to 75 mM NaCl (34.66), while the minimum was noted in runners exposed to 100 mM NaCl salinity (25.31). Gamma irradiation, particularly at doses 30 Gy, enhanced the number of fruits per plant under salinity stress, while 40 Gy and higher salinity with runners led to reduced fruit.

4.3.6 Days to bud

The data presented in (Table 4.3.3) reveals that, in the Winter Dawn variety, the combined effect of different physical mutagen doses and varying levels of induced salinity significantly influenced the days to bud initiation from planting. The pooled data of the two factors show

that early bud initiation was recorded in runners treated with a 30 Gy mutagen dose without NaCl (0 mM) salinity (48.33), late bud initiation was observed in control runners when exposed to 75 mM NaCl salinity (67.16). A similar trend was observed in 2023 and 2024, where early bud initiation was recorded in runners treated with 30 Gy physical mutagens dose, as opposed to those treated with 40 Gy.

4.3.7 Days to flower

The data pertaining to flower initiation days from planting in the Winter Dawn variety, as shown in the (Table 4.3.4), reveals the interaction effect of physical mutagen treatment under NaCl salinity stress. It is evident from the combined effect that early flower initiation at (56) was recorded in runners treated with a 30 Gy mutagen dose. However late flower initiation was observed in runners exposed to 75 mM salinity dose (75.16). A similar trend was observed during 2023 and 2024. 30 Gy gamma irradiation dose showed early flower initiation instead of 40 Gy dose.

4.3.8 Days to fruit

It is apparent from the data depicted in the (Table 4.3.4) that gamma irradiation treated runners significantly showed earlier fruit initiation in salinity exposed plants of the Winter Dawn variety. The combined effect shows early fruit initiation of both years in 30 Gy gamma irradiation treated runners exposed with 50 mM NaCl stress (79.83) and late fruit initiation was recorded in runners grown under 100 mM NaCl treatment (93). A significant trend was observed in 2023 and 2024 along with pooled data with respect to bud initiation days.

4.3.9 Fruit length

The inquisition of data presented in (Table 4.3.5) Illustrate plate 8 shows the combined effect of different gamma irradiation doses and varying levels of salinity on the tolerance of strawberry plants to salinity in winter Dawn variety. It is evident from the pooled data that the maximum fruit length was recorded in runners treated with a 30 Gy mutagen dose (43.97 mm) Illustrate in Figure 4.3 which was statistically at par with 20 Gy X 100 mM NaCl and 40 Gy X Control. The minimum fruit length was recorded in runners induced with 75 mM NaCl (32.12 mm). A similar trend was observed in 2023 and 2024, where the maximum fruit length was recorded in runners treated with 20 Gy doses instead of 40 Gy in salinity-exposed plants.

Table 4.3.2 Interaction effect of gamma irradiation doses under different levels of salinity on number of buds and flowers of strawberry cv. Winter Dawn

Treatment	Number of buds 2023	Number of buds 2024	Number of buds pooled data	Number of flowers 2023	Number of flowers 2024	Number of flowers pooled Data	Treatment	Number of buds 2023	Number of buds 2024	Number of buds pooled data	Number of flowers 2023	Number of flowers 2024	Number of flowers pooled data
20 Gy X Control	27	25	26	30	27	28.50	Control	26	26	26	30.66	30.66	30.66
20 Gy X 50 mM NaCl	24.33	29.21	26.77	31.33	29.33	30.33	Control X 50 mM NaCl	25.66	22	23.13	25.43	24.66	25.04
20 Gy X 75 mM NaCl	25.66	24.66	25.16	29	26	27.50	Control X 75 mM NaCl	25.21	21.33	23.27	26.35	22.46	24.40
20 Gy X 100 mM NaCl	26	24.66	25.33	28	24.66	26.33	Control X 100 mM NaCl	22.39	23.33	22.86	23.69	24.78	24.23
30 Gy X Control	26	25	25.50	31.66	29.66	30.66	C.D. for Years	0.313	1.104				
30 Gy X 50 mM NaCl	27.33	27	27.16	33.66	29	31.33	C.D. for Treatment	2.189	2.957				
30 Gy X 75 mM NaCl	27.66	28.66	28.16	32.33	31.33	31.83	C.D. for Years X Treatments	3.130	2.767				
30 Gy X 100 mM NaCl	25	23	24	28	27	27.50							
40 Gy X Control	25.66	25.66	25.66	28.66	27	27.83							
40 Gy X 50 mM NaCl	27.33	28	27.66	33.66	29	30.33							
40 Gy X 75 mM NaCl	26.33	24.33	25.33	31	29	30							
40 Gy X 100 mM NaCl	26	25.33	25.66	25.43	26.19	25.81							

Table 4.3.3 Interaction effect of gamma irradiation doses under different levels of salinity on number of fruits and days to bud of strawberry cv. Winter Dawn

Treatment	No. of fruits 2023	No. of fruits 2024	No. of fruits pooled data	Days to bud 2023	Days to bud 2024	Days to bud pooled data	Treatment	No. of fruits 2023	No. of fruits 2024	No. of fruits pooled data	Days to bud 2023	Days to bud 2024	Days to bud pooled data	
20 Gy X Control	32.66	30	31.33	61	51.33	56.16	Control	30	30.33	30.16	67	60	63.50	
20 Gy X 50 mM NaCl	30.66	29	29.83	50.33	50.66	50.50	Control X 50 mM NaCl	32	31	31.50	69.33	60.66	65	
20 Gy X 75 mM NaCl	32	30.33	31.16	49	57.66	53.33	Control X 75 mM NaCl	32.33	29.33	30.83	68	66.33	67.16	
20 Gy X 100 mM NaCl	30	28	29	59.33	54.66	57	Control X 100 mM NaCl	25.31	23.19	24.25	62.33	60.66	61.50	
30 Gy X Control	32.33	31.33	31.83	46.66	50	48.33	C.D. for Years	1.069	2.426					
30 Gy X 50 mM NaCl	33.66	32	32.83	46.33	53	49.66	C.D. for Treatment	2.644	11.063					
30 Gy X 75 mM NaCl	33.41	33	33.33	60.33	59.33	59.83	C.D. for Years X Treatments	2.696	5.713					
30 Gy X 100 mM NaCl	30.33	30.33	30.33	62	62	62								
40 Gy X Control	32.66	31	31.83	58.66	56	57.33								
40 Gy X 50 mM NaCl	33	31.66	32.33	59.66	60	59.83								
40 Gy X 75 mM NaCl	34.66	29	31.83	59.66	55	57.33								
40 Gy X 100 mM NaCl	30.33	29.66	30	64.66	50.66	57.66								

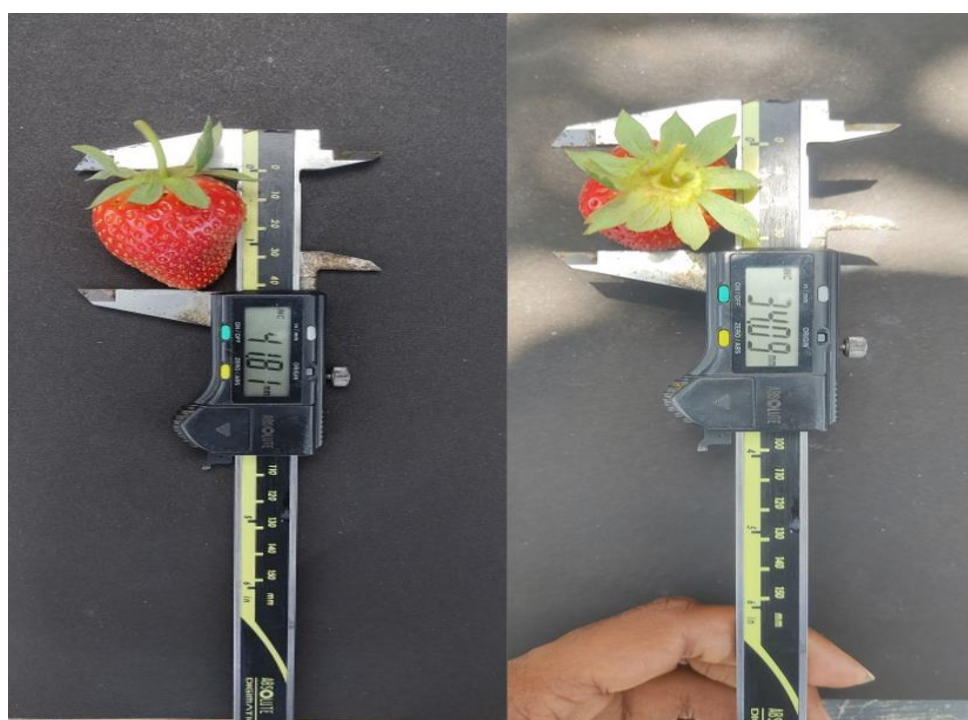
Table 4.3.4 Interaction effect of gamma irradiation doses under different levels of salinity on days to flower and fruits of strawberry cv.

Winter Dawn

Treatment	Days to flowers 2023	Days to flowers 2024	Days to flower pooled data	Days to fruits 2023	Days to fruits 2024	Days to fruit pooled data	Treatment	Days to flowers 2023	Days to flowers 2024	Days to flower pooled data	Days to fruits 2023	Days to fruits 2024	Days to fruit pooled data
20 Gy X Control	70	63	66.50	84.33	82	84.16	Control	77.66	69	73.36	90	89.33	89.66
20 Gy X 50 mM NaCl	61.33	61.66	61.50	88.33	86.33	87.33	Control X 50 mM NaCl	80.66	67	73.83	89.76	90.43	90.19
20 Gy X 75 mM NaCl	59.66	59.66	59.66	89.33	89	89.16	Control X 75 mM NaCl	79.33	71	75.16	93.65	91.76	92.70
20 Gy X 100 mM NaCl	64.33	60	62.16	90.66	87.33	89	Control X 100 mM NaCl	72	71	71.50	93.33	92.66	93
30 Gy X Control	56	56	56	72	87.66	79.83	C.D. for Years	2.30	1.80				
30 Gy X 50 mM NaCl	60.66	59	59.83	84	83.33	83.66	C.D. for Treatment	8.33	8.70				
30 Gy X 75 mM NaCl	67	64	65	91	88.66	87.83	C.D. for Years X Treatments	5.56	14.44				
30 Gy X 100 mM NaCl	73	65	69	87.66	89	88.33							
40 Gy X Control	69	64.33	66.17	90	85.66	87.83							
40 Gy X 50 mM NaCl	61	61	61	90	86	88							
40 Gy X 75 mM NaCl	71	61.33	66.16	86.66	84	85.33							
40 Gy X 100 mM NaCl	74.33	64.66	69.41	88.33	88.66	88.50							

4.3.10 Fruit diameter

The analysis of the data reveals that gamma irradiation significantly influenced the fruit diameter of Winter Dawn strawberry variety. The pooled data (Table 4.3.5) shows that the maximum fruit diameter was recorded in runners treated with a 30 Gy gamma irradiation dose induced with 50 mM salinity condition (33.47 mm) Illustrate in Plate 4.3, which was statistically at par with 30 Gy X Control and 40 Gy X 50 mM NaCl the minimum fruit diameter was observed in runners exposed to 75 mM NaCl (25.22 mm). However, in 2023, the maximum fruit diameter was recorded in runners treated with 20 Gy under 75 mM NaCl (35.21 mm), while the minimum diameter occurred in runners exposed to 100 mM salinity level (24.39 mm). A similar trend was recorded in 2024 the maximum fruit diameter was recorded under 100 mM NaCl (33.12 mm), while the minimum was recorded in runners induced against 50 mM salinity level (22.79 mm).



41.81 mm Fruit length in
30 Gy X Control

34.09 mm Fruit diameter in
30 Gy X 50 mM NaCl

Plate 8. Fruit length and fruit diameter

4.3.11 Fruit weight

It is inquisition from the (Table 4.3.6) that fruit weight was significantly affected in runners of the Winter Dawn strawberry variety. The pooled data showed the maximum fruit weight was recorded in runners treated with a 30 Gy dose exposed to 50 mM salinity condition (13.32 gm),

which was statistically at par with 20 Gy X Control and 20 Gy X 50 mM NaCl. However, the minimum fruit weight was recorded in plants exposed to 100 mM salinity level (10.17 gm). A similar trend was recorded 2023 and 2024. From the above results, it was observed that the 20 Gy and 30 Gy gamma irradiation doses resulted in the maximum fruit weight compared to the 40 Gy dose and control plants.

4.3.12 Fruit yield

It is evident from the data that fruit yield was significantly affected in control runners and 40 Gy dose-treated runners of the winter Dawn strawberry variety (Table 4.3.6). The pooled results of gamma irradiation-treated plants, subjected to varying levels of salinity, show the maximum fruit yield was recorded in plants treated with a 30 Gy dose exposed to 50 mM salinity level (350.33 gm), which was statistically at par with 20 Gy X 50 mM NaCl and 30 Gy X 75 mM NaCl. However, the minimum fruit yield was recorded in runners induced against 100 mM NaCl (257.16 gm). However, in 2023, the mutagen-treated runners showed the maximum fruit yield under the 50 mM salinity level (388.66 gm), while the minimum yield was recorded in runners exposed to 75 mM NaCl (257.66 gm). During 2024, the maximum fruit yield was recorded in runners treated with a 20 Gy dose and exposed to 100 mM NaCl (326 gm), while the minimum yield of (219.33 gm) was observed in runners exposed to 100 mM salinity level.

4.3.13 Shoot fresh weight

The analysis of the data reveals that gamma irradiation treated runners significantly influenced the shoot fresh weight of Winter Dawn strawberry plants (Table 4.3.7) The pooled data shows that the maximum shoot fresh weight was recorded in plants treated with a combination of 20 Gy gamma irradiation against 50 mM salinity level (14.53 gm), which was statistically at par with 30 Gy X 50 mM NaCl and 30 Gy X 75 mM NaCl. Conversely, the minimum shoot fresh weight was observed in runners exposed to 100 mM salinity treatment (10.18 gm). A similar trend was observed in 2024, where the treated runners exhibited the maximum shoot fresh weight under the 50 mM NaCl salinity level (13.88 gm), while the minimum shoot fresh weight was recorded under the 75 mM NaCl salinity condition (9.32 gm). But during 2023, the maximum shoot fresh weight was recorded in plants treated with 30 Gy exposed to 75 mM NaCl level (16.10 gm), while the minimum weight was recorded in runners exposed to 100 mM salinity level (10.10 gm).

4.3.14 Shoot dry weight

The analysis of the data reveals that gamma irradiation-treated runners induced against salinity stress significantly influenced the shoot dry weight of Winter Dawn strawberry plants (Table 4.3.7) The pooled data shows that the maximum shoot dry weight was recorded in plants treated with 20 Gy gamma irradiation treated runners under 50 mM salinity level (3.51 gm), while the minimum shoot dry weight was observed in plants exposed to 100 mM salinity level (1.98 gm). In 2023, the maximum shoot dry weight was recorded in runners treated with 30 Gy gamma irradiation treated runners induced against 100 mM NaCl (3.91 gm), while the minimum shoot dry weight was found in plants exposed to 75 mM NaCl salinity condition (1.78 gm). In 2024, a similar trend was observed, with the treated runners showing the highest shoot dry weight (3.43 gm) under the 50 mM NaCl salinity condition, while the lowest was recorded in runners subjected to 100 mM salinity stress (2.11 gm). Thus, it can be concluded that 20 Gy and 30 Gy treatments resulted in the most significant increase in shoot dry weight under salinity-stressed conditions, instead to 40 Gy dose.

4.3.15 Root fresh weight

It is evident from the data that root fresh weight in the Winter Dawn strawberry variety was significantly affected by salinity stress in 40 Gy dose and control runners (Table 4.3.8) The pooled data values show the highest root fresh weight was recorded in runners treated with a 30 Gy dose exposed to 75 mM NaCl (14.59 gm). Which was statistically at par with 20 Gy X 50 mM NaCl, 30 Gy X 75 mM NaCl and 30 Gy X 75 mM NaCl. However, the minimum root fresh weight was recorded in runners exposed to 100 mM NaCl (10.57 gm). In 2023, the maximum root fresh weight of was observed in the combination of a 20 Gy dose against 75 mM NaCl (15.63 gm), while the minimum was recorded in plants exposed to 100 mM NaCl (11.39 gm).

Table 4.3.5 Interaction effect of gamma irradiation doses under different levels of salinity on fruit length and diameter of strawberry cv. Winter Dawn

Treatment	Fruit length 2023	Fruit length 2024	Fruit length pooled data	Fruit diameter 2023	Fruit diameter 2024	Fruit diameter pooled data	Treatment	Fruit length 2023	Fruit length 2024	Fruit length pooled data	Fruit diameter 2023	Fruit diameter 2024	Fruit diameter pooled data
20 Gy X Control	39.19	39.74	39.46	30.44	30.43	30.44	Control	39.98	39.42	39.70	30.44	26.91	28.67
20 Gy X 50 mM NaCl	42.84	40.29	40.10	30.93	28.59	29.76	Control X 50 mM NaCl	37.83	37.52	37.68	30.69	22.79	26.74
20 Gy X 75 mM NaCl	41.50	38.9	39.76	35.21	29.39	30.10	Control X 75 mM NaCl	37.64	36.49	37.12	25.76	24.69	25.22
20 Gy X 100 mM NaCl	42.58	41.59	42.39	29.46	33.12	30.15	Control X 100 mM NaCl	37.69	37.55	37.62	24.39	28.59	26.49
30 Gy X Control	43.76	44.19	43.97	34.13	31.25	32.69	C.D. for Years	0.646	0.387				
30 Gy X 50 mM NaCl	42.57	40.36	41.46	34.48	32.47	33.47	C.D. for Treatment	2.463	2.208				
30 Gy X 75 mM NaCl	40.54	37.59	39.10	31.12	28.53	29.83	C.D. for Years X Treatments	2.126	2.353				
30 Gy X 100 mM NaCl	42.7	39.51	40.79	30.58	29.59	30.9							
40 Gy X Control	40.92	40.92	40.92	30.63	27.69	28.22							
40 Gy X 50 mM NaCl	38.32	38.57	38.44	33.89	31.32	30.60							
40 Gy X 75 mM NaCl	40.58	38.17	39.37	31.20	28.70	29.52							
40 Gy X 100 mM NaCl	40.48	38.61	39.55	30.51	26.86	27.87							

Table 4.3.6 Interaction effect of gamma irradiation doses under different levels of salinity on fruits weight and fruit yield of strawberry cv. Winter Dawn

Treatment	Fruit weight 2023	Fruit weight 2024	Fruit weight pooled data	Fruit yield 2023	Fruit yield 2024	Fruit yield pooled data	Treatment	Fruit weight 2023	Fruit weight 2024	Fruit weight pooled data	Fruit yield 2023	Fruit yield 2024	Fruit yield pooled data				
20 Gy X Control	12.46	11.69	12.17	354.66	292	323.33	Control	11.88	9.92	10.96	385.66	277.66	331.66				
20 Gy X 50 mM NaCl	11.84	10.74	11.29	388.66	296	342.33	Control X 50 mM NaCl	11.54	9.89	10.71	341.66	248.33	295				
20 Gy X 75 mM NaCl	11.46	10.61	11.13	367	250.66	308.83	Control X 75 mM NaCl	10.71	9.70	10.20	257.29	263.10	260.19				
20 Gy X 100 mM NaCl	12.21	10.35	11.28	279.66	326	302.83	Control X 100 mM NaCl	10.10	10.24	10.17	295	219.33	257.16				
30 Gy X Control	11.95	9.29	10.62	361.33	296.33	328.83	C.D. for Years	0.328	9.407								
30 Gy X 50 mM NaCl	12.78	13.87	13.32	385.33	315.33	350.33	C.D. for Treatment	1.261	49.849								
30 Gy X 75 mM NaCl	11.72	10.27	10.99	377	290	333.5	C.D. for Years X Treatments	1.285	74.38								
30 Gy X 100 mM NaCl	13.44	10.36	11.19	329.66	284	306.83											
40 Gy X Control	10.8	9.73	10.26	310.33	294.33	302.33											
40 Gy X 50 mM NaCl	10.92	10.34	10.63	380.33	295.66	338											
40 Gy X 75 mM NaCl	11.71	9.85	10.78	379	278.33	328.66											
40 Gy X 100 mM NaCl	9.87	11.32	10.59	343.66	272	307.83											

Table 4.3.7 Interaction effect of gamma irradiation doses under different levels of salinity on shoot fresh weight and dry weight of strawberry cv. Winter Dawn

Treatment	Shoot fresh wt. 2023	Shoot fresh wt. 2024	Shoot fresh wt. pooled data	Shoot dry wt. 2023	Shoot dry wt. 2024	Shoot dry wt. pooled data	Treatment	Shoot fresh wt. 2023	Shoot fresh wt. 2024	Shoot fresh wt. pooled data	Shoot dry wt. 2023	Shoot dry wt. 2024	Shoot dry wt. pooled data	
20 Gy X Control	12.14	14.10	13.12	3.21	3.19	3.15	Control	10.53	10.17	10.35	3.15	2.36	2.70	
20 Gy X 50 mM NaCl	15.19	13.88	14.53	3.78	3.25	3.51	Control X 50 mM NaCl	11.14	9.50	10.32	2.94	2.88	2.91	
20 Gy X 75 mM NaCl	13.96	11.71	12.84	3.25	2.88	3.07	Control X 75 mM NaCl	12.13	9.32	10.67	1.78	2.96	2.37	
20 Gy X 100 mM NaCl	10.87	10.70	10.78	2.81	2.99	2.90	Control X 100 mM NaCl	10.10	10.27	10.18	1.86	2.11	1.98	
30 Gy X Control	12.43	10.84	11.63	3.27	2.95	3.11	C.D. for Years	0.665	0.212					
30 Gy X 50 mM NaCl	15.35	13.28	14.31	3.27	3.43	3.35	C.D. for Treatment	2.537	0.729					
30 Gy X 75 mM NaCl	16.10	10.89	13.49	3.34	2.61	2.97	C.D. for Years X Treatments	1.426	0.937					
30 Gy X 100 mM NaCl	10.78	10.9	10.42	3.91	2.95	3.43								
40 Gy X Control	12.98	11.57	12.28	3.22	2.9	2.64								
40 Gy X 50 mM NaCl	15.8	10.86	12.96	3.71	2.99	3.35								
40 Gy X 75 mM NaCl	14.1	10.54	12.32	2.96	2.9	2.48								
40 Gy X 100 mM NaCl	11.20	10.25	10.72	2.93	2.19	2.56								

A Similar trend was recorded in 2024. In conclusion, a 30 Gy dose of gamma irradiation shows promising results for enhancing root fresh weight under salinity stress conditions.

4.3.16 Root dry weight

The results from the study indicate that gamma radiation had a significant impact on root dry weight, with notable differences observed between treated and runners under salinity stress conditions in Winter Dawn strawberry variety (Table 4.3.8) The pooled analysis revealed that the highest root dry weight was observed in plants treated with a 20 Gy dose induced with 50 mM NaCl (4.27 gm), as well as the lowest root dry weight was recorded in plants exposed to 75 mM NaCl (2.80 gm).

A similar trend was recorded during 2023 and 2024. These results highlight the efficacy of gamma radiation, particularly at a 20 Gy dose, in enhancing root development under salinity stress conditions instead of 40 Gy dose.

4.3.17 Number of leaves

The analysis of data in (Table 4.3.9) demonstrates that, in the Winter Dawn variety, the combined influence of varying gamma irradiation doses and salinity levels significantly affected the number of leaves, reflecting the plant tolerance to salinity stress. The pooled data reveal that the highest number of leaves, was observed in plants treated with a combination of 30 Gy gamma irradiation dose induced against 50 mM NaCl (11.83), which was statistically at par with 20 Gy X Control and 30 Gy X Control in contrast, the lowest leaf count was recorded in runners exposed to 100 mM NaCl (8). In 2023, the treated runners had the highest number of leaves recorded under the 50 mM salinity level (11.83), while the lowest number of leaves was observed in plants treated with 40 Gy and exposed to 75 mM salinity condition (2.66). Similar trend was observed during 2024.

4.3.18 Leaf relative water content

(Table 4.3.9) represents that gamma radiation significantly influenced leaf relative water content (LRWC) in the Winter Dawn strawberry variety under salinity stress, with substantial differences between treated and control runners. The pooled analysis revealed that the highest LRWC was recorded in plants treated with a 30 Gy dose under 75 mM salinity stress conditions (81.69 %), which was statistically at par with 30 Gy, while the lowest LRWC was observed in runners exposed to 100 mM NaCl (72.37 %) (Table 4.3.9).

Table 4.3.8 Interaction effect of gamma irradiation doses under different levels of salinity on root fresh weight and dry weight of strawberry cv. Winter Dawn

Treatment	Root fresh wt. 2023	Root fresh wt. 2024	Root fresh wt. pooled data	Root dry wt. 2023	Root dry wt. 2024	Root dry wt. pooled data	Treatment	Root fresh wt. 2023	Root fresh wt. 2024	Root fresh wt. pooled data	Root dry wt. 2023	Root dry wt. 2024	Root dry wt. pooled data
20 Gy X Control	14.35	11.80	13.11	3.84	3.46	3.65	Control	12.77	9.98	11.95	3.87	2.43	3.15
20 Gy X 50 mM NaCl	14.76	13.89	14.32	4.58	3.96	4.27	Control X 50 mM NaCl	13.43	10.11	11.37	4.22	2.10	3.16
20 Gy X 75 mM NaCl	15.63	10.49	13.16	3.96	2.81	3.38	Control X 75 mM NaCl	10.76	13.15	11.75	4.13	1.56	2.80
20 Gy X 100 mM NaCl	13.88	11.85	12.87	4.85	2.34	3.59	Control X 100 mM NaCl	11.39	9.76	10.57	2.63	1.86	2.24
30 Gy X Control	13.82	11.96	12.89	3.85	3.23	3.54	C.D. for Years	0.539	0.189				
30 Gy X 50 mM NaCl	15.32	13.87	14.59	4.84	3.68	4.26	C.D. for Treatment	1.495	1.533				
30 Gy X 75 mM NaCl	15.22	12.26	13.65	4.12	2.97	3.54	C.D. for Years X Treatments	1.806	0.922				
30 Gy X 100 mM NaCl	13.37	10.65	12.9	3.77	3.42	3.60							
40 Gy X Control	14.11	10.18	12.12	4.12	1.92	2.97							
40 Gy X 50 mM NaCl	15.28	11.50	13.39	3.23	2.86	3.4							
40 Gy X 75 mM NaCl	15.13	10.94	13.9	4.36	1.85	3.10							
40 Gy X 100 mM NaCl	13.37	11.67	12.52	3.77	2.25	3.10							

represents that gamma radiation significantly influenced leaf relative water content (LRWC) in the Winter Dawn strawberry variety under salinity stress, with substantial differences between treated and control runners. The pooled analysis revealed that the highest LRWC was recorded in plants treated with a 30 Gy dose under 75 mM salinity stress conditions (81.69 %), which was statistically at par with 20 Gy mutagen dose, while the lowest LRWC was observed in runners exposed to 100 mM NaCl (72.37 %). A similar trend was observed during 2023 and 2024. These findings show the positive impact of gamma irradiation, particularly at a 30 Gy dose, in enhancing LRWC under salinity stress conditions.

4.3.19 Leaf area

The results of this observation indicate that a specific dose of gamma irradiation maximizes leaf area under induced salinity stress with NaCl treatment (Table 4.3.10). In the pooled analysis, the maximum leaf area was recorded in plants treated with a 30 Gy dose against 100 mM salinity condition (21.45 mm), which was recorded statistically at par with 30 Gy X 75 mM NaCl and 20 Gy X 50 mM NaCl, while the minimum leaf area was observed in runners under the 100 mM salinity level (13.47 mm). A similar trend was recorded during 2023 mutagen treated runners shows maximum leaf area recorded under 75 mM salinity condition (23.49 mm) and the minimum was recorded in runners induced with 75 mM salinity level (12.55 mm). However, in 2024, the maximum leaf area was recorded in runners treated with a 20 Gy dose against 75 mM NaCl (21.32 mm), while the minimum leaf area was observed in runners induced with 50 mM salinity level (10.78 mm).

Table 4.3.9 Interaction effect of gamma irradiation doses under different levels of salinity on number of leaves and leaf relative water content of strawberry cv. Winter Dawn

Treatment	No. of leaves 2023	No. of leaves 2024	No. of leaves pooled data	LRWC 2023	LRWC 2024	LRWC pooled data	Treatment	No. of leaves 2023	No. of leaves 2024	No. of leaves pooled data	LRWC 2023	LRWC 2024	LRWC pooled data
20 Gy X Control	13.33	9.33	11.33	80.37	78.14	79.25	Control	12	8	10	80.62	67.81	74.21
20 Gy X 50 mM NaCl	10.66	8	9.33	80.68	78.53	79.61	Control X 50 mM NaCl	12.33	8.33	10	80.88	69.55	75.22
20 Gy X 75 mM NaCl	12	7	9.50	81.68	77.76	79.72	Control X 75 mM NaCl	12	6	9	80.47	67.22	73.85
20 Gy X 100 mM NaCl	10	7.66	8.83	80.48	77.23	78.85	Control X 100 mM NaCl	7	9	8	79.58	65.16	72.37
30 Gy X Control	12	8.23	10.11	82.18	77.88	80.03	C.D. for Years	0.53	1.11				
30 Gy X 50 mM NaCl	12.66	11	11.83	83.24	79.82	81.53	C.D. for Treatment	2.48	8.23				
30 Gy X 75 mM NaCl	11.33	6.33	8.83	82.93	80.45	81.69	C.D. for Years X Treatments	2.54	2.95				
30 Gy X 100 mM NaCl	10.33	7.33	8.83	80.53	77.22	78.87							
40 Gy X Control	11	8	9.50	79.88	72.17	76.03							
40 Gy X 50 mM NaCl	11	9	10	82.11	73.14	77.62							
40 Gy X 75 mM NaCl	2.66	7	9.83	81.19	71.20	76.19							
40 Gy X 100 mM NaCl	11	6.66	8.83	79.61	70.76	75.18							

4.3.20 Malondialdehyde content

(Table 4.3.10) In the pooled analysis, the minimum malonaldehyde content was found in runners treated with a 20 Gy dose of gamma radiation under 100 mM NaCl stress (2.05 $\mu\text{mol/g}$) while the maximum malonaldehyde content was recorded in runners exposed with 100 mM NaCl stress (2.05 $\mu\text{mol/g}$). The results of the study indicate that gamma radiation significantly impacted malonaldehyde content, with notable differences observed between treated and strawberry runners of the Winter Dawn variety under salinity stress. In 2023, the minimum malonaldehyde content was recorded in runners treated with a 30 Gy dose of gamma irradiation under 75 mM NaCl stress (1.95 $\mu\text{mol/g}$). In contrast, the maximum malonaldehyde content was observed in runners subjected to the same 75 mM salinity conditions (9.49 $\mu\text{mol/g}$). In 2024, a similar trend was observed, with the treated runners showing the minimum malondialdehyde content (1.84 $\mu\text{mol/g}$) under 100 mM salinity stress, while the maximum malondialdehyde content was recorded in runners under the same 100 mM NaCl stress condition (10.6 $\mu\text{mol/g}$).

4.3.21 Electrolyte leakage

The results from the study indicate that physical mutagenesis had a significant effect on electrolyte leakage in the Winter Dawn strawberry variety under salinity conditions (Table 4.3.11) The pooled analysis revealed that the lowest electrolyte leakage was observed in plants treated with a 30 Gy dose exposed to 50 mM NaCl (24.56 %), while the maximum electrolyte leakage was recorded in plants exposed to 100 mM NaCl (34.17 %). A similar trend was observed during 2023 and 2024. These results emphasize the role of gamma radiation, particularly at a 30 Gy dose, in reducing electrolyte leakage under salinity stress instead of runners and 40 Gy dose.

4.3.22 Proline content

The results from the study indicate that gamma radiation had a significant impact on proline content, with notable differences observed between treated and control plants under salinity stress in the Winter Dawn variety strawberry (Table 4.3.11) The pooled analysis revealed that the highest proline content was recorded in runners treated with a 30 Gy dose exposed to 50 mM NaCl (3.82 $\mu\text{mol/gm}$), which was statistically at par with 20 Gy X 100 mM NaCl. On the other hand, the lowest proline content was recorded in plants exposed to 75 mM

Table 4.3.10 Interaction effect of gamma irradiation doses under different levels of salinity on leaf area and malonaldehyde content of strawberry cv. Winter Dawn

Treatment	Leaf area 2023	Leaf area 2024	Leaf area pooled data	Malondi aldehyd e 2023	Malond ialdehy de 2024	Malond ialdehy de pooled Data	Treatment	Leaf area 2023	Leaf area 2024	Leaf area pooled	Malo ndiald ehyde 2023	Malo ndiald ehyde 2024	Malond ialdehy de pooled data	
20 Gy X Control	19.63	20.10	19.86	4.57	4.07	4.32	Control	18.49	19.78	19.17	4.57	2.73	3.65	
20 Gy X 50 mM NaCl	21.46	20.33	20.89	5.62	3.22	4.42	Control X 50 mM NaCl	16.55	10.78	13.66	8.36	9.26	8.81	
20 Gy X 75 mM NaCl	20.79	21.32	21.05	3.18	1.95	2.57	Control X 75 mM NaCl	12.55	15.21	13.88	9.49	8.93	9.21	
20 Gy X 100 mM NaCl	19.42	20.11	19.76	2.21	1.89	2.05	Control X 100 mM NaCl	14.56	12.39	13.47	9.35	10.6	9.99	
30 Gy X Control	18.79	19.36	19.07	4.29	2.15	3.22	C.D. for Years	0.67	0.23					
30 Gy X 50 mM NaCl	19.67	20.23	19.95	3.35	2.57	2.96	C.D. for Treatment	5.43	3.10					
30 Gy X 75 mM NaCl	23.49	19.10	21.29	1.95	2.22	2.08	C.D. for Years X Treatments	1.79	1.20					
30 Gy X 100 mM NaCl	22.67	20.24	21.45	2.41	1.84	2.12								
40 Gy X Control	16.79	18.12	17.45	4.04	6.19	5.12								
40 Gy X 50 mM NaCl	13.78	17.39	15.58	9.36	6.02	7.69								
40 Gy X 75 mM NaCl	15.37	14.22	14.89	9.96	6.89	8.42								
40 Gy X 100 mM NaCl	17.69	18.20	17.94	9.89	7.033	8.46								

Table 4.3.11 Interaction effect of gamma irradiation doses under different levels of salinity on electrolyte leakage and proline content of strawberry cv. Winter Dawn

Treatment	E L % 2023	E L % 2024	E L % pooled data	Proline content 2023	Proline content 2024	Proline content pooled data	Treatment	E L % 2023	E L % 2024	E L % pooled data	Proline content 2023	Proline content 2024	Proline content pooled data
20 Gy X Control	30.22	28.43	29.32	2.97	4.22	3.59	Control	30.69	33.71	32.20	2.64	3.33	2.98
20 Gy X 50 mM NaCl	25.50	24.29	24.89	3.40	3.40	3.40	Control X 50 mM NaCl	29.11	35.08	32.19	2.93	2.95	2.94
20 Gy X 75 mM NaCl	30.03	26.75	28.39	2.84	2.84	2.84	Control X 75 mM NaCl	29.65	38.49	34.17	2.34	2.74	2.54
20 Gy X 100 mM NaCl	29.51	25.30	27.41	3.84	3.54	3.69	Control X 100 mM NaCl	28.99	36.50	32.74	2.60	2.60	2.60
30 Gy X Control	26.78	25.49	26.13	3.12	3.67	3.40	C.D. for Years	1.17	0.23				
30 Gy X 50 mM NaCl	26.55	22.56	24.56	3.32	4.33	3.82	C.D. for Treatment	10.73	0.79				
30 Gy X 75 mM NaCl	27.06	34.70	30.88	3.35	3.76	3.56	C.D. for Years X Treatments	2.74	1.07				
30 Gy X 100 mM NaCl	26.55	27.83	27.19	3	3.40	3.20							
40 Gy X Control	28.68	32.49	30.59	2.96	3.44	3.20							
40 Gy X 50 mM NaCl	26.42	35.10	30.76	2.91	3.07	2.99							
40 Gy X 75 mM NaCl	26.57	35.34	30.95	3.11	3.91	3.51							
40 Gy X 100 mM NaCl	26.19	28.58	27.38	2.74	2.95	2.85							

salinity condition (2.54 $\mu\text{mol/g}$). In 2024, a similar trend was observed, with treated runners exhibiting the highest proline content (4.33 $\mu\text{mol/g}$) under 50 mM salinity stress. Conversely, the lowest proline content was recorded in runners exposed to the highest salinity stress of 100 mM (2.60 $\mu\text{mol/g}$). In 2023, the maximum proline content was recorded in plants treated with a 20 Gy dose induced with 100 mM NaCl (3.84 $\mu\text{mol/gm}$), while the minimum was observed in runners exposed against to 75 mM NaCl (2.34 $\mu\text{mol/gm}$), further highlighting the effectiveness of gamma radiation, particularly at 20 Gy and 30 Gy doses, in enhancing proline accumulation under salinity stress conditions.

4.3.23 Membrane stability index

The results from the study indicate that physical mutagen had a significant impact on the membrane stability index (MSI), with notable differences observed between treated and control plants under salinity stress in winter variety of strawberry (Table 4.3.12). The pooled analysis revealed that the highest MSI was observed in plants treated with a 20 Gy dose exposed to 50 mM NaCl (80.99 %), while the lowest MSI was recorded in plants exposed to 100 mM NaCl (74.4 %). However, in 2023, the maximum MSI (84.53 %) was recorded in the combination of 30 Gy mutagen dose against 50 mM NaCl, while the minimum was found in runners under 100 mM NaCl (68.32 %). A similar trend was recorded during 2024 treated runners showed maximum membrane stability index under 75 mM NaCl (80.95 %) and minimum membrane stability index was recorded in runners induce with 75 mM salinity stress (69.61 %). These findings highlight the effectiveness of gamma radiation, particularly at 20 Gy and 30 Gy doses, in enhancing membrane stability under salinity stress conditions instead of 40 Gy and control runners.

4.3.24 Leaf scorching

It is evident from the data that leaf scorching in the Winter Dawn strawberry variety was significantly affected by salinity levels represented in (Table 4.3.12) and Illustrate in Figure 4.

The results indicate that the physical mutagen significantly reduced leaf scorching in strawberry plants under salinity stress conditions. The pooled analysis revealed that the highest leaf scorching was observed in plants exposed to 100 mM NaCl (22.98 %), while the lowest leaf scorching was recorded in plants treated with 30 Gy of gamma radiation against with 75 mM NaCl (13.94%). A similar trend was observed in both 2023 and 2024, where salinity stress caused the highest leaf scorching in salinity induced plants. These findings demonstrate

Table 4.3.12 Interaction effect of gamma irradiation doses under different levels of salinity on membrane stability index and leaf scorching of strawberry cv. Winter Dawn

Treatment	MSI 2023	MSI 2024	MSI pooled data	Leaf scorchin g 2023	Leaf scorchi ng 2024	Leaf scorchi ng pooled data	Treatment	MSI 2023	MSI 2024	MSI pooled data	Leaf scorchi ng 2023	Leaf scorchi ng 2024	Leaf scorchi ng pooled data
20 Gy X Control	80.41	78.50	79.45	17.66	14.48	16.11	Control	82.6	69.71	75.88	17.7	11.88	14.79
20 Gy X 50 mM NaCl	82.48	79.50	80.99	14.17	17.55	15.86	Control X 50 mM NaCl	83.84	71.77	77.80	21.49	20.76	21.12
20 Gy X 75 mM NaCl	81	75.41	78.20	15.84	12.86	14.35	Control X 75 mM NaCl	81.53	69.61	75.57	22.56	23.19	22.87
20 Gy X 100 mM NaCl	80.40	77.74	79.07	17.68	11.88	14.78	Control X 100 mM NaCl	68.32	80.49	74.41	24.63	21.33	22.98
30 Gy X Control	80.96	74.85	77.91	17.83	15.42	16.63	C.D. for Years	0.71	1.04				
30 Gy X 50 mM NaCl	84.53	71.91	78.22	13.67	17.31	15.49	C.D. for Treatment	7.01	13.08				
30 Gy X 75 mM NaCl	73.45	80.95	77.70	13.51	13.48	13.49	C.D. for Years X Treatments	2.43	2.04				
30 Gy X 100 mM NaCl	80.48	73.86	77.17	27.40	12.88	20.14							
40 Gy X Control	79.98	71.17	75.57	29.47	11.75	20.61							
40 Gy X 50 mM NaCl	82.90	75.29	79.10	20.30	12.26	16.28							
40 Gy X 75 mM NaCl	82.14	70.83	76.48	21.81	10.93	16.37							
40 Gy X 100 mM NaCl	80.61	70.17	75.39	20.65	15.95	18.30							

the effectiveness of gamma radiation, particularly at a 30 Gy dose, in mitigating leaf scorching under salinity stress conditions.



Plate 9. Showed leaf scorching effect on control runners plant as compare to gamma irradiation treated

4.3.25 Mineral analysis sodium Na^+

The (Table 4.3.13) presents the concentration of sodium Na in gamma irradiation treated strawberry plant against induced salinity NaCl concentration. In sodium analysis, it was observed that the plant treated with 20 Gy and 30 Gy mutagen doses showed minimum sodium instead of 40 Gy and control runners.

As well as in pooled results the maximum sodium content was recorded in runners induced with 100 mM NaCl (99.45 ppm) and minimum sodium content was recorded in runners treated with 30 Gy mutagen dose exposed with 50 mM NaCl (24.51 ppm). A similar trend was recorded during 2024 sodium was recorded maximum in runners against 50 mM NaCl (99.10

ppm) and minimum sodium was recorded in gamma irradiation treated runners against 50 mM NaCl (25.43 ppm).

However, in 2023 the sodium was maximum recorded in plant against 75 mM NaCl (101.29 ppm) and minimum sodium recorded in runners treated with 20 Gy physical mutagen dose under 0 mM salinity condition (23.39 ppm).

4.3.26 Mineral analysis potassium K⁺

The potassium was estimated from leaves of gamma irradiation treated runners against induced salinity NaCl concentration added plants to check their tolerance against salinity (Table 4.3.13). In potassium analysis, it was observed that the plant treated with 20 Gy and 30 Gy mutagen doses showed maximum potassium ion as compared to 40 Gy and control runners showed minimum potassium ion.

In pooled results maximum potassium was recorded in runners treated with 30 Gy gamma irradiation exposed with 50 mM NaCl treatment (27.76 ppm) and minimum potassium was found in runners against 100 mM NaCl (12.15 ppm). A similar trend was during 2023 potassium was observed maximum in mutagen treated runners against 50 mM NaCl (26.37 ppm) and minimum recorded in runners against 75 mM NaCl (11.69 ppm).

However, in 2024 potassium was found maximum in runners treated with 20 Gy dose induced with 75 mM NaCl (25.43 ppm) and minimum potassium was recorded in runners under 50 mM NaCl (10.64 ppm).

Table 4.3.13 Interaction effect of gamma irradiation doses under different levels of salinity on sodium content and potassium ion of strawberry cv. Winter Dawn

Treatment	Sodium Na ppm 2023	Sodium Na ppm 2024	Sodium Na ppm pooled data	Potassium K ppm 2023	Potassium K ppm 2024	Potassium K ppm pooled data	Treatment	Sodium Na ppm 2023	Sodium Na ppm 2024	Sodium Na ppm pooled data	Potassium K ppm 2023	Potassium K ppm 2024	Potassium K ppm pooled data				
20 Gy X Control	23.39	27.86	25.62	22.76	23.43	23.09	Control	29.64	31.12	30.38	15.78	14.39	15.08				
20 Gy X 50 mM NaCl	50.68	49.67	50.17	16.37	17.46	16.91	Control X 50 mM NaCl	78.39	99.10	88.74	18.67	10.64	14.66				
20 Gy X 75 mM NaCl	66.49	69.32	67.90	29.63	30.17	29.9	Control X 75 mM NaCl	101.29	93.11	97.2	11.69	12.63	12.11				
20 Gy X 100 mM NaCl	71.29	78.53	74.91	22.19	23.33	22.76	Control X 100 mM NaCl	100.2	98.7	99.45	12.67	11.64	12.15				
30 Gy X Control	24.91	26.23	25.27	21.37	22.29	21.83	C.D. for Years	2.67	1.79								
30 Gy X 50 mM NaCl	23.59	25.43	24.51	26.37	29.16	27.76	C.D. for Treatment	3.94	2.439								
30 Gy X 75 mM NaCl	78.39	81.22	79.80	27.56	26.19	26.87	C.D. for Years X Treatments	8.11	1.73								
30 Gy X 100 mM NaCl	52.37	53.44	52.90	18.76	16.59	17.67											
40 Gy X Control	34.23	36.37	35.3	19.24	20.33	19.78											
40 Gy X 50 mM NaCl	71.52	73.44	72.48	21.39	22.67	22.03											
40 Gy X 75 mM NaCl	84.33	86.23	85.28	15.33	14.67	15											
40 Gy X 100 mM NaCl	61.33	63.77	62.7	14.62	12.49	13.55											

4.4 Interaction effect of ethyl methane sulphonate doses under varying levels of salinity in the Winter Dawn strawberry variety

4.4.1 Plant height

The analysis of the data reveals that different EMS doses combined with varying levels of salinity significantly influenced the plant height of Winter Dawn strawberry plants (Table 4.4.1). The pooled data shows that the maximum plant height was recorded in runners treated with 2% EMS induced with 50 mM NaCl (24.82 cm), which was statistically at par with 1 % EMS dose. While the minimum plant height was observed in runners exposed under 100 mM salinity condition (20.15 cm). A similar trend was recorded during 2023 treated runners exposed with 75 mM NaCl shows the maximum plant height (20.51 cm), However minimum plant height was recorded runners exposed with 75 mM salinity condition (20.51 cm). However also in 2024 similar trend was recorded treated runners showed maximum plant height (23.44 cm), while the minimum height of was noted in runners exposed to 50 mM salinity condition (17.93 cm). In this result, it was observed that the plant treated with a 2% EMS chemical mutagen dose had greater tolerance to salinity and was not affected in terms of plant height.

4.4.2 Chlorophyll content index

It is evident from the (Table 4.4.1) that the chlorophyll content index was significantly lower in non-mutagen-treated plants compared to those treated with a 3% EMS dose in the Winter Dawn strawberry variety. In the pooled analysis, the maximum chlorophyll content index value was recorded in runners treated with 2% EMS dose against 75 mM NaCl (55.74), which was statistically at par with 1% EMS dose. The minimum value was observed in runners subjected to the highest salinity condition concentration of 100 mM (51.8). A similar trend was recorded during 2023 when chemical mutagen treated runners showed a maximum chlorophyll under 100 mM salinity conditions (62.42) and minimum chlorophyll was recorded in runners exposed with 75 mM NaCl (46.17). A similar trend was recorded during 2024.

4.4.3 Number of buds

It is apparent from the data presented in (Table 4.4.2) that chemical mutagen treatment against induced salinity NaCl stress significantly influenced bud formation in winter Dawn strawberry plants.

Table 4.4.1 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on plant height and chlorophyll of strawberry cv. Winter Dawn

Treatment	Plant height 2023	Plant height 2024	Plant height pooled Data	Chlorophyll 2023	Chlorophyll 2024	Chlorophyll pooled Data	Treatment	Plant height 2023	Plant height 2024	Plant height pooled Data	Chlorophyll 2023	Chlorophyll 2024	Chlorophyll pooled Data				
1% EMS X Control	23.66	21.52	22.59	60.83	49.91	55.41	Control	23.85	18.5	21.17	56.4	47.39	51.89				
1% EMS X 50 mM NaCl	25.7	21.48	23.59	59.8	46.81	53.30	Control X 50 mM NaCl	22.61	17.93	20.27	56.8	46.28	51.54				
1% EMS X 75 mM NaCl	24.12	19.53	21.82	57.13	47.9	52.9	Control X 75 mM NaCl	20.51	20.87	20.69	59	46.17	52.58				
1% EMS X 100 mM NaCl	22.74	19.47	21.10	56.56	47.64	52.10	Control X 100 mM NaCl	21.71	18.58	20.15	55.33	48.26	51.8				
2% EMS X Control	23.65	21.38	22.52	55.5	48.92	52.21	C.D. for Years	1.08	0.93								
2% EMS X 50 mM NaCl	26.19	23.44	24.82	54.36	52.41	53.39	C.D. for Treatment	2.57	5.086								
2% EMS X 75 mM NaCl	26.40	18.78	22.59	61.93	49.55	55.74	C.D. for Years X Treatments	2.03	3.23								
2% EMS X 100 mM NaCl	24.5	17.50	21.4	62.46	45.88	54.17											
3% EMS X Control	23.82	19.95	21.88	60.46	47.7	54.12											
3% EMS X 50 mM NaCl	24.49	20.94	22.71	60.93	48.77	54.85											
3% EMS X 75 mM NaCl	23.87	19.37	21.62	59.3	51.68	55.49											
3% EMS X 100 mM NaCl	24.58	18.51	21.54	58.7	46.49	52.597											

The pooled analysis revealed that the maximum number of buds was observed in plants treated with a combination of 2% EMS induced with 50 mM salinity level (26.16), while the minimum was recorded in runners exposed to 100 mM salinity condition (20.5). A similar trend was recorded during 2023 the mutagen treated runners shows highest number of buds was recorded under 75 mM salinity level (28.33), with the minimum occurring in the without mutagen treated runners against to the same salinity level (20.33). In 2024, a maximum of buds was recorded in plants treated with 1% EMS under 50 mM NaCl (24.33), whereas the minimum number of buds was observed in the runners exposed to 100 mM NaCl (18.67). The findings indicate that chemical mutagen treatment, particularly with 2% EMS dose against NaCl salinity levels to 100 mM NaCl significantly enhances bud formation in Winter Dawn strawberry plants under induced salinity stress. The data show that treated runners with 2% EMS dose consistently produced a higher number of buds compared to controls, shows the potential of EMS treatment in improving bud development.

4.4.4 Number of flowers

It is evident from the (Table 4.4.2) that the number of flowers was significantly affected in 2% EMS dose and control plants compared to those treated with 1% EMS and 2% EMS in the Winter Dawn strawberry variety. The pooled results of chemical mutagen EMS treated plants, subjected to varying levels of salinity, show a maximum flower count was recorded in plants treated with 2% EMS dose exposed to 50 mM NaCl (27.22). Which was statistically at par with 1% EMS dose. However, the minimum number of flowers was recorded in plants exposed to 100 mM NaCl (20.5). A similar trend was recorded during 2023, in treated runners the maximum number of flower was recorded under 75 mM salinity level (31), while the minimum was recorded in without mutagen treated runners against to 75 mM NaCl (20.26). In 2024, the maximum flower recorded was observed in the combination of 1% EMS exposed to 50 mM NaCl (25.67), However, the minimum flower count of was recorded in runners exposed to 50 mM salinity level (20).

4.4.5 Number of fruit

It is apparent from the data presented in (Table 4.4.3) that chemical mutagen treatment significantly influenced the number of fruits in Winter Dawn strawberry plants subjected to induced salinity NaCl stress. In both years of the experiment, the maximum average number of fruits recorded

Table 4.4.2 Interaction effect of ethyl methane sulfonate doses under different levels of salinity on plant height and chlorophyll of strawberry cv. Winter Dawn

Treatment	Number of bud 2023	Number of bud 2024	Number of bud pooled data	Number of flowers 2023	Number of flowers 2024	Number of flowers pooled data	Treatment	Number of bud 2023	Number of bud 2024	Number of bud pooled data	Number of flowers 2023	Number of flowers 2024	Number of flowers pooled data
1% EMS X Control	25	23.66	24.33	25.66	24.67	25.16	Control	23	21	22	23.30	20.46	24.83
1% EMS X 50 mM NaCl	27	24.33	25.66	28.66	25.67	27.16	Control X 50 mM NaCl	24.67	22.33	23.5	27.66	20	23.83
1% EMS X 75 mM NaCl	25.33	22.33	23.83	27.33	21	24.16	Control X 75 mM NaCl	20.33	21.67	21	20.26	21	20.63
1% EMS X 100 mM NaCl	26.66	20	23.33	28	19.33	23.66	Control X 100 mM NaCl	21.33	18.67	20.5	21.14	20.21	20.67
2% EMS X Control	26	23	24.5	27.66	21.17	24.33	C.D. for Years	0.88	0.92				
2% EMS X 50 mM NaCl	27.33	25	26.16	30.33	25.11	27.72	C.D. for Treatment	5.17	3.05				
2% EMS X 75 mM NaCl	28.33	18	23.16	31	20.87	25.93	C.D. for Years X Treatments	3.08	3.05				
2% EMS X 100 mM NaCl	25	20	22.5	27.33	20.33	23.83							
3% EMS X Control	25.66	18	21.83	29.33	19.67	24.5							
3% EMS X 50 mM NaCl	25.66	21	23.33	30.9	20.67	25.78							
3% EMS X 75 mM NaCl	25.66	17.33	21.5	26	20	23							
3% EMS X 100 mM NaCl	26	17.667	21.833	27.67	20	22.83							

was observed in plants treated with a 2% EMS dose against 50 mM NaCl (29) Which was statistically at par with 1% EMS dose. While the minimum number of fruit was found in those exposed to 75 mM salinity level (21.82). A similar trend was recorded during 2023 mutagen treated runners showed maximum fruits under 75 mM salinity level (32.33) and minimum fruit number recorded in 2% EMS dose under 0 mM salinity condition. During 2024, the maximum number of buds was found in plants treated with a 1% EMS dose (27). However, the minimum was recorded in those subjected to 100 mM salinity level (19.33). These observations indicate that plants treated with 2% EMS doses demonstrate greater tolerance to induced salinity stress and exhibit higher buds.

4.4.6 Days to buds

It is evident from the pooled data that bud initiation was significantly influenced by the interaction of various EMS doses and different salinity levels in winter Dawn strawberry plants (Table 4.4.3). The combined results show that early bud initiation was recorded in plants treated with a 2% EMS dose exposed to 50 mM NaCl (30.88), while late bud initiation occurred in runners exposed to 100 mM NaCl (50.83). A similar trend was recorded during 2023 the treated runners showed early bud initiation under 100 mM salinity conditions (32.10). However late bud initiation was recorded in runners exposed to 100 mM salinity conditions (49.10). A similar trend was observed during 2024.

4.4.7 Days to flowers

The pooled data clearly indicates that flower initiation in Winter Dawn strawberry plants was significantly impacted by the interaction between varying EMS doses induced with different salinity levels on winter Dawn variety strawberry (Table 4.4.4). The combined results reveal that the earliest flower initiation has occurred in runners treated with a 2% EMS dose under 50 mM NaCl (54.78), while the late initiation was recorded in runners under 100 mM salinity level (63.66). A similar trend was observed during 2023 the mutagen-treated runners induced with 100 mM salinity conditions showed early flower initiation was observed at (56). While runners under the same salinity level showed late initiation at (73) days. Similar trend was observed during 2024.

4.4.8 Days to fruit set

It is evident from the pooled data that days to the first fruit set were significantly influenced by the interaction of various EMS doses and different salinity levels in Winter Dawn strawberry

Table 4.4.3 Interaction effect of ethyl methane sulfonate doses under different levels of salinity on number of fruits and days to bud of strawberry cv. Winter Dawn

Treatment	No. of fruits 2023	No. of fruits 2024	No. of fruits pooled data	Days to bud 2023	Days to bud 2024	Days to bud pooled data	Treatment	No. of fruits 2023	No. of fruits 2024	No. of fruits pooled data	Days to bud 2023	Days to bud 2024	Days to bud pooled data				
1% EMS X Control	26.66	27	26.83	33.29	32.46	32.87	Control	30.33	20.33	25.33	39.23	38.22	38.72				
1% EMS X 50 mM NaCl	30.66	21.33	26	34.29	33.19	33.74	Control X 50 mM NaCl	29	24	26.5	45.47	46.22	45.84				
1% EMS X 75 mM NaCl	28	20	24	38.23	37.63	37.93	Control X 75 mM NaCl	21.64	22	21.82	47.23	51.27	49.25				
1% EMS X 100 mM NaCl	29.33	19	24.16	35.42	34.22	34.82	Control X 100 mM NaCl	29	19.33	24.16	49.10	52	50.83				
2% EMS X Control	32.33	22.66	27.5	40.10	29.37	34.73	C.D. for Years	0.92	0.60								
2% EMS X 50 mM NaCl	31.66	26.33	29	32.19	29.57	30.88	C.D. for Treatment	3.05	2.32								
2% EMS X 75 mM NaCl	30.66	21	25.83	39.11	28.46	33.78	C.D. for Years X Treatments	3.05	2.90								
2% EMS X 100 mM NaCl	28	24.66	26.33	32.10	33.23	32.21											
3% EMS X Control	30	22.33	26.16	39.18	40.33	39.75											
3% EMS X 50 mM NaCl	30.66	22	26.33	40.56	41.32	40.94											
3% EMS X 75 mM NaCl	30	19.33	24.66	42.68	41.29	41.98											
3% EMS X 100 mM NaCl	29	19	24	41.63	40.79	41.21											

Table 4.4.4 Interaction effect of ethyl methane sulfonate doses under different levels of salinity on days to flower and fruits of strawberry cv. Winter Dawn

Treatment	Days to flowers 2023	Days to flowers 2024	Days to flower pooled data	Days to fruits 2023	Days to fruits 2024	Days to fruit pooled data	Treatment	Days to flowers 2023	Days to flowers 2024	Days to flower pooled data	Days to fruits 2023	Days to fruits 2024	Days to fruit pooled data
1% EMS X Control	67	59.33	63.16	82.33	75.33	78.83	Control	61	60	60.5	88.66	88.66	88.66
1% EMS X 50 mM NaCl	61	52	56.5	84.66	75.66	80.16	Control X 50 mM NaCl	61	60.33	60.66	87.33	88.33	87.83
1% EMS X 75 mM NaCl	61	57	59	89.33	78	83.66	Control X 75 mM NaCl	62	61.66	61.83	82.33	90.33	86.33
1% EMS X 100 mM NaCl	58.33	53.33	55.83	90	76.33	82.5	Control X 100 mM NaCl	73	54.33	63.66	89.33	89	89.16
2% EMS X Control	60.33	58	59.16	89.33	76.33	82.83	C.D. for Years	1.68	3.52				
2% EMS X 50 mM NaCl	56.38	53.19	54.78	91.66	75	84	C.D. for Treatment	9.58	16.72				
2% EMS X 75 mM NaCl	61.33	51.66	56.5	78.33	71	74.66	C.D. for Years X Treatments	4.44	9.39				
2% EMS X 100 mM NaCl	56	55.33	58.33	78	90	84							
3% EMS X Control	60.33	58.33	59.33	86.33	87.33	86.83							
3% EMS X 50 mM NaCl	60.33	59	59.66	87	86	86.5							
3% EMS X 75 mM NaCl	62	64.66	63.33	79.33	88.33	83.83							
3% EMS X 100 mM NaCl	59.333	55.667	57.8	85.667	89	87.333							

plants (Table 4.4.4) The combined results show that early fruit set was recorded in treated runners with a 2% EMS dose exposed to 50 mM salinity level (74.66), while late fruit set occurred in runners exposed to 100 mM NaCl (89.16).

A similar trend was recorded during 2023 treated runners shows early fruit set under the 100 mM salinity level (78) and late bud initiation was found when plant induced with same salinity level (89.33). However, during 2024 similar trend was recorded.

4.4.9 Fruit length

The analysis of the data reveals that the combined effect of different EMS doses and varying salinity levels significantly influenced the fruit length of winter Dawn strawberry plants (Table 4.4.5). The pooled data shows that the maximum fruit length was recorded in plants treated with 2% EMS dose under 75 mM NaCl (89.46 mm) which was statistically at par with 1% EMS dose. However minimum fruit length found in plants exposed to 100 mM NaCl (74.66 mm).

In 2023 the runners treated with 1% EMS dose showed maximum fruit length was recorded under 50 mM salinity level length (91.66 mm), and minimum fruit length was recorded in plants exposed to 100 mM salinity level (71 mm).

However similar trend was observed during 2024 the treated runners showed maximum fruit length under the 100 mM NaCl salinity level (90.33 mm) and length was found in runners induced with 100 mM salinity condition (74.66 mm).

4.4.10 Fruit diameter

The data analysis indicates that the interaction between different EMS concentrations induced with varying levels of NaCl salinity level significantly affected the fruit diameter of the Winter Dawn variety of strawberry plants (Table 4.4.5) The pooled data reveals that the maximum fruit diameter was recorded in the combination of 2% EMS induced with 75 mM salinity condition (30.49 mm), which was statistically at par with the 2% EMS dose. Conversely, the minimum diameter was observed in runners under a 100 mM salinity level (24.16 mm).

A similar trend was noted in 2024, where treated runners showed a maximum fruit diameter found under 75 mM salinity (29.85 mm), while the minimum fruit diameter was recorded in runners exposed to 100 mM salinity (19.62 mm).

Table 4.4.5 Interaction effect of ethyl methane sulfonate doses under different levels of salinity on fruit length and diameter of strawberry cv. Winter

Treatment	Fruit length 2023	Fruit length 2024	Fruit length pooled data	Fruit diameter 2023	Fruit diameter 2024	Fruit diameter pooled data	Treatment	Fruit length 2023	Fruit length 2024	Fruit length pooled data	Fruit diameter 2023	Fruit diameter 2024	Fruit diameter pooled data
1% EMS X Control	90	75	82.5	31.50	24.62	28.11	Control	84.66	75.66	80.16	29.51	20.77	25.14
1% EMS X 50 mM NaCl	88.66	88.66	88.66	32.43	22.48	27.45	Control X 50 mM NaCl	76.3	79.21	77.78	33.17	21.57	27.37
1% EMS X 75 mM NaCl	89.33	78	83.66	34.98	23.43	29.21	Control X 75 mM NaCl	82.33	75.33	78.83	29.12	20.35	24.73
1% EMS X 100 mM NaCl	82.31	80.56	81.43	29.81	22.11	25.96	Control X 100 mM NaCl	71	78.33	74.66	30.7	19.62	24.16
2% EMS X Control	89.33	76.33	82.83	30.26	25.2	27.73	C.D. for Years	3.52	1.18				
2% EMS X 50 mM NaCl	91.66	76.33	84	30.5	25.81	28.15	C.D. for Treatment	16.72	3.60				
2% EMS X 75 mM NaCl	89.33	89	89.16	31.13	29.85	30.49	C.D. for Years X Treatments	8.49	2.38				
2% EMS X 100 mM NaCl	82.33	90.33	86.33	29.90	21.99	25.95							
3% EMS X Control	87.33	88.33	87.83	29.36	21.44	25.4							
3% EMS X 50 mM NaCl	80.63	81.19	80	30.37	21.60	25.99							
3% EMS X 75 mM NaCl	79.33	88.33	83.83	30.38	20.26	25.32							
3% EMS X 100 mM NaCl	82.23	81.34	81.78	30.32	20.54	25.43							

However, during 2023 where treated runners with a 1 % EMS dose showed a maximum fruit diameter of with a combination of 1% EMS induced with 75 mM salinity condition (34.98 mm) However, the minimum fruit diameter was found in runners exposed to a 75 mM salinity level (29.12 mm). These findings indicate that a specific dose of 2% EMS can enhance fruit development in strawberry plants under salinity condition.

4.4.11 Fruit weight

The (Table.4.4.6) clearly demonstrates the impact of EMS treatment combined with NaCl-induced salinity stress on the fruit weight of runners from the Winter Dawn strawberry variety. In the pooled analysis, the highest fruit weight was recorded in runners treated with a 2% EMS dose under 50 mM NaCl stress (12.32 gm) which was statistically at par with 1% EMS dose, whereas the lowest fruit weight was recorded in runners induced with 100 mM NaCl salinity level (9.29 gm). A similar trend was observed in 2023 mutagen treated showed maximum fruit weight under 75 mM NaCl (13.10 gm), While the minimum was found in runners induced with 50 mM NaCl (9.75 gm).

However, in 2024, the highest fruit weight was recorded in runners treated with a 1% EMS dose under 100 mM NaCl (12.50 gm), and the lowest fruit weight was recorded in runners exposed to 100 mM salinity condition (8.12 gm).

4.4.12 Fruit yield

It is evident from the data that fruit yield in the Winter Dawn strawberry variety was significantly affected by different EMS chemical mutagen doses and salinity levels (Table 4.4.6). The pooled data values show the highest fruit yield in runners treated with a 2% EMS dose and exposed to 50 mM NaCl stress (350.66 gm) Which was statistically at par with 1% EMS dose. followed by a minimum yield recorded in runners exposed to 100 mM salinity condition (256.66 gm).

A similar trend was recorded in 2024 the treated runners showed maximum fruit yield found under the 100 mM salinity condition (351.67 gm) and minimum fruit yield recorded in runners exposed to 75 mM salinity condition (192.66 gm).

However, in 2023, the maximum fruit yield was observed in runners treated with a 1% EMS dose and exposed to 50 mM salinity level (376.67 gm) and the minimum yield was recorded in runners exposed to 50 mM salinity level (289.33 gm).

Table 4.4.6 Interaction effect of ethyl methane sulfonate doses under different levels of salinity on fruits weight and fruit yield of strawberry cv. Winter Dawn

Treatment	Fruit weight 2023	Fruit weight 2024	Fruit weight pooled data	Fruit yield 2023	Fruit yield 2024	Fruit yield pooled data	Treatment	Fruit weight 2023	Fruit weight 2024	Fruit weight pooled data	Fruit yield 2023	Fruit pooled 2024	Fruit yield pooled data				
1% EMS X Control	12.03	9.88	10.95	358.67	284.66	321.66	Control	11.54	8.40	9.97	303.33	247.3	275.33				
1% EMS X 50 mM NaCl	11.4	10.44	10.92	376.67	318.33	347.5	Control X 50 mM NaCl	9.75	9.86	9.80	289.33	270	279.66				
1% EMS X 75 mM NaCl	11.69	10.48	11.08	358.67	225.33	292	Control X 75 mM NaCl	10.65	8.12	9.38	343.66	192.66	268.16				
1% EMS X 100 mM NaCl	10.56	12.50	11.53	324.67	272.33	298.5	Control X 100 mM NaCl	10.36	8.22	9.29	292	221.33	256.66				
2% EMS X Control	12.17	10.68	11.43	351.67	291	321.33	C.D. for Years	0.33	1.53								
2% EMS X 50 mM NaCl	12.87	11.77	12.32	391	310.33	350.66	C.D. for Treatment	2.52	7.47								
2% EMS X 75 mM NaCl	13.10	9.54	11.32	372.67	286.66	329.66	C.D. for Years X Treatments	1.55	6.45								
2% EMS X 100 mM NaCl	10.46	10.51	10.48	300.33	351.67	326											
3% EMS X Control	11.56	9.06	10.31	304.33	257.33	280.83											
3% EMS X 50 mM NaCl	11.9	9.56	10.73	365.67	271.33	318.5											
3% EMS X 75 mM NaCl	11.47	8.25	9.86	376.67	225.66	301.16											
3% EMS X 100 mM NaCl	12.66	7.41	10.03	371.33	288	329.67											

4.4.13 Shoot fresh weight

The interaction between various EMS doses and salinity levels significantly affected the salinity tolerance of Winter Dawn strawberry plants, specifically in terms of shoot fresh weight (Table 4.4.7). The pooled data indicated that the maximum shoot fresh weight was observed in runners treated with a 2% EMS dose under a 75 mM salinity condition (12.90 gm) which was statistically at par with 1 % EMS dose. However, the minimum shoot fresh weight was recorded in runners exposed to a 100 mM salinity level (8.38 gm). A similar trend was recorded 2024, the treated runners demonstrated a maximum shoot fresh weight under a 100 mM salinity condition (12.63 gm), with the minimum shoot fresh weight observed at the 100 mM salinity level (8.22 gm). During 2023 the 1% EMS dose treated runners showed a maximum shoot fresh weight under a 50 mM salinity condition (14.66 gm) and minimum shoot fresh weight was recorded in runners against 75 mM salinity condition (8.12).

4.4.14 Shoot dry weight

After measuring the shoot fresh weight, the shoot dry weight was recorded for both years in the Winter Dawn variety of strawberry (Table 4.4.7) In the pooled analysis, the maximum shoot dry weight was recorded in runners treated with a 2% EMS dose under 75 mM NaCl salinity (3.37 gm) which was statistically at par with the 1% EMS dose. Conversely, the minimum shoot dry weight was observed in runners exposed to the same salinity level (1.89 gm). A similar trend was recorded in 2024, where treated runners exhibited a maximum shoot dry weight under 75 mM salinity (2.96 gm), while the minimum was observed in plants exposed to 75 mM NaCl salinity (1.74 gm). However, in 2023, the maximum shoot dry weight was recorded in runners treated with 1% EMS dose induced with 75 mM salinity level (4.21 gm), while the minimum was observed in plants exposed to 75 mM NaCl salinity (1.10 gm).

4.4.15 Root fresh weight

The analysis of data on the interaction between chemical mutagen EMS doses and varying salinity levels reveals a significant impact on the root fresh weight of Winter Dawn strawberry plants (Table 4.4.8) The pooled results indicate that the highest root fresh weight was recorded in runners treated with a 2% EMS dose exposed with a 75 mM NaCl salinity condition (14.35 gm) which was statistically at par with 1 % EMS dose. While the lowest root fresh weight was observed in runners exposed to 100 mM salinity condition (9.63 gm) similar

Table 4.4.7 Interaction effect of ethyl methane sulfonate doses under different levels of salinity on shoot fresh weight and dry weight of strawberry cv. Winter Dawn

Treatment	Shoot fresh wt. 2023	Shoot fresh wt. 2024	Shoot fresh wt. pooled Data	Shoot dry wt. 2023	Shoot dry wt. 2024	Shoot dry wt. pooled Data	Treatment	Shoot fresh wt. 2023	Shoot fresh wt. 2024	Shoot fresh wt. pooled Data	Shoot dry wt. 2023	Shoot dry wt. 2024	Shoot dry wt. pooled data
1% EMS X Control	12.51	11.13	11.82	3.34	2.33	2.84	Control	10.69	9.84	10.26	3.32	2.24	2.78
1% EMS X 50 mM NaCl	11	10.10	10.55	3.25	1.76	2.50	Control X 50 mM NaCl	9.93	8.46	9.19	1.10	3.29	2.26
1% EMS X 75 mM NaCl	12.97	10.69	11.83	4.21	1.88	3.11	Control X 75 mM NaCl	8.19	9.73	8.96	3.14	1.74	2.44
1% EMS X 100 mM NaCl	12.85	8.48	10.62	2.81	1.84	2.33	Control X 100 mM NaCl	8.54	8.22	8.38	2.96	1.81	2.38
2% EMS X Control	11.93	10.55	11.24	3	1.80	2.43	C.D. for Years	0.68	0.19				
2% EMS X 50 mM NaCl	14.66	11.79	12.23	3.25	2.28	2.76	C.D. for Treatment	2.77	0.53				
2% EMS X 75 mM NaCl	13.41	12.38	12.90	3.78	2.96	3.37	C.D. for Years X Treatments	1.49	0.90				
2% EMS X 100 mM NaCl	10.12	12.63	11.37	2.94	1.67	2.30							
3% EMS X Control	12.58	9.12	10.85	3.1	2.14	2.62							
3% EMS X 50 mM NaCl	13.44	9.56	11.5	3.15	2.26	2.71							
3% EMS X 75 mM NaCl	10.35	10.65	10.50	3.14	1.91	2.53							
3% EMS X 100 mM NaCl	10.73	8.23	9.48	2.81	1.97	2.39							

trend was recorded in 2023, where the maximum root fresh weight was found in runners treated with a 2% EMS dose induced with a 100 mM NaCl salinity level (14.80 gm), contrasting with the lowest root fresh weight was observed in runners exposed to 75 mM NaCl (11.72 gm).

However, in 2024, the maximum root fresh weight was recorded in runners treated with a 1% EMS dose exposed with a 50 mM NaCl condition (14.83 gm), whereas the minimum root fresh weight was observed in runners under a 100 mM NaCl condition (8.98 gm). These results highlight the significant effects of 2% EMS dose treatments on enhancing salinity tolerance and increasing root fresh weight in the Winter Dawn strawberry variety.

4.4.16 Root dry weight

After measuring root fresh weight, root dry weight was recorded in the Winter Dawn strawberry variety over both years (Table 4.4.8) In the pooled analysis, the highest root dry weight was observed in runners treated with a 2% EMS dose and exposed to 75 mM NaCl (3.42 gm) which was statistically at par with 3% EMS dose. While the lowest root dry weight was recorded under 100 mM salinity condition (1.13 gm). A similar trend was observed in 2023, where EMS-treated runners showed a maximum root dry weight under the 50 mM salinity level (4.56 gm), and the minimum was recorded in runners exposed to 100 mM NaCl (1.6 gm). However, in 2024 maximum root dry weight was recorded in runners treated with 1 % EMS dose induced with 50 mM salinity level (2.85 gm) while the minimum root dry weight was found in runners under 75 mM salinity level (1.23 gm).

4.4.17 Number of leaves

The pooled two-year analysis demonstrates the combined effect of different doses of the chemical mutagen EMS and varying salinity levels on the number of leaves in the Winter Dawn Strawberry variety (Table 4.4.9) In the pooled analysis, the maximum number of leaves was recorded in runners treated with a 2% EMS dose under 50 mM NaCl (12.76) which was statistically at par with 3% EMS dose. In contrast, the minimum number of leaves was observed in runners exposed to 100 mM salinity (8.83). A similar trend was seen in 2023, where treated runners exhibited the maximum number of leaves under the same salinity level (13), and the minimum number of leaves was found under 75 mM salinity (8.33). This trend was consistent in 2024. These results indicate that the 2% EMS treatment was effective in enhancing leaf production and improving tolerance to salinity stress.

Table 4.4.8 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on root fresh weight and dry weight of strawberry cv. Winter Dawn

Treatment	Root fresh wt. 2023	Root fresh wt. 2024	Shoot fresh wt. pooled data	Root dry wt. 2023	Root dry wt. 2024	Root dry wt. pooled data	Treatment	Root fresh wt. 2023	Root fresh wt. 2024	Root fresh wt. pooled data	Root dry wt. 2023	Root dry wt. 2024	Root dry wt. pooled data
1% EMS X Control	13.22	11.87	12.54	3.88	2.24	3.12	Control	13.52	9.92	11.72	3.25	2.37	2.81
1% EMS X 50 mM NaCl	12.17	14.83	13.5	3.99	2.25	3.12	Control X 50 mM NaCl	13.86	9.62	10.74	3.23	1.23	1.23
1% EMS X 75 mM NaCl	12.29	13.49	12.89	3.77	2.16	2.97	Control X 75 mM NaCl	11.72	10.20	10.96	3.6	2.73	2.39
1% EMS X 100 mM NaCl	12.89	10.21	11.55	4.21	1.84	3.11	Control X 100 mM NaCl	12.28	8.98	9.63	1.6	2.11	1.13
2% EMS X Control	13.21	10.5	11.78	3.71	3.4	3.38	C.D. for Years	0.5	0.34				
2% EMS X 50 mM NaCl	14.10	12.34	13.22	3.26	2.85	3.12	C.D. for Treatment	2.50	1.33				
2% EMS X 75 mM NaCl	14.15	14.56	14.35	4.56	2.28	3.42	C.D. for Years X Treatments	1.36	0.94				
2% EMS X 100 mM NaCl	14.81	8.81	11.81	3.10	1.81	2.42							
3% EMS X Control	13.35	11.21	12.28	3.39	1.94	2.66							
3% EMS X 50 mM NaCl	13.17	10.29	11.73	2.66	2.12	2.39							
3% EMS X 75 mM NaCl	13.73	10.16	11.94	2.92	1.98	2.45							
3% EMS X 100 mM NaCl	12.27	9.97	11.12	2.20	1.99	1.59							

4.4.18 Leaf relative water content

The analysis of data on leaf relative water content (LRWC) in Winter Dawn strawberry plants treated with the chemical mutagen EMS and subjected to varying salinity levels reveals a significant impact (Table 4.4.9). The pooled results indicate that the highest LRWC was recorded in runners treated with a 2% EMS dose combined with 50 mM NaCl (80.96%), which was statistically at par with 1% EMS dose and the lowest LRWC was observed in runners exposed to 100 mM NaCl (73.21%). A similar trend was recorded in 2024, with treated runners showing a maximum LRWC of (79.21%) in runners treated with 2% EMS dose under 0 mM condition, and a minimum LRWC was found in runners under the 100 mM NaCl (71.56 %).

In 2023, the highest LRWC was found in plants treated with 1% EMS under 50 mM salinity condition (84.16%), contrasting with the lowest LRWC in plants exposed to 50 mM salinity level (73.44%). These results demonstrate the significant effects of 2% EMS treatments in enhancing salinity tolerance and maintaining leaf relative water content in strawberry plants.

4.4.19 Electrolyte leakage %

The analysis of data on electrolyte leakage in Winter Dawn strawberry plants treated with chemical mutagen EMS under NaCl salinity stress shows significant effects across both years (Table 4.4.10). The pooled results indicate that the minimum electrolyte leakage was recorded in runners treated with 2% EMS dose exposed with 100 mM NaCl (25.71%), while the maximum electrolyte leakage was observed in runners exposed to 100 mM NaCl (36.23%).

In 2024, a similar trend was observed, treated runners showed the lowest electrolyte leakage recorded in plants treated with 2% EMS under 100 mM salinity (21.90%), whereas the highest leakage was found in runners under 0 mM condition (36.78%).

However, in 2023, the lowest electrolyte leakage was observed in runners treated with 1% EMS under 50 mM NaCl (26.19%) and the highest electrolyte leakage value was recorded in runners exposed to 100 mM NaCl (36.23%). These results demonstrate that EMS treatments, particularly at 2%, effectively reduce electrolyte leakage and enhance salinity tolerance in Winter Dawn strawberry plants.

4.4.20 Proline content $\mu\text{mol/g}$

Table 4.4.9 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on number of leaves and leaf relative water content of strawberry cv. Winter Dawn

Treatment	No. of leaves 2023	No. of leaves 2024	No. of leaves pooled data	LRWC 2023	LRWC 2024	LRWC pooled data	Treatment	No. of leaves 2023	No. of leaves 2024	No. of leaves pooled data	LRWC 2023	LRWC 2024	LRWC pooled data
1% EMS X Control	11	10.33	10.66	80.13	78.46	79.27	Control	9	10	9.5	80.28	74.71	77.49
1% EMS X 50 mM NaCl	11.33	9.33	10.33	84.16	77.07	80.62	Control X 50 mM NaCl	10	8.67	9.33	73.44	81.55	77.5
1% EMS X 75 mM NaCl	11.66	10	10.83	81.11	77.46	79.28	Control X 75 mM NaCl	8.33	11.66	9.99	79.74	69.44	74.59
1% EMS X 100 mM NaCl	10.66	10.67	10.66	79.59	78.15	78.87	Control X 100 mM NaCl	10.66	7	8.83	74.87	71.56	73.21
2% EMS X Control	11.66	10	10.83	81.65	79.09	80.37	C.D. for Years	0.81	0.55				
2% EMS X 50 mM NaCl	12.37	13.16	12.76	83.52	78.41	80.96	C.D. for Treatment	3.04	5.16				
2% EMS X 75 mM NaCl	11.33	12	11.66	82.69	75.38	79.13	C.D. for Years X Treatments	2.87	3.70				
2% EMS X 100 mM NaCl	13	9.33	11.16	80.48	76.48	78.48							
3% EMS X Control	11	9	10	80.39	77.84	79.12							
3% EMS X 50 mM NaCl	11.33	9.67	10.5	80.52	78.90	79.71							
3% EMS X 75 mM NaCl	13	7.33	10.16	81.10	73.69	77.35							
3% EMS X 100 mM NaCl	10.33	8.33	9.33	80.38	74.81	77.59							

The analysis of data on the interaction between chemical mutagen EMS doses and varying salinity levels reveals a significant impact on the proline content of Winter Dawn strawberry plants (Table 4.4.10). The pooled results indicate that the maximum proline content was recorded in runners treated with a 1% EMS dose combined with 50 mM NaCl (4.22 $\mu\text{mol/g}$), while the minimum proline content was observed in runners exposed to 100 mM NaCl (2.48 $\mu\text{mol/g}$). A similar trend was observed in 2023, with EMS-treated runners showing the highest proline content under 0 mM condition (3.92 $\mu\text{mol/g}$) and the lowest proline content recorded in runners exposed to 50 mM NaCl (2.24 $\mu\text{mol/g}$). The trend continued in 2024. These results demonstrate the significant effects of 1% EMS treatment in enhancing salinity tolerance by increasing proline content in strawberry plants.

4.4.21 Leaf scorching %

The interaction effects between different doses of the chemical mutagen EMS and varying levels of salinity on the tolerance of Winter Dawn strawberry plants to salinity, specifically in terms of leaf scorching, was analysed in this study (Table 4.4.11). A review of the data reveals a significant influence of various EMS doses and application methods on leaf scorching in strawberries over both years of the investigation. In the pooled analysis, maximum leaf scorching was recorded under the 100 mM salinity condition (19.91%), whereas minimum leaf scorching was observed with the 2% EMS dose combined with 75 mM salinity level (12.17%).

This trend was consistent in 2023, where runners exhibited the highest leaf scorching under 50 mM salinity stress (20.76%), while EMS-treated runners with 2% EMS dose showed the lowest leaf scorching under 75 mM induced salinity stress (9.61%). A similar pattern was observed in 2024.

4.4.22 Membrane stability index %

The Membrane Stability Index (MSI) was assessed in Winter Dawn strawberry plants treated with the chemical mutagen EMS under induced salinity stress, as summarized in the accompanying (Table 4.4.11). In the pooled analysis, the highest MSI was observed in plants treated with a 2% EMS dose induced with 50 mM salinity condition (83.89%) which was statistically at par with 1% EMS dose. However, the lowest MSI was recorded in runners exposed to 100 mM salinity condition (74.82%). A similar pattern was noted in 2024, with a maximum MSI recorded in runners treated with a 2% EMS dose exposed to 75 mM salinity level.

Table 4.4.10 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on electrolyte leakage and proline content of strawberry cv. Winter Dawn

Treatment	E L % 2023	E L % 2024	E L % pooled data	Proline content 2023	Proline content 2024	Proline content pooled data	Treatment	E L % 2023	E L % 2024	E L % pooled data	Proline content 2023	Proline content 2024	Proline content pooled data
1% EMS X Control	30.22	28.80	29.51	3.92	3.62	3.77	Control	30.69	36.78	33.73	2.95	2.95	2.95
1% EMS X 50 mM NaCl	26.19	28.11	27.15	3.68	4.76	4.22	Control X 50 mM NaCl	35.49	34.46	34.97	2.24	2.87	2.55
1% EMS X 75 mM NaCl	26.55	29.94	28.25	2.50	4.24	3.37	Control X 75 mM NaCl	33.59	32.63	33.11	2.50	2.50	2.50
1% EMS X 100 mM NaCl	29.88	30.69	30.28	3.23	4.22	3.72	Control X 100 mM NaCl	36.23	34.90	35.56	2.32	2.65	2.48
2% EMS X Control	27.86	26.49	27.17	3.78	2.87	3.32	C.D. for Years	1.36	3.31				
2% EMS X 50 mM NaCl	26.78	32.12	29.40	3.36	4.22	3.79	C.D. for Treatment	7.678	3.20				
2% EMS X 75 mM NaCl	26.55	25.35	25.95	3.78	4.58	4.18	C.D. for Years X Treatments	3.739	2.58				
2% EMS X 100 mM NaCl	29.51	21.90	25.71	3.12	3.12	3.12							
3% EMS X Control	28.68	31.26	29.97	3.12	4.22	3.67							
3% EMS X 50 mM NaCl	27.10	32.12	29.54	3.62	4.31	3.96							
3% EMS X 75 mM NaCl	30.59	29.86	30.22	3.24	3.81	3.53							
3% EMS X 100 mM NaCl	31.56	32.43	31.99	3.36	3.92	3.64							

Table 4.4.11 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on membrane stability index and leaf scorching of strawberry cv. Winter Dawn

Treatment	MSI 2023	MSI 2024	MSI pooled data	Leaf scorchin g 2023	Leaf scorchi ng 2024	Leaf scorchi ng pooled data	Treatment	MSI 2023	MSI 2023	MSI pooled data	Leaf scorchi ng 2023	Leaf scorchi ng 2024	Leaf scorchi ng pooled data
1% EMS X Control	80.10	78.84	79.47	12.78	13.65	13.21	Control	79.45	80.46	79.54	15.43	17.23	16.33
1% EMS X 50 mM NaCl	81.65	79.71	80.68	14.53	10.46	12.49	Control X 50 mM NaCl	81.7	79.58	79.64	18.46	20.76	17.87
1% EMS X 75 mM NaCl	80.51	77.52	79.11	15.21	12.82	14.13	Control X 75 mM NaCl	74.46	76.78	75.62	19.68	18.55	19.11
1% EMS X 100 mM NaCl	84.69	79.56	82.12	14.7	11.89	13.29	Control X 100 mM NaCl	76.39	73.59	74.82	19.39	20.43	19.91
2% EMS X Control	81.56	79.63	80.59	16.62	11.74	14.18	C.D. for Years	1.13	0.47				
2% EMS X 50 mM NaCl	83.28	80.16	83.89	13.74	12.83	13.28	C.D. for Treatment	3.53	6.54				
2% EMS X 75 mM NaCl	81.54	82.36	81.95	14.50	9.61	12.17	C.D. for Years X Treatments	4.82	2.39				
2% EMS X 100 mM NaCl	79.92	77.90	78.91	11.84	12.89	12.36							
3% EMS X Control	79.77	79.12	79.41	19.52	11.35	15.43							
3% EMS X 50 mM NaCl	81.37	78.32	79.85	16.11	17.30	16.70							
3% EMS X 75 mM NaCl	81.7	77.41	79.55	18.64	12.77	15.70							
3% EMS X 100 mM NaCl	79.52	74.55	77.19	17.85	14.12	15.99							

(82.36%), and a minimum MSI was observed in runners exposed to 100 mM salinity condition (73.59%).

In 2023, however, the highest MSI was recorded in runners treated with a 1% EMS dose exposed to 100 mM salinity level (84.69%), However, the lowest MSI was observed in runners exposed to 75 mM salinity level (74.46%). From these results, it was observed that a 2% EMS dose significantly enhanced membrane stability under salinity stress compared to a 3% EMS dose.

4.4.23 Malondialdehyde content

The analysis of malondialdehyde content in Winter Dawn strawberry runners treated with the chemical mutagen EMS and subjected to varying salinity levels revealed a significant impact (Table 4.4.12) In the pooled analysis, the minimum malondialdehyde content was recorded in runners treated with a 2% EMS dose exposed to 100 mM NaCl (1.87 nmol/g) and the maximum was observed in runners induced to 100 mM salinity level (10.68 nmol/g).

During 2023, the minimum malondialdehyde content was recorded in runners treated with a 1% EMS dose induced with 75 mM NaCl (1.95 nmol/g), whereas the maximum was found in runners treated with 75 mM salinity condition (11.57 nmol/g). A similar trend was observed in 2024, where the minimum malondialdehyde content was recorded in runners treated with 1% EMS under the 100 mM salinity condition (1.49 nmol/g), and the maximum was found in runners induced to 50 mM induced salinity stress (11.49 nmol/g). EMS chemical mutagen significantly reduces malondialdehyde content under salinity stress, with the 2% EMS doses being the most effective in mitigating salinity stress.

4.4.25 Leaf area

The data on leaf area in Winter Dawn strawberry runners treated with a chemical mutagen under NaCl salinity stress indicates significant variations across both years (Table 4.4.12) In the pooled results, the maximum Leaf Area Index was observed in runners treated with a 2% EMS dose induced with 75 mM NaCl (21.46 mm), while the minimum LAI was recorded in runners induced to 100 mM induced salinity stress (10.68 mm).

In 2023, the highest leaf area index was recorded in runners treated with a 1% EMS dose under 75 mM NaCl (22.46 mm), whereas the lowest leaf area index was observed in runners exposed to 75 mM salinity level (9.86 mm). Similarly in 2024, the treated runners showed a

Table 4.4.12 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on leaf area index and malonaldehyde content of strawberry cv. Winter Dawn

Treatment	LAI 2023	LAI 2024	LAI pooled	Malondia ldehyde 2023	Malond ialdehy de 2024	Malond ialdehy de pooled	Treatment	LAI 2023	LAI 2024	LAI pooled	Malondia ldehyde 2023	Malond ialdehy de 2024	Malondia ldehyde Pooled
1% EMS X Control	17.66	19.29	18.47	5.36	2.13	3.74	Control	12.49	11.37	11.93	9.47	7.77	8.62
1% EMS X 50 mM NaCl	18.39	24.31	21.35	7.85	2.63	5.24	Control X 50 mM NaCl	11.39	10.78	11.08	7.03	11.49	9.26
1% EMS X 75 mM NaCl	22.46	18.42	20.44	1.95	2.19	2.07	Control X 75 mM NaCl	9.86	12.33	11.09	11.57	9.53	10.55
1% EMS X 100 mM NaCl	19.65	18.22	18.93	5.21	1.49	3.35	Control X 100 mM NaCl	10.43	11.29	10.86	10.62	10.74	10.68
2% EMS X Control	20.39	19.46	19.92	2.73	2.47	2.60	C.D. for Years	5.47	3.47				
2% EMS X 50 mM NaCl	18.69	20.11	19.4	2.96	2.41	2.68	C.D. for Treatment	6.46	8.66				
2% EMS X 75 mM NaCl	20.76	22.16	21.46	5.32	2.93	4.13	C.D. for Years X Treatments	9.21	7.19				
2% EMS X 100 mM NaCl	19.76	18.22	18.99	2.06	1.67	1.87							
3% EMS X Control	16.49	15.47	15.98	7.92	5.74	6.83							
3% EMS X 50 mM NaCl	16.46	15.35	15.90	9.95	6.33	8.14							
3% EMS X 75 mM NaCl	15.63	14.74	15.18	10.59	8.85	9.72							
3% EMS X 100 mM NaCl	14.76	16.22	15.49	9.36	9.55	9.45							

maximum leaf area index under 50 mM salinity condition (21.31 mm). However, the minimum leaf area index was recorded in runners induced with 50 mM salinity level (10.78 mm). The results demonstrate that chemical mutagen 2% EMS doses significantly improve leaf area under salinity stress in strawberry runners.

4.5 Interaction effect of gamma irradiation doses under varying levels of salinity in the Chandler Strawberry Variety

4.5.1 Plant height

The analysis of the data reveals that different doses of gamma irradiation doses against with varying levels of salinity significantly influenced the plant height of the Chandler variety of strawberry (Table 4.5.1) In the pooled analysis, the maximum plant height was recorded in runners treated with 30 Gy of gamma radiation against 50 mM NaCl (23.13 cm), which was statistically at par with 20 Gy dose. While the minimum plant height was observed in plants exposed to 100 mM NaCl (17.6 cm). A similar trend was recorded during 2023 treated runners showed maximum plant height was recorded in runners induced with 100 mM salinity condition (23.73 cm). The minimum plant height was recorded in runners against 100 mM salinity condition (16.83 cm).

In 2024, the maximum plant height was recorded in plants treated with 20 Gy induced with 100 mM NaCl (23.43 cm), while the minimum height was noted in runners under 0 mM condition (17.8 cm). This result suggests that runners treated with 30 Gy of gamma radiation had greater tolerance to salinity and maintained higher plant height under stress conditions.

4.5.2 Chlorophyll content index

The pertained data on the effect of different physical mutagens, specifically gamma irradiation, and varying levels of salinity significantly impacted the chlorophyll content index in the Chandler variety of strawberry plants (Table 4.5.1) The pooled data revealed that the maximum Chlorophyll content index was recorded in the 30 Gy gamma irradiation dose treated runners exposed with 50 mM NaCl (59.8), which was statistically at par with 20 Gy dose. while the minimum chlorophyll content index was observed in runners induced with 100 mM NaCl (45.8). A similar trend was noted in 2024 treated runners showed maximum chlorophyll content index recorded under 50 mM salinity level (60.46) and a minimum chlorophyll content index was recorder runners exposed to 100 mM salinity level

Table 4.5.1 Interaction effect of gamma irradiation doses under different levels of salinity on plant height and chlorophyll of strawberry cv. Chandler

Treatment	Plant height 2023	Plant height 2024	Plant height pooled data	Chlorophyll 2023	Chlorophyll 2024	Chlorophyll pooled data	Treatment	Plant height 2023	Plant height 2024	Plant height pooled data	Chlorophyll 2023	Chlorophyll 2024	Chlorophyll pooled data
20 Gy X Control	19.66	21.6	20.63	51.2	55.7	53.45	Control	24.9	17.8	21.35	53.26	47.43	50.35
20 Gy X 50 mM NaCl	19.16	23.33	21.25	60	56.4	58.64	Control X 50 mM NaCl	18.2	19.36	18.78	49.11	50.36	49.7
20 Gy X 75 mM NaCl	18.23	19.43	18.83	54.83	52.8	53.81	Control X 75 mM NaCl	18.36	17.16	17.76	52.8	46.96	49.88
20 Gy X 100 mM NaCl	20.39	23.43	21.92	56.93	48.7	52.81	Control X 100 mM NaCl	16.83	18.36	17.6	50.23	41.36	45.8
30 Gy X Control	17.53	19.10	18.3	51.73	51.8	51.76	C.D. for Years	0.97	0.82				
30 Gy X 50 mM NaCl	23.63	22.63	23.13	59.13	60.46	59.8	C.D. for Treatment	5.15	7.2				
30 Gy X 75 mM NaCl	23.2	22.23	22.71	58.16	50.26	54.21	C.D. for Years X Treatments	1.62	2.84				
30 Gy X 100 mM NaCl	23.73	20.6	22.16	54.63	49.7	52.16							
40 Gy X Control	17.3	17.46	17.38	49.86	49.12	49.45							
40 Gy X 50 mM NaCl	18.53	19.96	19.25	53.16	49.5	51.33							
40 Gy X 75 mM NaCl	19.8	19.23	19.51	51.8	47	49.4							
40 Gy X 100 mM NaCl	17.4	16.76	17.13	50.26	48.63	49.45							

(41.36). During 2023, the maximum chlorophyll content index was recorded in the combination of 20 Gy dose treated runners exposed with 75 mM NaCl (60), while the minimum was recorded in runners induced with 50 mM NaCl (49.11). This indicates that 30 Gy gamma irradiation was effective against salinity levels can enhance Chlorophyll development in strawberry plants.

4.5.3 Number of buds

It is evident from the pooled data that the number of buds was significantly influenced by the combined effects of different physical mutagen gamma irradiation doses and varying levels of salinity on the tolerance of strawberry plants in the Chandler variety (Table 4.5.2) The maximum number of buds was recorded in plants treated with a 30 Gy dose exposed to 50 mM NaCl (24.36) which was statistically at par with 20 Gy dose, while the minimum was found in runners exposed to 100 mM salinity level (17.33). A similar trend was recorded during 2023 treated runners showed a maximum number of buds in 0 mM condition (25.33) and minimum buds were found in runners induced with 75 mM salinity level (18.79).

In the 2024 season, the highest bud count was observed in the combination of 20 Gy dose against 50 mM NaCl (25.66), whereas the minimum was recorded in plants exposed to 100 mM salinity level (15.33).

4.5.4 Number of flowers

It is inquisition from the data that the number of flowers was significantly influenced in gamma irradiation-treated plants compared to those that were control in the Chandler strawberry variety (Table 4.5.2) The pooled results of gamma irradiation-treated runners subjected to varying levels of salinity reveal the maximum flower recorded in plants treated with a 30 Gy dose exposed to 50 mM salinity condition (27.33) which was statistically at par with 20 Gy dose. Conversely, the minimum flower count was observed in runners exposed to 100 mM salinity level (19.87). A similar trend was observed during 2023 in treated runners showed number of flowers in 0 mM salinity condition (28.79), while a minimum was recorded in runners against 75 mM salinity condition (19).

However, in 2024, the maximum flower count was recorded in the combination of a 20 Gy dose exposed to 50 mM NaCl (27). However, the minimum flower count was observed in runners exposed to 50 salinity conditions (18.33).

Table 4.5.2 Interaction effect of gamma irradiation doses under different levels of salinity on number of buds and flowers of strawberry cv. Chandler

Treatment	Number of Bud 2023	Number of bud 2024	Number of bud pooled data	Number of flowers 2023	Number of flowers 2024	Number of flowers pooled data	Treatment	Number of bud 2023	Number of bud 2024	Number of bud pooled data	Number of flowers 2023	Number of flowers 2024	Number of flowers pooled data
20 Gy X Control	24.66	22.33	23.5	26	24.33	25.16	Control	22	18.66	20.33	23	20	21.5
20 Gy X 50 mM NaCl	23	25.66	24.33	22.66	27	24.83	Control X 50 mM NaCl	22	18	20	20.33	18.33	20.33
20 Gy X 75 mM NaCl	23.33	19	21.12	25	23.66	24.33	Control X 75 mM NaCl	18.79	17.59	19.33	19	21	20
20 Gy X 100 mM NaCl	23.66	21.32	22.5	21	24.33	22.66	Control X 100 mM NaCl	19.33	15.33	17.33	20.26	19.49	19.87
30 Gy X Control	25.33	21.66	23.5	28.79	24	26.39	C.D. for Years	0.54	0.46				
30 Gy X 50 mM NaCl	24.87	23.86	24.36	28.66	26	27.33	C.D. for Treatment	2.09	4.09				
30 Gy X 75 mM NaCl	25	19.33	22.16	27	20.66	23.83	C.D. for Years X Treatments	2.55	2.74				
30 Gy X 100 mM NaCl	24.33	19.33	21.83	25.33	21.33	23.33							
40 Gy X Control	22.33	20.33	21.33	23.33	23	23.16							
40 Gy X 50 mM NaCl	19.88	17	18.44	25	22.66	23.83							
40 Gy X 75 mM NaCl	20.66	18	19.33	22.33	21.33	21.83							
40 Gy X 100 mM NaCl	19.66	17.31	18.5	23	21	22							

4.5.5 Number of fruit

It is evident from the (Table 4.5.3) that the combined effect of different doses of physical mutagen, specifically gamma irradiation, and varying levels of salinity significantly influenced the number of fruits produced by the Chandler variety of strawberry plant. The data demonstrate that the application of gamma irradiation, in conjunction with salinity, had a marked impact on fruit production. In the pooled analysis, the maximum number of fruits was recorded in 30 Gy gamma irradiation treated runners induced with 50 mM salinity level (28.66) which was statistically at par with 20 Gy dose. However, the minimum was observed in the runners exposed to 100 mM salinity condition (21.14). A similar trend was recorded during the 2023 mutagen treated runners showed maximum fruits in 0 mM salinity condition (29.33) and the minimum was recorded in runners against 75 mM salinity level (21.64).

However, in 2024, the maximum number of fruits was recorded again in the combination of 20 Gy gamma irradiation dose-treated runners exposed to 50 mM salinity level, whereas the minimum was found in runners exposed to 100 mM salinity condition (19.86).

4.5.6 Days to bud

The pooled data indicate that the days to bud initiation were influenced by the combined effects of different physical mutagen gamma irradiation doses and varying levels of salinity on the salinity tolerance of the Chandler variety of strawberry (Table 4.5.3) In the pooled analysis, early bud initiation was recorded in the treatment of 30 Gy dose exposed with 75 mM NaCl (39.33), while late bud initiation occurred in 100 mM highest salinity level (59.39). A comparable pattern was noted in 2024, with treated runners exhibiting early bud initiation under 75 mM salinity conditions (39.76), while runners showed delayed bud initiation under 100 mM NaCl (60.11). In 2023, early bud initiation was recorded in the treatment of 20 Gy dose against 75 mM salinity condition (24.66), and late bud initiation was recorded in runners induced with 75 mM salinity stress (60.33).

4.5.7 Days to flower

It is evident from the (Table 4.5.4) that days to flower were significantly influenced by the combined effect of gamma irradiation and varying levels of salinity in the Chandler variety of strawberry.

Table 4.5.3 Interaction effect of gamma irradiation doses under different levels of salinity on number of fruits and days to bud of strawberry cv. Chandler

Treatment	No. of fruits 2023	No. of fruits 2024	No. of fruits pooled data	Days to bud 2023	Days to bud 2024	Days to bud pooled data	Treatment	No. of fruits 2023	No. of fruits 2024	No. of fruits pooled data	Days to bud 2023	Days to bud 2024	Days to bud pooled data				
20 Gy X Control	26.33	28	27.16	26.33	59.66	43	Control	22.66	23.37	23.16	36	57	46.5				
20 Gy X 50 mM NaCl	20.33	29.33	24.83	48	47.333	47.66	Control X 50 mM NaCl	22.66	21.60	22.16	56.66	55	55.83				
20 Gy X 75 mM NaCl	24.33	26.66	25.5	24.66	52	38.33	Control X 75 mM NaCl	21.64	21.33	21.48	60.33	52	56.16				
20 Gy X 100 mM NaCl	26.66	25	25.83	41	52.53	46.83	Control X 100 mM NaCl	22.43	19.86	21.14	58.68	60.11	59.39				
30 Gy X Control	29.33	23.67	26.5	32.33	51	41.66	C.D. for Years	0.70	8.80								
30 Gy X 50 mM NaCl	28.33	29	28.66	42.23	50	46.16	C.D. for Treatment	5.38	26.81								
30 Gy X 75 mM NaCl	26.66	24	25.33	30.68	39.76	39.33	C.D. for Years X Treatments	2.92	37.63								
30 Gy X 100 mM NaCl	22.61	22.66	22.66	34	53.66	43.83											
40 Gy X Control	22.67	25.67	24.17	42	56	49											
40 Gy X 50 mM NaCl	24.62	27.33	26	45.66	52	48.83											
40 Gy X 75 mM NaCl	25.33	24.33	24.83	38.66	53	45.83											
40 Gy X 100 mM NaCl	22.76	23.11	22.93	51.66	52.33	52											

The combined results indicate that runners treated with a 30 Gy dose induced to 50 mM NaCl exhibited early flower initiation (50.48), whereas runners exposed to 100 mM salinity conditions showed delayed flower initiation (70.5). A similar trend was observed during 2023 early flower initiation was recorded in treated runners under 0 mM salinity condition (49.78) and late was observed in runners exposed to 75 mM NaCl salinity level (74.12). During 2024, the earliest flower initiation was recorded in runners treated with a 20 Gy dose exposed to 50 mM salinity level (49.78), while the late flower initiation occurred in runners exposed to 50 mM salinity level (70.11).

4.5.8 Days to fruit

The data clearly shows that fruit initiation days were significantly affected in runners treated with a 20 Gy and 30 Gy dose of the Chandler strawberry variety, compared to those control and 40 Gy (Table 4.5.4) The pooled results of gamma irradiation treated runners, induced with varying levels of salinity, showed the earliest fruit initiation was recorded in plants treated with a 30 Gy dose exposed to 50 mM NaCl (79), while the late fruit initiation was recorded in plants exposed to 100 mM salinity condition (99.39). A similar trend was observed in 2023 treated runners showed early fruit initiation under 0 mM salinity condition (72.43). However late fruit initiation found in runners against 50 mM salinity condition (99). Likewise, in 2024, fruit initiation was earlier found in runners treated with a 20 Gy dose under a 50 mM salinity level (80.66), whereas delayed fruit initiation was observed under a 75 mM salinity condition (101.11). The results indicate that 40 Gy dose and control runners of the Chandler strawberry variety show significant delays in bud, flower, and fruit initiation under salinity stress. In contrast, treatments with 30 Gy doses of gamma radiation led to earlier initiation of this stage.

4.5.9Fruit length

The data in (Table 4.5.5) indicates that fruit length in the Chandler strawberry variety was significantly influenced by varying doses of the physical mutagen gamma irradiation, demonstrating an impact on tolerance to salinity stress. The pooled results show that the maximum fruit length was observed in runners treated with a 20 Gy dose exposed to salinity stress 75 mM level (39.33 mm) which was statistically at par with a 30 Gy dose. While the minimum fruit length was recorded in runners exposed to the highest salinity stress 100 mM level (29.82 mm). A similar trend was recorded in 2024 the treated runners showed maximum fruit length

Table 4.5.4 Interaction effect of gamma irradiation doses under different levels of salinity on days to flower and fruits of strawberry cv. Chandler

Treatment	Days to flowers 2023	Days to flowers 2024	Days to flower pooled data	Days to fruits 2023	Days to fruits 2024	Days to fruit pooled data	Treatment	Days to flowers 2023	Days to flowers 2024	Days to flower pooled data	Days to fruits 2023	Days to fruits 2024	Days to fruits pooled data	
20 Gy X Control	54.73	63.32	59.16	90.26	81.46	86	Control	63.22	61.23	62.22	88.10	88.62	85.86	
20 Gy X 50 mM NaCl	52.64	49.78	51.21	84	80.66	82.33	Control X 50 mM NaCl	61.23	70.11	65.67	99	93.33	96.16	
20 Gy X 75 mM NaCl	50.47	67.33	58.83	83.19	86.33	84.76	Control X 75 mM NaCl	74.12	62.32	68.22	97.32	101.11	99.21	
20 Gy X 100 mM NaCl	53.23	62.11	57.67	79.68	91.12	85.4	Control X 100 mM NaCl	72	69	70.5	98.67	100.12	99.39	
30 Gy X Control	49.78	61.76	55.77	72.43	81.39	76.91	C.D. for Years	1.41	0.54					
30 Gy X 50 mM NaCl	49.86	51.10	50.48	72.45	85.31	79	C.D. for Treatment	2.91	3.45					
30 Gy X 75 mM NaCl	46.38	58.54	52.46	78.66	82.66	80.66	C.D. for Years X Treatments	4.51	3.32					
30 Gy X 100 mM NaCl	49.58	61.33	55.45	80.21	81.32	80.76								
40 Gy X Control	68.49	63.37	65.93	89.33	82.33	85.83								
40 Gy X 50 mM NaCl	61.46	67.49	64.47	84.63	83.49	84.06								
40 Gy X 75 mM NaCl	58.67	69.46	64.1	89.19	97.28	93.23								
40 Gy X 100 mM NaCl	67.66	64.61	66.16	90.46	99.22	91.34								

Table 4.5.5 Interaction effect of gamma irradiation doses under different levels of salinity on fruit length and fruit diameter of strawberry cv. Chandler

Treatment	Fruit length 2023	Fruit length 2024	Fruit length pooled data	Fruit diameter 2023	Fruit diameter 2024	Fruit diameter pooled data	Treatment	Fruit length 2023	Fruit length 2024	Fruit length pooled data	Fruit diameter 2023	Fruit diameter 2024	Fruit diameter Pooled data
20 Gy X Control	36.95	30.45	33.70	32.96	29.15	31.05	Control	38.25	28.48	33.36	27.68	22.10	24.89
20 Gy X 50 mM NaCl	37.67	36.12	36.84	30.29	33.22	31.75	Control X 50 mM NaCl	29.44	27.12	28.28	33.95	19.41	26.68
20 Gy X 75 mM NaCl	39.35	31.59	35.47	33.80	31.12	32.46	Control X 75 mM NaCl	28.43	30.11	29.27	20.89	21.44	21.16
20 Gy X 100 mM NaCl	41.64	29.50	35.57	31.72	29.69	30.70	Control X 100 mM NaCl	30.19	29.45	29.82	21.64	23.11	22.37
30 Gy X Control	40.48	30.32	35.40	32.76	30.11	31.43	C.D. for Years	0.59	0.14				
30 Gy X 50 mM NaCl	30.97	40.42	35.69	32.78	33.11	32.94	C.D. for Treatment	7.72	8.49				
30 Gy X 75 mM NaCl	39.14	39.62	39.33	32.54	29.11	30.82	C.D. for Years X Treatments	2.88	2.20				
30 Gy X 100 mM NaCl	40.22	36.93	38.61	30.12	31.29	30.70							
40 Gy X Control	37.41	29.6	33.50	30.44	21.95	26.20							
40 Gy X 50 mM NaCl	36.24	32.61	34.77	30.61	25.57	28.09							
40 Gy X 75 mM NaCl	38.35	27.49	32.92	31.61	27.58	29.09							
40 Gy X 100 mM NaCl	37.6	33.63	35.61	29.68	30.23	29.95							

in 50 mM salinity condition (40.42 mm) and minimum was found in runners exposed to 50 mM NaCl (20.89 mm). In 2023, the highest fruit length was recorded in runners treated with a 30 Gy dose under 100 mM salinity level (41.64 mm), while the lowest was observed under runners induced with salinity treatment 75 mM level (28.43 mm).

4.5.10 Fruit diameter

It is evident from the pooled data that fruit diameter was significantly affected by the varying salinity levels instead of 20 Gy and 30 Gy doses in the Chandler strawberry variety (Table 4.5.5) The maximum fruit diameter was recorded in plants treated with a 30 Gy dose exposed to 50 mM salinity stress (32.94 mm), which was statistically at par with 20 Gy dose. The minimum fruit diameter was observed in runners exposed to 100 mM salinity condition (22.37 mm). In 2023, the maximum fruit diameter was recorded in runners treated with a 20 Gy dose against 75 mM salinity condition (33.80 mm). However, the minimum was recorded in runners exposed to 75 mM salinity level (20.89 mm). A similar trend was recorded in 2024 treated runners showing maximum fruit diameter was observed in runners exposed to 50 mM salinity level (33.22 mm), while the minimum diameter was recorded in plants exposed to 50 mM salinity treatment (19.41 mm).

4.5.11 Fruit weight

In the present study on the Chandler variety of strawberry (Table 4.5.6) the interaction between different doses of gamma irradiation and varying levels of salinity was examined with respect to fruit weight as an indication of salinity tolerance. The pooled data revealed that the highest fruit weight was observed in the treatment combination of 30 Gy gamma irradiation dose induced with 50 mM salinity conditions (11.51gm), which was statistically at par with 20 Gy. The lowest fruit weight was recorded in 100 mM salinity level (8.51gm). During the 2024023, the maximum fruit weight of was recorded in the combination of 20 Gy gamma irradiation dose induced with 50 mM NaCl (11.80 gm), whereas the minimum fruit weight was noted in runners against 50 mM salinity condition (9.12 gm). A similar trend was observed in the 2024024, treated runners showed maximum fruit weight under 75 mM salinity condition (11.33 gm). However, the minimum weight of the fruit was found runners under the 75 mM salinity level (7.62 gm). Confirming the consistency of the results across both years. In conclusion, 30 Gy gamma irradiation treated runners showed maximum fruit weight per plant under salinity conditions.

Table 4.5.6 Interaction effect of gamma irradiation doses under different levels of salinity on fruits weight and fruit yield of strawberry cv. Chandler

Treatment	Fruit weight 2023	Fruit weight 2024	Fruit weight pooled data	Fruit yield 2023	Fruit yield 2024	Fruit yield pooled data	Treatment	Fruit weight 2023	Fruit weight 2024	Fruit weight pooled data	Fruit yield 2023	Fruit yield 2024	Fruit yield pooled data				
20 Gy X Control	11.24	9.4	10.36	355.66	269.33	312.5	Control	10.66	7.90	9.28	338	253	295.5				
20 Gy X 50 mM NaCl	11.80	10.10	10.95	387	318.66	352.83	Control X 50 mM NaCl	9.12	9.73	10.21	287.66	206	246.83				
20 Gy X 75 mM NaCl	10.18	11.33	10.75	354.66	262.33	308.3	Control X 75 mM NaCl	9.58	7.62	8.60	327.59	221.66	274.66				
20 Gy X 100 mM NaCl	11.74	9.80	10.13	366.33	277.66	322	Control X 100 mM NaCl	9.13	7.89	8.51	288.66	217	252.83				
30 Gy X Control	10.29	9.19	9.74	320.66	361.33	341	C.D. for Years	0.15	2.53								
30 Gy X 50 mM NaCl	11.74	11.28	11.51	376	385.33	380.66	C.D. for Treatment	1.46	10.63								
30 Gy X 75 mM NaCl	11.18	9.10	10.14	313.33	390	351.16	C.D. for Years X Treatments	1.19	8.57								
30 Gy X 100 mM NaCl	9.79	8.39	9.09	301.33	329.66	315.5											
40 Gy X Control	9.66	8.25	8.96	349.66	273.33	311.5											
40 Gy X 50 mM NaCl	9.35	8.44	8.89	306	256	281											
40 Gy X 75 mM NaCl	10.06	9.21	9.63	335.33	247.66	291.5											
40 Gy X 100 mM NaCl	9.94	8.82	9.38	377.66	273.33	325.5											



Plate 10. Salinity affected fruits in control runners and good quality fruits in 20Gy and 30 Gy

4.5.12 Fruit yield

The perusal data in (Table 4.5.6) shows that fruit yield in the Chandler strawberry variety was significantly influenced by the interaction of gamma irradiation doses and varying levels of salinity. The pooled and individual results show that gamma irradiation had a noticeable impact on fruit yield under salinity stress. In the pooled analysis, the highest fruit yield was recorded in plants treated with a 30 Gy dose induced with 50 mM salinity level (380.66 gm), which was statistically at par with 20 Gy dose. The lowest yield was observed in runners exposed to 100 mM highest salinity condition (252.83 gm). A similar pattern was observed in 2024, with treated runners achieving the highest fruit yield under 75 mM NaCl salinity conditions (390 gm), while runners exhibited the lowest fruit yield under 50 mM NaCl (287.66 gm). In 2023, the maximum fruit yield was noted in the combination of 20 Gy dose induced with 50 mM salinity level (387 gm). However, the lowest yield was recorded in runners exposed to 50 mM salinity condition (287.66 gm).

4.5.13 Shoot fresh weight

The data pertaining to indicate that gamma radiation had a significant impact on shoot fresh weight, with notable differences observed between treated and plants under salinity stress in the Chandler strawberry variety (Table 4.5.7) The pooled analysis showed that runners

Table 4.5.7 Interaction effect of gamma irradiation doses under different levels of salinity on shoot fresh weight and dry weight of strawberry cv. Chandler

Treatment	Shoot fresh wt. 2023	Shoot fresh wt. 2024	Shoot fresh wt. pooled data	Shoot dry wt. 2023	Shoot dry wt. 2024	Shoot dry wt. pooled data	Treatment	Shoot fresh wt. 2023	Shoot fresh wt. 2024	Shoot fresh wt. pooled data	Shoot dry wt. 2023	Shoot dry wt. 2024	Shoot dry wt. pooled data
20 Gy X Control	8.18	9.83	9.00	3.17	1.90	2.54	Control	7.77	8.48	8.12	3.01	1.77	2.39
20 Gy X 50 mM NaCl	10.26	10.26	10.26	3.90	2.23	3.07	Control X 50 mM NaCl	8.21	7.33	7.77	2.10	2.89	2.49
20 Gy X 75 mM NaCl	11.40	11.60	11.5	4.07	2.92	3.5	Control X 75 mM NaCl	6.32	8.64	7.48	2.64	1.35	1.99
20 Gy X 100 mM NaCl	10.17	10.21	10.19	2.65	1.66	2.15	Control X 100 mM NaCl	6.85	7.59	7.22	2.21	1.55	1.85
30 Gy X Control	10.32	9.82	10.17	2.97	2.18	2.57	C.D. for Years	0.26	0.76				
30 Gy X 50 mM NaCl	11.36	13.06	12.21	4.01	3.11	3.56	C.D. for Treatment	1.91	3.64				
30 Gy X 75 mM NaCl	10.17	13.69	11.93	2.21	3.52	2.86	C.D. for Years X Treatments	1.51	9.78				
30 Gy X 100 mM NaCl	9.13	8.79	8.96	2.24	1.99	2.12							
40 Gy X Control	8.77	8.48	8.62	3.06	1.84	2.45							
40 Gy X 50 mM NaCl	7.83	9.83	8.83	3.65	1.88	2.76							
40 Gy X 75 mM NaCl	8.00	8.53	8.26	2.7	1.70	2.20							
40 Gy X 100 mM NaCl	8.84	7.59	8.22	2.737	1.47	2.10							

treated with a 30 Gy dose under 50 mM NaCl achieved the highest shoot fresh weight (12.21 gm) which was statistically at par with 20 Gy mutagen dose. As well as the lowest was recorded in plant under 100 mM NaCl (7.22 gm). A similar trend was observed in 2024, where treated runners exhibited the maximum shoot fresh weight (13.69 gm) under 75 mM salinity, whereas runners recorded the minimum shoot fresh weight under 50 mM salinity (7.33 gm).

In 2023, the maximum shoot fresh weight of was recorded in plants treated with a 20 Gy dose against 75 mM NaCl (11.40 gm) and the minimum shoot fresh weight was found in runners exposed with 75 mM salinity condition (6.32 gm). These findings emphasize the effectiveness of gamma radiation, particularly at 20 Gy and 30 Gy doses, in promoting shoot fresh weight under salinity stress conditions.

4.5.14 Shoot dry weight

The results from the study indicate that gamma radiation had a significant impact on shoot dry weight, with notable differences observed between treated and control runners under salinity stress in the Chandler strawberry variety (Table 4.5.7). The pooled analysis revealed that the maximum shoot dry weight was recorded in runners treated with a 30 Gy dose exposed to 75 mM NaCl (3.52 gm) which was statistically at par with 20 Gy mutagen dose. The minimum shoot dry weight was observed in without mutagen-treated runners against 100 mM NaCl (1.85 gm). A similar trend was observed during 2024 treated runners showed maximum shoot dry weight recorded in runners induced with 75 mM salinity level (3.52 gm), while the minimum shoot dry weight observed in runners against 50 mM salinity condition (2.10 gm). However, in 2023, the maximum shoot dry weight was recorded in the interaction of 20 Gy dose induced with 50 mM salinity condition (3.90 gm), while the minimum was noted runners exposed to 50 mM NaCl (2.10 gm).

4.5.15 Root fresh weight

The study results show that gamma radiation significantly affected root fresh weight, with clear differences between treated and Chandler strawberry runners under salinity stress (Table 4.5.8). The pooled analysis revealed that the maximum root fresh weight was recorded in plants treated with a 20 Gy dose exposed to 75 mM NaCl (13.29 gm) which was statistically at par with 30 Gy mutagen dose. The minimum root fresh weight was observed in runners exposed to 100 mM NaCl (10.66 gm). A similar trend was observed during 2023

Table 4.5.8 Interaction effect of gamma irradiation doses under different levels of salinity on root fresh weight and dry weight of strawberry cv. Chandler

Treatment	Root fresh wt. 2023	Root fresh wt. 2024	Root fresh wt. pooled data	Root dry wt. 2023	Root dry wt. 2024	Root dry wt. pooled data	Treatment	Root fresh wt. 2023	Root fresh wt. 2024	Shoot fresh wt. pooled data	Root dry wt. 2023	Root dry wt. 2024	Root dry wt. pooled data
20 Gy X Control	14.07	10.58	12.32	5.1	2.27	3.70	Control	12.17	9.40	10.78	4.39	1.84	3.12
20 Gy X 50 mM NaCl	15.87	10.39	13.13	5.03	3.44	4.23	Control X 50 mM NaCl	13.83	9.6	11.71	3.86	1.56	2.71
20 Gy X 75 mM NaCl	15.36	11.22	13.29	4.76	3.95	4.35	Control X 75 mM NaCl	10.89	9.87	10.38	3.8	1.99	2.89
20 Gy X 100 mM NaCl	14.42	10.40	12.41	4	2.30	3.15	Control X 100 mM NaCl	11.10	10.10	10.6	3.68	1.69	2.68
30 Gy X Control	14.27	10.65	12.46	4.14	2.88	3.51	C.D. for Years	0.38	0.41				
30 Gy X 50 mM NaCl	10.11	11.40	10.75	3.43	4.10	3.76	C.D. for Treatment	1.27	1.74				
30 Gy X 75 mM NaCl	14.24	10.07	12.16	3.10	3.01	3.05	C.D. for Years X Treatments	1.52	0.78				
30 Gy X 100 mM NaCl	13.41	9.66	11.54	3.88	2.46	3.17							
40 Gy X Control	14.71	10.12	12.42	4.91	1.55	3.23							
40 Gy X 50 mM NaCl	13.22	9.31	11.19	4.18	2.81	3.49							
40 Gy X 75 mM NaCl	13.03	9.51	11.26	3.88	1.74	2.81							
40 Gy X 100 mM NaCl	11.81	10.52	10.66	3.13	2.16	2.65							

treated runners showed maximum root fresh weight under 50 mM salinity level (15.87 gm) also minimum root fresh weight was recorded in runners induced with 75 mM salinity level (10.89 gm). However, in 2024, the maximum root fresh weight was recorded in the combination of 30 Gy mutagen dose treated runners against 50 mM NaCl (11.40 gm) and the minimum was observed in runners exposed to 50 mM salinity level (9.6 gm). These results highlight the effectiveness of gamma radiation, particularly at 20 Gy dose, in enhancing root development under salinity stress conditions.

4.5.16 Root dry weight

After evaluating root fresh weight, root dry weight was measured in the interaction between the physical mutagen and induced salinity stress NaCl (Table 4.5.8). The pooled analysis showed a maximum root dry weight in the 20 Gy mutagen dose treated runners against 75 mM salinity condition (4.35 gm) and minimum root dry weight was recorded in runners against 50 mM salinity level (2.71 gm). A similar trend was recorded during 2023, the maximum root dry weight was recorded in runners treated with 20 Gy gamma irradiation dose under 50 mM NaCl (5.03 gm), while the lowest was found in runners against 100 mM NaCl treatment (2.71 gm).

However, in 2024 the treated runners with 30 Gy showed maximum root dry weight recorded (4.10 gm) under 100 mM induced salinity condition and a minimum was found in runners induced against 50 mM salinity level (1.56 gm).

4.5.17 Number of leaves

In the Chandler variety of strawberry, the number of leaves was recorded in gamma irradiation-treated plants subjected to induced salinity stress (Table 4.5.9). In both years of the experiment, the maximum number of leaves was observed in plants treated with a combination of 20 Gy gamma irradiation treated runners against 50 mM NaCl (10.33). which was statistically at par with 30 Gy dose. In contrast, the minimum number of leaves was recorded in runners exposed to 100 mM salinity level (5.96). A similar trend was noted in 2023 mutagen treated runners showed a maximum number of leaves under 50 mM salinity level (11) and a minimum was found in runners exposed with 100 mM NaCl (6.17). However, during 2024, the maximum number of leaves was recorded in plants treated with 30 Gy gamma irradiation dose against 50 mM NaCl (12.33), and the minimum of leaves was recorded runners exposed with 100 mM NaCl (5.76).

Tabel 4.5.9 Interaction effect of gamma irradiation doses under different levels of salinity on number of leaves and leaf relative water content of strawberry cv. Chandler

Treatment	No. of leaves 2023	No. of leaves 2024	No. of leaves pooled data	LRWC 2023	LRWC 2024	LRWC pooled data	Treatment	No. of leaves 2023	No. of leaves 2024	No. of leaves pooled data	LRWC 2023	LRWC 2024	LRWC pooled data
20 Gy X Control	10	11	10.5	78.88	76.95	77.92	Control	9.33	8	8.66	76.98	69.33	73.15
20 Gy X 50 mM NaCl	11	9.53	10.33	87.21	82.29	84.75	Control X 50 mM NaCl	8	8.42	8.33	68.21	64.32	66.26
20 Gy X 75 mM NaCl	9	9	9	88.13	79.23	83.68	Control X 75 mM NaCl	6.51	6.33	6.5	65.11	67.16	66.08
20 Gy X 100 mM NaCl	8.33	8.61	8.5	67.82	64.62	66.22	Control X 100 mM NaCl	6.17	5.76	5.96	65.27	66.82	66.04
30 Gy X Control	8.33	11.31	9.83	78.29	72.52	75.40	C.D. for Years	0.97	1.20				
30 Gy X 50 mM NaCl	8	12.33	10.16	75.79	84.67	80.23	C.D. for Treatment	4.38	9.04				
30 Gy X 75 mM NaCl	8	9	8.5	79.28	79.43	79.35	C.D. for Years X Treatments	2.20	3.55				
30 Gy X 100 mM NaCl	7.67	8.33	8	67.65	71.58	69.62							
40 Gy X Control	10	9.33	9.66	68.29	67.75	68.02							
40 Gy X 50 mM NaCl	8.33	9.66	9	78.69	75.81	77.25							
40 Gy X 75 mM NaCl	6.61	9	7.83	79.15	71.14	75.14							
40 Gy X 100 mM NaCl	5.58	8.33	7	78.85	62.62	70.73							

4.5.18 Leaf relative water content

The analysis of the data reveals that leaf relative water content LRWC was significantly influenced by physical mutagen gamma irradiation treated runners combined with induced salinity stress in the Chandler variety of strawberry (Table 4.5.9) The pooled data shows that the maximum LRWC was recorded in runners treated with a combination of 20 Gy gamma irradiation dose exposed with 50 mM NaCl (84.75%), while the minimum LRW was observed in plants exposed to 100 mM NaCl (66.04%). A similar trend was observed during 2023 in mutagen treated runners showing maximum LRWC was recorded under 75 mM salinity level (88.13%) and minimum LRWC was found in runners against 75 mM salinity condition (65.11%).

However, during 2024 the maximum LRWC was observed in plants treated with 30 Gy gamma irradiation treated runners against 50 mM NaCl (84.67%), whereas the minimum LRWC was recorded in 50 mM salinity conditions (65.11%).

4.5.19 Electrolyte leakage

The results from the study indicate that electrolyte leakage serves as a useful indicator of salt tolerance in the Chandler variety of strawberry (Table 4.5.10) The interaction of different doses of gamma irradiation against induced salinity stress was assessed to evaluate salinity tolerance. In the pooled analysis, the lowest electrolyte leakage was observed in runners treated with 20 Gy dose exposed to 100 mM NaCl (26.31%) and the highest electrolyte leakage was recorded in runners exposed to 100 mM NaCl (34.21%). A similar trend was observed during 2023 the treated runners showed minimum electrolyte leakage was observed in plants treated with 20 Gy under 50 mM NaCl (25.94%), while the maximum was recorded in runners exposed to 75 mM salinity level (35.47%).

However, in 2024, the lowest electrolyte leakage was observed in plants treated with 30 Gy dose exposed to 100 mM NaCl (23.78%), whereas the highest was recorded in plants exposed to 100 mM salinity condition (33.61%). These findings highlight the effectiveness of gamma radiation, particularly at 20 Gy dose, in reducing electrolyte leakage under 100 mM induced salinity stress concentrations.

4.5.20 Proline content

Table 4.5.10 Interaction effect of gamma irradiation doses under different levels of salinity on electrolyte leakage and proline content of strawberry cv. Chandler

Treatment	E L % 2023	E L % 2024	E L % pooled data	Proline content 2023	Proline content 2024	Proline content pooled data	Treatment	E L % 2023	E L % 2024	E L % pooled data	Proline content 2023	Proline content 2024	Proline content pooled data	
20 Gy X Control	29.69	28.28	28.98	2.27	2.66	2.46	Control	29.03	24.39	26.71	2.14	1.87	2.08	
20 Gy X 50 mM NaCl	25.94	26.19	26.06	2.51	3.21	2.86	Control X 50 mM NaCl	34.79	29.33	32.06	1.42	2.11	1.71	
20 Gy X 75 mM NaCl	31	25.1	28.08	1.99	2.39	2.19	Control X 75 mM NaCl	35.47	31.62	33.54	2.13	1.56	1.84	
20 Gy X 100 mM NaCl	28.24	24.38	26.31	3.62	2.34	2.98	Control X 100 mM NaCl	34.81	33.61	34.21	1.77	1.80	1.78	
30 Gy X Control	29.24	27.24	28.24	2.24	2.60	2.42	C.D. for Years	0.41	0.23					
30 Gy X 50 mM NaCl	30.53	23.96	27.24	3.48	3.10	3.29	C.D. for Treatment	1.74	1.11					
30 Gy X 75 mM NaCl	32.36	24.38	28.37	2.83	2.43	2.63	C.D. for Years X Treatments	0.78	0.62					
30 Gy X 100 mM NaCl	28.88	23.78	26.33	1.52	2.80	2.16								
40 Gy X Control	29.14	28.27	28.71	2.35	1.88	2.11								
40 Gy X 50 mM NaCl	33.88	26.41	30.14	2.33	2.14	2.23								
40 Gy X 75 mM NaCl	32.84	26.77	29.81	2.9	1.77	2.33								
40 Gy X 100 mM NaCl	29.22	26.27	27.74	1.96	2.20	2.08								

The results from the study indicate that proline content was significantly affected in Chandler variety strawberry plants treated with physical mutagens under induced salinity stress (Table 4.5.10) The pooled analysis showed that the maximum proline content was recorded, in runners treated with mutagen 30 Gy dose exposed to 50 mM NaCl (3.29 $\mu\text{mol/g}$) which was statistically at par with 20 Gy gamma irradiation dose. On the other hand, the lowest proline content was recorded in runners exposed to 100 mM salinity level (1.78 $\mu\text{mol/g}$).

During 2023 the maximum proline was recorded in plants treated with a 20 Gy dose against 100 mM salinity condition (3.62 $\mu\text{mol/g}$), while the minimum proline content was found in runners against 50 mM salinity condition (1.42 $\mu\text{mol/g}$). However, a Similar trend was recorded in the 2024 the mutagen treated runners against 50 mM NaCl showed maximum proline content (3.21 $\mu\text{mol/g}$) and minimum proline was recorded in runners against 75mM salinity level (1.56 $\mu\text{mol/g}$).

4.5.21 Membrane stability index

The results from the study indicate that gamma radiation had a significant impact on membrane stability index (MSI), with notable differences observed between mutagen treated and control plant under salinity stress in the Chandler strawberry variety (Table 4.5.11) Based on the pooled data, the maximum Membrane Stability Index was observed in the treatment of 20 Gy dose against 50 mM NaCl (84.37 %), which was statistically at par with 30 Gy dose. While the minimum MSI of was recorded in the without mutagen treated runners against 100 mM NaCl treatment (79.10 %). A similar trend was recorded in 2024 maximum MSI was observed in the 20 Gy dose against 75 mM NaCl treatment (85.12%), while the lowest MSI was recorded in control runners under the 100 mM NaCl treatment (78.41%).

In 2023, the highest MSI was noted in the 30 Gy dose exposed with 50 mM NaCl treatment (85.16 %) and the lowest MSI recorded in the 75 mM NaCl treatment (78.36 %).

4.5.22 Leaf scorching

Leaf scorching was recorded in the Chandler variety of strawberry. The (Table 4.5.11) presents interaction data on the effects of different doses of the physical mutagen gamma irradiation combined with varying levels of salinity on leaf scorching. The data show that the application of gamma irradiation and salinity had a significant influence on the leaf scorching of strawberry plants. In the pooled analysis, the minimum leaf scorching was recorded in the combination of 20 Gy dose against 75 mM NaCl (12.28 %), while the maximum scorching was observed in

Table 4.5.11 Interaction effect of gamma irradiation doses under different levels of salinity on membrane stability index and leaf scorching of strawberry cv. Chandler

Treatment	MSI 2023	MSI 2024	MSI pooled data	Leaf Scorchi ng 2023	Leaf Scorchi ng 2024	Leaf Scorchi ng pooled data	Treatment	MSI 2023	MSI 2024	MSI pooled data	Leaf Scorchi ng 2023	Leaf Scorchi ng 2024	Leaf Scorchi ng pooled data
20 Gy X Control	84.9	81.15	83.02	20.81	20.24	20.52	Control	83.1	79.71	81.40	32.54	37.11	34.28
20 Gy X 50 mM NaCl	84.23	84.52	84.37	16.66	9.15	12.90	Control X 50 mM NaCl	84.43	80.21	82.32	42.23	44.19	43.21
20 Gy X 75 mM NaCl	80.32	85.12	81.63	14.39	10.18	12.28	Control X 75 mM NaCl	78.36	79.40	79.38	50.11	37.46	43.78
20 Gy X 100 mM NaCl	79.93	80.43	80.18	12.64	19.32	15.98	Control X 100 mM NaCl	79.18	78.41	79.10	49.33	40.19	44.76
30 Gy X Control	81.8	82.16	81.98	14.88	17.85	16.36	C.D. for Years	0.81	0.96				
30 Gy X 50 mM NaCl	85.16	83.3	84.23	16.86	15.18	16.02	C.D. for Treatment	3.05	6.04				
30 Gy X 75 mM NaCl	83.03	83.12	83.08	16.29	18.15	17.22	C.D. for Years X Treatments	1.92	2.76				
30 Gy X 100 mM NaCl	79.8	79.84	79.82	19.32	21.19	20.25							
40 Gy X Control	83.13	80.39	81.76	29.43	28.19	28.81							
40 Gy X 50 mM NaCl	81.7	80.56	81.13	25.32	27.11	26.21							
40 Gy X 75 mM NaCl	81.76	80.5	81.13	27.67	28.49	28.08							
40 Gy X 100 mM NaCl	80.16	78.79	79.48	26.49	29.18	27.83							

the 100 mM NaCl treatment (44.76 %). A similar trend was recorded during both 2023 and 2024. In this observation, we found that a gamma irradiation dose of 20 Gy had positive effects on salinity tolerance, resulting in reduced leaf scorching in the plants.

4.5.23 Malonaldehyde content

In (Table 4.5.12) shows the tolerance of physical mutagen gamma irradiation treated runners against induced salinity NaCl concentration in the Chandler variety of strawberry. In this observation, it was observed that the plant treated with mutagen gamma irradiation doses shows tolerance to salinity and found minimum malonaldehyde content instead of control runners and 40 Gy dose treated runners. In Pooled analysis minimum malonaldehyde content was recorded in 20 Gy dose treated runners against 75 mM NaCl (1.50 $\mu\text{mol/g}$) and maximum malonaldehyde content was found under control runners induced with 100 mM salinity condition (11.59 $\mu\text{mol/g}$). A similar trend was recorded during 2024 the treated runners showed minimum malonaldehyde content was found under 75 mM salinity condition (1.24 $\mu\text{mol/g}$) and a maximum was recorded under 50 mM salinity level (11.75 $\mu\text{mol/g}$). However, in 2023 minimum malonaldehyde content was recorded minimum in runners treated with 30 Gy dose treated runners against 50 mM NaCl (1.70 $\mu\text{mol/g}$) and maximum malonaldehyde content was found in plant under 75 mM NaCl condition (11.59 $\mu\text{mol/g}$).

4.5.23 Leaf area

The leaf area was recorded in gamma-irradiated runners exposed to induced salinity through NaCl concentration in the Chandler variety of strawberry plants (Table 4.5.12) In the analysis, it was observed that plants treated with 20 Gy and 30 Gy doses showed maximum leaf area, rather than the 40 Gy and control runners. In pooled analysis, the maximum leaf area was recorded in runners treated with a 30 Gy dose against 100 mM NaCl (24.79 mm) and the minimum leaf area was recorded in runners exposed to 100 mM NaCl (10.84 mm).

During 2023, the maximum leaf area was found in runners treated with a 20 Gy dose against 50 mM NaCl (26.17 mm). However the minimum leaf area was recorded in runners exposed to 75 mM NaCl (9.87 mm).

A similar trend was recorded during 2024 maximum leaf area was recorded in runners treated with a 20 Gy dose induced with 75 mM NaCl (21.32 mm), whereas the minimum leaf area, was found in runners exposed to 50 mM salinity condition (10.78 mm).

Table 4.5.12 Interaction effect of gamma irradiation doses under different levels of salinity on leaf area index and malonaldehyde content of strawberry cv. Chandler

Treatment	LAI 2023	LAI 2024	LAI pooled	Malondi aldehyd e 2023	Malond ialdehy de 2024	Malond ialdehy de pooled	Treatment	LAI 2023	LAI 2024	LAI pooled	Malond ialdehy de 2023	Malond ialdehy de 2024	Malond ialdehy de pooled
20 Gy X Control	24.79	21.46	23.12	3.0	2.96	2.98	Control	18.74	16.67	17.70	10.26	10.62	10.44
20 Gy X 50 mM NaCl	26.17	21.30	23.73	3.48	3.43	3.45	Control X 50 mM NaCl	11.78	10.34	11.06	10.21	11.75	10.98
20 Gy X 75 mM NaCl	19.36	27.11	23.23	1.76	1.24	1.50	Control X 75 mM NaCl	9.87	10.63	10.25	11.83	10.21	11.02
20 Gy X 100 mM NaCl	21.78	20.19	20.98	2.94	3.33	3.13	Control X 100 mM NaCl	10.24	11.45	10.84	11.67	11.52	11.59
30 Gy X Control	23.58	21.87	22.72	1.80	2.05	1.92	C.D. for Years	1.37	0.79				
30 Gy X 50 mM NaCl	24.49	25.10	24.79	1.70	1.92	1.81	C.D. for Treatment	9.43	8.64				
30 Gy X 75 mM NaCl	22.63	23.10	22.86	2.02	1.99	2.01	C.D. for Years X Treatments	6.19	7.46				
30 Gy X 100 mM NaCl	21.35	22.49	21.92	3.91	3.61	3.76							
40 Gy X Control	19.23	21.34	20.28	7.66	8.51	8.08							
40 Gy X 50 mM NaCl	16.57	18.43	17.5	8.05	9.64	8.84							
40 Gy X 75 mM NaCl	17.38	16.29	16.83	8.60	10.06	9.33							
40 Gy X 100 mM NaCl	18.53	19.32	18.92	9.36	10.67	10.02							



Plate 11. Salinity causes leaf edge burn, necrosis, nutrient imbalance, or specific ion toxicity, as a reduction in fruit quality, yield as well as plant growth and death of the plant

Interaction effect of ethyl methanesulfonate doses under varying levels of salinity in the Chandler Strawberry Variety

4.6.1 Plant height

The data related to the effect of different doses of the chemical mutagen ethyl methane sulfonate on the tolerance of the Chandler variety of strawberry under induced salinity stress conditions in plant height was presented in the (Table 4.6.1) In the pooled analysis, the maximum plant height was recorded in the chemical mutagen doses of 1% EMS induced with 75 mM salinity concentration (20.96 cm), which was statistically at par with 2% EMS dose. However, minimum height was observed in runners against 75 mM NaCl (16.53 cm). This trend was consistently noted in 2024 mutagen treated runners showed maximum plant height under the 50 mM NaCl salinity level (20.66 cm) and minimum plant height was found in runners induced with 75 mM salinity condition (16.53 cm).

Table 4.6.1: Interaction effect of ethyl methane sulphonate doses under different levels of salinity on plant height and chlorophyll of strawberry cv. Chandler

Treatment	Plant height 2023	Plant height 2024	Plant height pooled data	Chlorophyll 2023	Chlorophyll 2024	Chlorophyll pooled data	Treatment	Plant height 2023	Plant height 2024	Plant height pooled data	Chlorophyll 2023	Chlorophyll 2024	Chlorophyll pooled data				
1% EMS X Control	21.2	20.43	20.81	59.28	54.94	57.11	Control	18.66	15.43	17.05	45.41	40.27	42.84				
1% EMS X 50 mM NaCl	20.56	20.66	20.61	61.94	62.56	62.25	Control X 50 mM NaCl	19.12	16.86	17.95	46.39	40.5	43.44				
1% EMS X 75 mM NaCl	22.6	19.33	20.96	58.31	67.96	63.13	Control X 75 mM NaCl	18.11	15	16.53	40.15	42.33	41.24				
1% EMS X 100 mM NaCl	19.2	17.83	18.51	61.75	61.22	61.48	Control X 100 mM NaCl	17.69	20.43	19.06	40.61	40.83	40.72				
2% EMS X Control	19.9	18.9	19.4	61.04	63.79	62.41	C.D. for Years	0.375	1.75								
2% EMS X 50 mM NaCl	20.83	19.86	20.35	61.63	60.97	61.3	C.D. for Treatment	2.964	7.55								
2% EMS X 75 mM NaCl	22.93	18	20.46	65.40	60.64	63.02	C.D. for Years X Treatments	1.539	3.69								
2% EMS X 100 mM NaCl	22.36	17.8	20.08	64.52	66.17	65.35											
3% EMS X Control	18.7	15.36	17.03	48.46	52.48	50.47											
3% EMS X 50 mM NaCl	19.13	18.73	18.93	46.90	51.13	49.01											
3% EMS X 75 mM NaCl	20.36	17.3	18.83	44.27	50.3	47.28											
3% EMS X 100 mM NaCl	19.66	18.76	19.21	41.45	49.77	45.61											

However, in 2023 plant height was found maximum in EMS treated runners with 2% EMS dose induced with 75 mM NaCl (22.93 cm). as well as minimum plant height recorded in runners induced with 100 mM salinity condition (17.69 cm).

4.6.2 Chlorophyll content index

In the Chandler variety of strawberry, chlorophyll content index value was measured over two years in runners treated with the chemical mutagen EMS combined with varying levels of salinity to assess their tolerance (Table 4.6.1) In the pooled analysis, the highest chlorophyll content index was recorded found in runners treated with 2% EMS dose exposed to 100 mM salinity level (65.35). which was statistically at par with 1 % EMS dose, while the lowest was observed in runners exposed to 100 mM salinity condition (40.72). A similar trend was observed in 2023 the maximum chlorophyll content index was recorded in plants treated with 2% EMS dose induced with 75 mM salinity level (65.40), while the minimum chlorophyll content index was observed in runners exposed to 75 mM salinity condition (40.15). However, in 2024 the highest chlorophyll content index was observed in the combination of 1% EMS dose induced with 75 mM NaCl (67.96). The minimum chlorophyll content index was recorded in without mutagen treated plant plants exposed to 50 mM NaCl (40.5).

4.6.3 Number of buds

It is evident from the pooled data that the number of buds was significantly influenced by the combined effects of different chemical mutagen EMS doses and varying levels of salinity on the tolerance of strawberry plants in the Chandler variety (Table 4.6.2). The maximum number of buds recorded was in plants treated with a 2% EMS dose exposed to 50 mM NaCl (28.16), while the minimum was noted in runners exposed to 100 mM salinity level (21.33). A similar pattern was observed during 2023 treated runners showed maximum buds under 75 mM NaCl (34.33). The minimum buds were recorded in runners induced with 50 mM salinity level (24.33). However, in 2024 season, the highest bud count was observed in the combination of a 1% EMS dose exposed 50 mM NaCl (21.13), whereas the minimum was recorded in runners exposed to 100 mM salinity level (14.59).

4.6.4 Number of flowers

After counting the buds, the number of initiated flowers was documented in the Chandler variety to assess the impact of varying EMS chemical mutagen doses and their tolerance to

Table 4.6.2 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on number of buds and flowers of strawberry cv. Chandler

Treatment	Numbe r of bud 2023	Numbe r of bud 2024	Number of bud pooled data	Number of flowers 2023	Number of flowers 2024	Number of flowers pooled data	Treatment	Numbe r of bud 2023	Numbe r of bud 2024	Number of bud pooled data	Number of flowers 2023	Number of flowers 2024	Number of flowers pooled data				
1% EMS X Control	31.66	21.33	26.5	30.33	22.33	26.33	Control	26.66	19	22.83	27.10	21.31	24.20				
1% EMS X 50 mM NaCl	32.66	21.19	27.76	31.66	21.66	26.66	Control X 50 mM NaCl	24.33	22	23.16	23.49	21.32	22.40				
1% EMS X 75 mM NaCl	34.33	20.19	27.26	33.21	21.76	27.76	Control X 75 mM NaCl	25.33	19.61	22.49	26.59	20.39	23.49				
1% EMS X 100 mM NaCl	28.33	24.13	26.23	29	25.13	27.06	Control X 100 mM NaCl	28	14.59	21.33	27.66	15.49	21.57				
2% EMS X Control	27	20	23.5	28.66	20.67	24.66	C.D. for Years	0.61	0.12								
2% EMS X 50 mM NaCl	33	23.33	28.16	32	23.33	27.66	C.D. for Treatment	2.99	3.34								
2% EMS X 75 mM NaCl	28.33	21.46	24.89	30.66	20	25.33	C.D. for Years X Treatments	2.98	3.13								
2% EMS X 100 mM NaCl	29.33	18.33	23.83	28.67	19.33	24											
3% EMS X Control	27.66	20.66	24.16	27.68	21.56	24.62											
3% EMS X 50 mM NaCl	30.16	20.11	25.13	29.72	23.19	25.66											
3% EMS X 75 mM NaCl	31.33	19.33	25.33	27.49	23.12	25.30											
3% EMS X 100 mM NaCl	27.66	19	23.33	26.53	20.33	23.43											

induced salinity stress (Table 4.6.2). In the pooled analysis, the highest number of flower was observed in the treatment of 2% EMS combined with 50 mM NaCl (27.66), while the lowest number of flowers occurred in the runners with 100 mM NaCl (21.57). In 2023, the maximum flower count was recorded under the treatment of 1% EMS dose exposed with 100 mM salinity stress (33.21). The minimum was recorded in runners induced with 50 mM salinity stress (23.49). A similar trend was recorded in 2024.

4.6.5 Number of fruits

The pooled data shows that the number of fruits was significantly affected by the combined influence of varying EMS concentrations and different salinity levels on the tolerance of Chandler strawberry plants (Table 4.6.3) In the pooled analysis, the maximum number of fruits recorded in chemical mutagen 2% EMS dose treated runners induced with 75 mM salinity stress (31.46). In contrast, the minimum number of fruits was recorded in runners induced with 100 mM salinity level (23.83). A similar trend was observed in 2024 the treated runners showed the maximum number of fruits recorded under 75 mM salinity level (28.10).

While the minimum number of fruits found in runners induced with 100 mM salinity concentration (18.66). In 2023 the number of fruits was recorded as maximum in chemical mutagen EMS treated runners with 1% dose induced with 75 mM NaCl (35.79). However minimum fruit was recorded in runners induced with 75 mM NaCl (23.83).

4.6.6 Days to bud

In the Chandler variety of strawberry, the days taken for bud initiation were assessed in plants treated with the chemical mutagen EMS, focusing on their tolerance to induced salinity stress conditions (Table 4.6.3) The pooled analysis revealed that the earliest bud initiation occurred in runners treated with 2% EMS dose induced with 50 mM salinity level (48.97), While the late bud initiation was observed in without mutagen treated runners against 100 mM salinity level (61). Moreover, during the 2024, early bud initiation was recorded in runners treated with 1% EMS dose exposed to 50 mM salinity level (43),

However, late bud initiation was recorded in runners induced with 75 mM salinity level (61.33). A similar trend was recorded during 2024 treated runners showed early bud initiation under 100 mM salinity level (48.39) and late bud initiation was recorded under the 50 mM salinity level (62).

Table 4.6.3: Interaction effect of ethyl methane sulphonate doses under different levels of salinity on number of fruits and days to bud of strawberry cv. Chandler

Treatment	No. of fruits 2023	No. of fruits 2024	No. of fruits pooled data	Days to bud 2023	Days to bud 2024	Days to bud pooled data	Treatment	No. of fruits 2023	No. of fruits 2024	No. of fruits pooled data	Days to bud 2023	Days to bud 2024	Days to bud pooled data
1% EMS X Control	32.19	23.17	27.68	44.33	60	52.16	Control	30.33	21.33	25.83	52.33	61.33	56.83
1% EMS X 50 mM NaCl	33.10	24.22	28.66	43	59	51	Control X 50 mM NaCl	25.53	26.21	25.78	59.33	62	60.66
1% EMS X 75 mM NaCl	35.79	22.34	29.06	49.33	56.33	52.83	Control X 75 mM NaCl	28	21	24.5	61.33	55	58.16
1% EMS X 100 mM NaCl	28.10	25.34	26.72	59.43	48.39	56.27	Control X 100 mM NaCl	29	18.66	23.83	61	61	61
2% EMS X Control	30	22.66	26.33	59.33	53	56.16	C.D. for Years	0.25	0.89				
2% EMS X 50 mM NaCl	35.29	27.63	31.46	47.29	50.66	48.97	C.D. for Treatment	3.29	6.69				
2% EMS X 75 mM NaCl	30.66	28.10	29.38	51	55	53	C.D. for Years X Treatments	2.62	9.88				
2% EMS X 100 mM NaCl	29	20.33	24.66	51.33	52	51.66							
3% EMS X Control	31	26	28.5	46	63	54.5							
3% EMS X 50 mM NaCl	32	25	28.5	57.66	58	57.83							
3% EMS X 75 mM NaCl	31	19.33	25.16	56	57	56.5							
3% EMS X 100 mM NaCl	29.66	19	24.33	52.33	55.66	54							

4.6.7 Days to flowers

After counting the days to bud initiation, the days to flower initiation were recorded for the Chandler variety of strawberry in chemical mutagen treated runners to assess salinity tolerance over a two year experiment (Table 4.6.4) In the pooled analysis, early flower initiation occurred in the treatment with 2% EMS dose treated runners exposed with 50 mM NaCl (62.70), while late flower initiation was observed in runners induced with 100 mM NaCl (78.83).

However, in 2023 early flower initiation was recorded in chemical mutagen EMS treated runners with 1% dose induced with 50 mM salinity level and late flower initiation was found in runners exposed with 75 mM NaCl (72.10). A similar trend was observed during 2024 treated runners showed early flower initiation under the 100 mM salinity level (63), while the late flower initiation was observed in runners exposed to 50 mM NaCl (78.10).

4.6.8 Days to fruits

After counting the days for bud and flower initiation, we recorded the fruit initiation days in the Chandler variety of strawberry, specifically in plants treated with chemical mutagens, to assess their tolerance to salinity over two years of experimentation (Table 4.6.4). In the pooled analysis, early fruit initiation was observed at an average day in the runners treated with 2% EMS dose against 50 mM NaCl (90.21), while late fruit initiation was noted in runners exposed with 100 mM NaCl (104.39). However, during 2023 early fruit initiation recorded in 1% EMS dose exposed with 50 mM salinity level (87.63), as well as late fruit initiation recorded in runners induced with 100 mM salinity level (73.87). Similar trend was found during 2024 fruit initiation was recorded early in EMS treated runners with 1% dose (90.36) under 100 mM salinity level. Fruit initiation recorded late in runners induced with 50 mM salinity level (110.21).

4.6.9 Fruit length

In Chandler, a variety of strawberry fruit lengths was recorded in plants treated with the chemical mutagen EMS to assess their tolerance to induced salinity stress conditions (Table 4.6.5) In the pooled analysis, the maximum fruit length was observed at in the treatment of 1% EMS dose exposed with 100 mM NaCl (41.15 mm) Illustrate Figure , while the minimum fruit length was recorded in runners exposed with 100 mM NaCl (28.88 mm). A Similar trend was recorded during 2024 the treated runners showed maximum fruit length in the treatment

Table 4.6.4 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on days to flower and fruits of strawberry cv. Chandler

Treatment	Days to flowers 2023	Days to flowers 2024	Days to flower pooled data	Days to fruits 2023	Days to fruits 2024	Days to fruit pooled data	Treatment	Days to flowers 2023	Days to flowers 2024	Days to flower pooled data	Days to fruits 2023	Days to fruits 2024	Days to fruit pooled data
1% EMS X Control	68.66	68.33	68.5	90.12	92.17	91.14	Control	68.33	75.33	71.83	96.36	99.21	97.78
1% EMS X 50 mM NaCl	54.66	80	67.33	87.63	93.64	90.63	Control X 50 mM NaCl	67.11	78.10	72.60	97.35	110.21	103.78
1% EMS X 75 mM NaCl	68.29	70.13	69.21	88.23	93.19	90.71	Control X 75 mM NaCl	72.10	74.6	73.35	102.43	98.69	100.56
1% EMS X 100 mM NaCl	64.13	63	63.56	90.39	90.36	90.37	Control X 100 mM NaCl	70	77.66	73.83	99.68	109	104.39
2% EMS X Control	71.11	67	69.05	92.33	95.10	93.71	C.D. for Years	2.09	2.45				
2% EMS X 50 mM NaCl	63	62.41	62.70	89.32	91.10	90.21	C.D. for Treatment	16.38	16.17				
2% EMS X 75 mM NaCl	71.10	72.39	71.74	90.37	93.66	92.01	C.D. for Years X Treatments	7.54	6.24				
2% EMS X 100 mM NaCl	60.33	79.67	70	89.79	93.76	91.77							
3% EMS X Control	67.66	70.33	68.99	90.68	94.33	92.50							
3% EMS X 50 mM NaCl	63.33	69.33	66.33	91.69	91.79	91.74							
3% EMS X 75 mM NaCl	70.21	71.22	70.71	94.63	96.79	95.71							
3% EMS X 100 mM NaCl	68.53	70.19	69.36	97.66	99.10	98.38							

of 1% EMS dose exposed with 50 mM NaCl (42.10 mm) induced with 50 mM NaCl stress, with the minimum fruit length recorded in runners induced with 50 mM salinity level (26.76 mm). In 2023, the maximum fruit length was recorded at in the treatment of 2% EMS against 50 mM salinity level (43.58 mm), whereas the minimum fruit length was recorded in runners exposed with 75 mM NaCl (28.74 mm).

4.6.10 Fruit diameter

The pertaining data regarding the effect of different doses of the chemical mutagen EMS on the tolerance of the Chandler variety of strawberry to induced salinity stress conditions are presented in (Table 4.6.5). In the pooled analysis, the maximum fruit diameter was recorded in runners induced with 1% EMS dose under 100 mM NaCl (29.64 mm) Illustrate Figure, while the minimum fruit diameter was noted in without mutagen treated runners against 100 mM NaCl (24.83 mm). In the 2024/23, the maximum fruit diameter reached highest under the influence of 2% EMS dose induced with 50 mM NaCl salinity level (33.56 mm). The minimum diameter was consistently recorded in runners under 75 mM NaCl salinity stress (28.74 mm). A similar trend was found in 2024 EMS treated runners with 2% EMS dose under 0 mM salinity level (25.17 mm). However minimum fruit diameter recorded in runners induced with (18.51 mm).

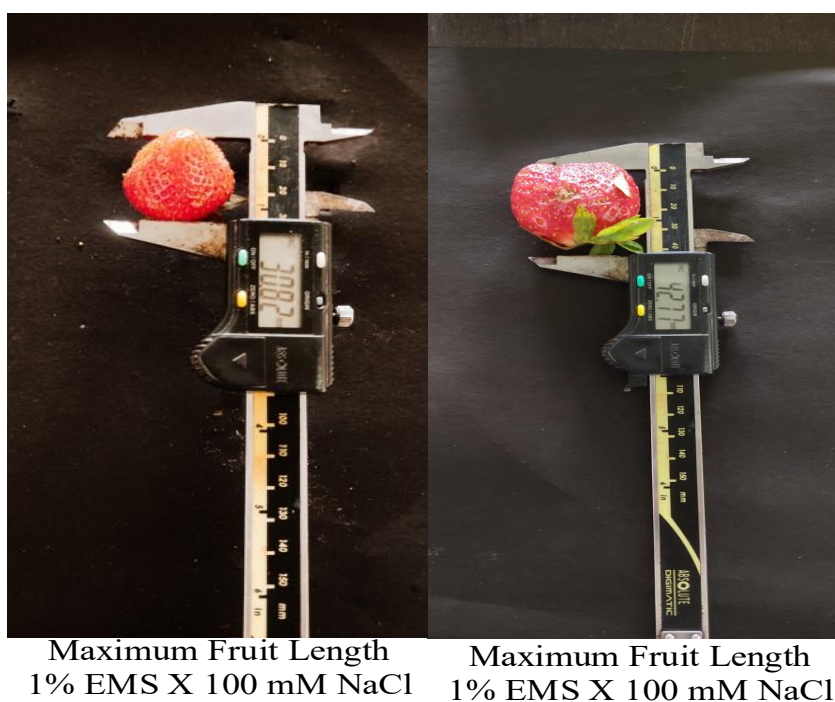


Plate 12. Fruit length and diameter in Chandler variety

Table 4.6.5 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on fruit length and fruit diameter of strawberry cv. Chandler

Treatment	Fruit length 2023	Fruit length 2024	Fruit length pooled data	Fruit diameter 2023	Fruit diameter 2024	Fruit diameter pooled data	Treatment	Fruit length 2023	Fruit length 2024	Fruit length pooled data	Fruit diameter 2023	Fruit diameter 2024	Fruit diameter pooled data
1% EMS X Control	38.51	33.63	36.07	32.17	26.84	29.50	Control	40.10	30.85	35.47	32.12	21.42	26.77
1% EMS X 50 mM NaCl	39.54	42.10	40.82	30.8	20.59	25.73	Control X 50 mM NaCl	35.11	26.76	30.93	31.42	18.51	24.96
1% EMS X 75 mM NaCl	39.73	37.59	38.66	32.13	22.75	27.44	Control X 75 mM NaCl	28.74	29.33	29.03	27.79	26.43	27.11
1% EMS X 100 mM NaCl	41.17	41.14	41.15	33.79	25.50	29.64	Control X 100 mM NaCl	30.22	27.54	28.88	29.78	19.88	24.83
2% EMS X Control	39.7	35.92	37.81	32.12	25.71	28.91	C.D. for Years	0.90	1.61				
2% EMS X 50 mM NaCl	43.58	32.16	37.87	33.56	25.17	29.36	C.D. for Treatment	6.61	3.05				
2% EMS X 75 mM NaCl	38.97	34.11	36.51	31.83	22.56	27.19	C.D. for Years X Treatments	2.44	1.42				
2% EMS X 100 mM NaCl	39.65	31.77	35.71	30.69	21.39	26.04							
3% EMS X Control	39.28	31.59	35.43	31.17	22.50	26.83							
3% EMS X 50 mM NaCl	39.79	37.44	38.62	32.76	24.51	28.63							
3% EMS X 75 mM NaCl	39.38	30.44	34.91	31.83	20.84	26.33							
3% EMS X 100 mM NaCl	38.66	30.11	34.38	33.19	23.45	28.23							

4.6.11 Shoot fresh weight

The data regarding the effect of different doses of the chemical mutagen EMS on the tolerance of the Chandler variety of strawberry under induced salinity stress, focusing on shoot fresh weight, are presented in the (Table 4.6.6). The study evaluated the interaction between EMS doses and varying levels of NaCl to determine their impact on plant growth under stress conditions.

In the pooled analysis, the maximum shoot fresh weight was recorded in the 2% EMS dose induced with 50 mM salinity stress (14.79 gm), while the minimum the minimum was recorded in runners exposed with 100 mM salinity stress treatment (11.21 gm). A similar trend was observed during the 2023 growing season, indicating that the treated runners showed maximum shoot fresh weight found under 50 mM salinity condition (17.37 gm). The minimum shoot fresh weight found in runners induced with 75 mM NaCl (10.79 gm).

However in 2024, the maximum shoot fresh weight was recorded in the 1% EMS dose against 50 mM NaCl treatment (13.18 gm), with the minimum being recorded in the without mutagen treated runners against 50 mM NaCl treatment (9.56 gm). This data suggests that EMS plays a crucial role in enhancing salinity tolerance in strawberries, with varying doses having differential effects depending on the level of salinity stress.

4.6.12 Shoot dry weigh

The interaction effect of the chemical mutagen EMS and induced salinity stress (NaCl) on shoot dry weight is presented in (Table 4.6.6) The shoot dry weight was measured after recording the fresh weight of the shoots in the Chandler variety of strawberry. In a pooled analysis, the highest shoot dry weight was observed in the treatment of 2% EMS dose exposed with 50 mM NaCl (3.76 gm), while the lowest dry weight was recorded in the runners induced with 100 mM NaCl (2.29 gm). A similar trend was observed in 2023 treated runners showed maximum shoot dry weight (4.57 gm) recorded under the 50 mM NaCl (4.57 gm) and minimum dry weight was found in runners induced with 75 mM NaCl (2.86 gm).

However, in 2024, the maximum shoot dry weight was recorded in the treatment of 1% EMS exposed to 50 mM NaCl (2.93 gm), while the minimum was found in the runners induced with 100 mM NaCl (1.93 gm). This suggests that salinity stress significantly affect shoot dry weight in strawberries, with reduced dry weight under higher salinity stress, particularly in 2% EMS dose.

Table 4.6.6 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on shoot fresh weight and dry weight of strawberry cv. Chandler

Treatment	Shoot fresh wt. 2023	Shoot fresh wt. 2024	Shoot fresh wt. pooled data	Shoot dry wt. 2023	Shoot dry wt. 2024	Shoot dry wt. pooled data	Treatment	Shoot fresh wt. 2023	Shoot fresh wt. 2024	Shoot fresh wt. pooled data	Shoot dry wt. 2023	Shoot dry wt. 2024	Shoot dry wt. pooled data	
1% EMS X Control	14.37	13.2	13.78	3.28	2.84	3.06	Control	13.85	10.91	12.38	4.22	1.54	2.88	
1% EMS X 50 mM NaCl	15.85	13.18	14.51	4.21	2.93	3.57	Control X 50 mM NaCl	13.29	9.56	11.42	4.57	1.67	3.12	
1% EMS X 75 mM NaCl	13.10	11.73	12.41	3.12	1.89	2.50	Control X 75 mM NaCl	10.79	14.13	12.46	2.86	1.89	2.37	
1% EMS X 100 mM NaCl	12.36	10.64	11.50	3.11	1.74	2.42	Control X 100 mM NaCl	12.30	10.13	11.21	3.19	1.39	2.29	
2% EMS X Control	13.4	11.87	12.63	3.42	2.75	3.09	C.D. for Years	0.13	0.33					
2% EMS X 50 mM NaCl	17.37	12.22	14.79	4.57	2.96	3.76	C.D. for Treatment	2.86	1.39					
2% EMS X 75 mM NaCl	14.97	10.85	12.91	3.54	2.40	2.97	C.D. for Years X Treatments	1.13	0.71					
2% EMS X 100 mM NaCl	12.92	10.57	11.74	3.40	2.18	2.79								
3% EMS X Control	14.37	10.66	12.51	3.25	1.86	2.55								
3% EMS X 50 mM NaCl	16.88	10.74	13.81	4.17	1.96	3.06								
3% EMS X 75 mM NaCl	14.44	10.15	12.29	3.45	1.74	2.59								
3% EMS X 100 mM NaCl	12.88	11.31	12.09	3.84	1.46	2.65								

4.6.13 Root fresh weight

The data pertaining to the effect of different EMS doses on root fresh weight in the Chandler strawberry variety under salinity stress conditions are provided in the (Table 4.6.7). The pooled analysis showed that the maximum root fresh weight was observed in plants treated with 2% EMS exposed to 100 mM NaCl (16.59 gm), while the minimum root fresh weight of was recorded in runners exposed to 100 mM NaCl (12.39 gm).

A similar trend was observed during 2023 the treated runners showed maximum root fresh weight in runners induced with 50 mM NaCl (17.33 gm) also the minimum root fresh weight was found in runners induced against 75 mM NaCl (10.25 gm).

However, in 2024, the highest root fresh weight was recorded in plants treated with 1% EMS exposed to 50 mM NaCl (15.35 gm), while the lowest root fresh weight was recorded in control plants exposed to 75 mM NaCl (10.25 gm).

4.6.14 Root dry weight

The root dry weight of EMS treated plants was measured after weighing the fresh root weight to assess their tolerance to induced salinity stress (NaCl) in the Chandler variety of strawberry. The results are presented in (Table 4.6.7) In the pooled analysis, the maximum root dry weight was observed in the treatment with 2% EMS induced with 100 mM NaCl (4.25 gm), while the minimum root dry weight was recorded in 100 mM salinity level (2.89 gm). A similar trend was noted during the 2023 treated runners showed maximum root dry weight found under the 50 mM NaCl (6.18 gm) and minimum weight found under the 75 mM NaCl (1.70 gm).

In 2024, the maximum root dry weight was found in the treatment with 1% EMS induced with 50 mM NaCl (4.24 gm), and the minimum root dry weight was recorded in runners exposed with 75 mM NaCl (1.70 gm). These findings suggest that different EMS concentrations, when applied under varying salinity levels, significantly impact the root dry weight of the Chandler strawberry, indicating enhanced salinity tolerance in EMS treated plants.

4.6.15 Number of leaves

Table 4.6.7 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on root fresh weight and dry weight of strawberry cv. Chandler

Treatment	Root fresh wt. 2023	Root fresh wt. 2024	Root fresh wt. pooled data	Root dry wt. 2023	Root dry wt. 2024	Root dry wt. pooled data	Treatment	Root fresh wt. 2023	Root fresh wt. 2024	Root fresh wt. pooled data	Root dry wt. 2023	Root dry wt. 2024	Root dry wt. poole d data
1% EMS X Control	16.69	10.1	13.42	6.13	3.14	4.63	Control	16.24	9.95	13.16	4.14	1.74	3.94
1% EMS X 50 mM NaCl	16.25	15.35	15.80	5.62	4.24	4.93	Control X 50 mM NaCl	15.31	9.89	12.60	3.18	2.96	3.50
1% EMS X 75 mM NaCl	15.26	13.80	14.53	5.17	3.92	4.54	Control X 75 mM NaCl	10.25	15.92	13.08	4.12	1.70	2.29
1% EMS X 100 mM NaCl	14.98	12.23	13.61	5.15	3.78	4.46	Control X 100 mM NaCl	15.81	8.97	12.39	3.49	2.29	2.89
2% EMS X Control	15.87	13.43	14.65	5.94	2.83	4.38	C.D. for Years	0.54	0.24				
2% EMS X 50 mM NaCl	17.33	11.41	14.37	6.18	4.12	5.15	C.D. for Treatment	3.93	2.23				
2% EMS X 75 mM NaCl	15.97	10.14	13.05	5.77	2.95	4.36	C.D. for Years X Treatments	1.32	0.78				
2% EMS X 100 mM NaCl	17.24	15.94	16.59	4.38	4.12	4.25							
3% EMS X Control	16.95	11.69	14.32	4.41	3.52	3.52							
3% EMS X 50 mM NaCl	17.18	11.88	14.53	4.49	3.22	3.88							
3% EMS X 75 mM NaCl	16.88	10.76	13.82	5.10	3.11	4.10							
3% EMS X 100 mM NaCl	15.25	10.60	12.93	5.07	2.68	3.38							

The analysis of the data reveals that different concentrations of the chemical mutagen EMS combined with varying levels of salinity significantly influenced the number of leaves in the Chandler variety of strawberry (Table 4.6.8). In the pooled analysis, the maximum number of leaves was recorded in plants treated with 1% EMS dose induced with 50 mM NaCl (13.5), while the minimum number of leaves was observed in plants exposed to 100 mM NaCl (7.66). A similar pattern was observed during 2024, the maximum number of leaves was recorded in plants treated with 1% EMS combined with 50 mM NaCl (14), while the minimum number leaves was noted in plants under 50 mM NaCl (9.67). In 2023, the maximum number of leaves was recorded in runners treated with 2% EMS induced with 50 mM NaCl (14), whereas the minimum leaves occurred in plants exposed to 75 mM NaCl (6.78). This result suggests that plants treated with 1% EMS dose had a better tolerance to salinity stress, shows a higher number of leaves under stress conditions.

4.6.16 Leaf relative water content

The analysis of the data reveals that different concentrations of EMS combined with varying levels of salinity significantly influenced the leaf relative water content (LRWC) of the Chandler variety of strawberry (Table 4.6.8). In the pooled analysis, the maximum LRWC was recorded in plants treated with 2% EMS with 50 mM NaCl (86.63 %), while the minimum LRWC was observed in plants exposed to 100 mM NaCl (77.26 %). During In 2024 similar trend was observed the maximum LRWC was recorded in plants treated with 2% EMS dose exposed with 75 mM NaCl (88.44 %), while the minimum LRWC of was noted in plants under 75 mM NaCl conditions (73.69 %). In 2023, the maximum LRWC was recorded in plants treated with 1% EMS dose under 50 mM NaCl (86.61 %), whereas the minimum LRWC occurred in plants exposed to 50 mM NaCl (79.67 %). This result suggests that plants treated with 2% EMS exhibited greater tolerance to salinity and maintained higher LRWC under stress conditions.

4.6.17 Electrolyte leakage

Electrolyte leakage was recorded during both years of the experiment in Chandler strawberry plants treated with the chemical mutagen EMS and exposed to different salinity levels using NaCl doses. This was done to assess their tolerance to salinity stress (Table 4.6.9). In the pooled analysis, the minimum electrolyte leakage was observed in runners treated with 1% EMS dose induced with 50 mM salinity level (20.97 %), while the maximum electrolyte leakage was recorded in the runners exposed to 75 mM NaCl (41.12%).

Table 4.6.8 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on number of leaves and leaf relative water content of strawberry cv. Chandler

Treatment	No. of leaves 2023	No. of leaves 2024	No. of leaves pooled data	LRWC 2023	LRWC 2024	LRWC pooled data	Treatment	No. of leaves 2023	No. of leaves 2024	No. of leaves pooled data	LRWC 2023	LRWC 2024	LRWC pooled data
1% EMS X Control	12	13.66	12.83	82.65	84.45	83.55	Control	8.33	9	8.66	84.67	78.99	81.83
1% EMS X 50 mM NaCl	13	14	13.5	86.61	84.98	85.79	Control X 50 mM NaCl	9	9.67	9.33	79.67	76.49	78.09
1% EMS X 75 mM NaCl	10	11.33	10.66	84.27	83.51	83.89	Control X 75 mM NaCl	6.78	9.33	8.05	82.55	73.69	78.12
1% EMS X 100 mM NaCl	9.33	10.33	9.83	83.93	81.73	82.83	Control X 100 mM NaCl	7	8.33	7.66	80.59	74.22	77.26
2% EMS X Control	11.33	11.66	11.5	82.42	82.64	82.53	C.D. for Years	0.37	1.05				
2% EMS X 50 mM NaCl	14	11	12.5	86.16	87.10	86.63	C.D. for Treatment	0.98	8.29				
2% EMS X 75 mM NaCl	9.66	11	10.33	81.10	88.44	84.77	C.D. for Years X Treatments	1.80	2.42				
2% EMS X 100 mM NaCl	9	9.33	9.16	85.63	78.73	82.18							
3% EMS X Control	9	10	9.5	81.4	79.50	80.45							
3% EMS X 50 mM NaCl	8.33	10.33	9.33	80.54	80.21	80.38							
3% EMS X 75 mM NaCl	8	9.66	8.83	82.85	77.11	79.96							
3% EMS X 100 mM NaCl	8	8.33	8.16	80.86	71.39	76.13							

However, in 2023 the electrolyte leakage was found minimum in runners treated with 2% EMS dose under the 50 mM NaCl (17.85 %) and maximum was observed in runners induced with 75 mM NaCl (41.12 %). In 2024 treated runners showed minimum electrolyte leakage observed in 2% EMS dose induced with 100 mM NaCl (18.25 %) and maximum was found runners exposed 75 mM NaCl (41.12 %). These findings suggest that the 1% EMS treatment may enhance salinity tolerance in Chandler strawberries by reducing electrolyte leakage under salinity stress conditions.

4.6.18 Proline content

The proline content was estimated in strawberry plants Chandler variety subjected to chemical mutagen treatment with EMS combined with induced salinity stress from different concentrations of sodium chloride. The experiment was conducted over two years, and the proline levels were recorded (Table 4.6.9). In the pooled analysis, the maximum proline content recorded in runners treated with 1% EMS dose with 75 mM NaCl (3.94 $\mu\text{mol/g}$), while the minimum was found in the runners exposed to 100 mM NaCl (1.50 $\mu\text{mol/g}$). A similar trend was observed in 2024, the proline content reached maximum in treated runners induce with 100 mM NaCl (4.86 $\mu\text{mol/g}$) and the minimum was found in the runners exposed to 100 mM NaCl (1.37 $\mu\text{mol/g}$). During 2023, the highest proline content was observed in runners treated with 2% EMS dose exposed to 75 mM NaCl (3.75 $\mu\text{mol/g}$), while the lowest was recorded in the plant with 50 mM NaCl (1.36 $\mu\text{mol/g}$). This suggests that 1% EMS treatment exposed with salinity stress increases proline accumulation in strawberry plants, with higher concentration NaCl generally leading to greater proline content.

4.6.19 Membrane stability Index

The membrane stability index was analysed over two years in an experiment on the Chandler variety of strawberry, focusing on plants treated with ethyl methanesulfonate under induced salinity stress using varying NaCl concentrations, as shown in the (Table 4.6.10). In the pooled analysis, the maximum membrane stability index was observed in plants treated with 1% EMS dose exposed to 50 mM salinity level (85.65 %), while the minimum membrane stability index was recorded in runners exposed to 100 mM salinity condition (77.79 %). However, in 2023, the maximum membrane stability index was recorded in plants treated with 2% EMS dose induced with 75 mM NaCl (89.43 %), while the minimum maximum membrane stability index was observed in the runners exposed to 50 mM NaCl (77.54 %). A similar

Table 4.6.9 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on electrolyte leakage and proline content of strawberry cv. Chandler

Treatment	E L % 2023	E L % 2024	E L % pooled data	Proline content 2023	Proline content 2024	Proline content pooled data	Treatment	E L % 2023	E L % 2024	E L % pooled data	Proline content 2023	Proline content 2024	Proline content pooled data
1% EMS X Control	29.35	19.63	24.49	3.26	1.72	2.49	Control	37.56	31.66	34.61	3.45	1.91	2.68
1% EMS X 50 mM NaCl	26.15	18.61	22.38	2.30	3.24	2.77	Control X 50 mM NaCl	36.49	34.32	35.40	1.36	1.94	1.65
1% EMS X 75 mM NaCl	20.26	21.69	20.97	3.32	4.55	3.94	Control X 75 mM NaCl	41.12	23.40	32.25	2.77	2.13	2.45
1% EMS X 100 mM NaCl	26.12	19.19	22.60	1.39	4.86	3.12	Control X 100 mM NaCl	39.12	33.72	36.37	1.63	1.37	1.50
2% EMS X Control	21.77	20.24	21	1.46	2.28	1.87	C.D. for Years	1.63	0.23				
2% EMS X 50 mM NaCl	17.85	24.58	21.21	2.36	2.77	2.56	C.D. for Treatment	12.69	2.18				
2% EMS X 75 mM NaCl	37.8	19.21	28.50	3.75	1.83	2.79	C.D. for Years X Treatments	2.25	0.50				
2% EMS X 100 mM NaCl	25.26	18.25	21.75	2.42	2.39	2.41							
3% EMS X Control	23.32	19.13	21.23	1.57	1.92	1.75							
3% EMS X 50 mM NaCl	26.69	19.48	23.11	1.37	1.57	1.47							
3% EMS X 75 mM NaCl	28.35	19.20	23.86	1.77	2.1	1.93							
3% EMS X 100 mM NaCl	40.61	28.99	34.80	2.12	1.79	1.91							

trend was observed during 2024 the maximum membrane stability index was found in treated runners induced with 100 mM NaCl (84.42 %), while the minimum maximum membrane stability index was observed in runners exposed to 75 mM NaCl (77.39 %). These findings indicate that EMS treatments can improve membrane stability in strawberry plants, especially at certain NaCl concentrations. Further studies may help optimize EMS to enhance stress tolerance in strawberries under varying environmental conditions.

4.6.20 Leaf Scorching

Leaf scorching was recorded in the Chandler variety of strawberry plant treated with the chemical mutagen Ethyl Methane sulfonate (EMS) under induced salinity stress conditions presented in (Table 4.6.10) Illustrated in plate 12. In pooled analysis, the minimum leaf scorching was recorded in runners treated in 1% EMS dose under 0 mM salinity level (15.75 %). In contrast, the maximum leaf scorching was observed in runners induced with 100 mM salinity condition (27.25 %). A similar trend was observed during the 2023 growing season, the minimum leaf scorching was recorded in runners treated with 1% EMS dose exposed with 50 mM NaCl (12.13 %). Maximum leaf scorching was found in the runners induced with 75 mM salinity concentration (28.13 %).

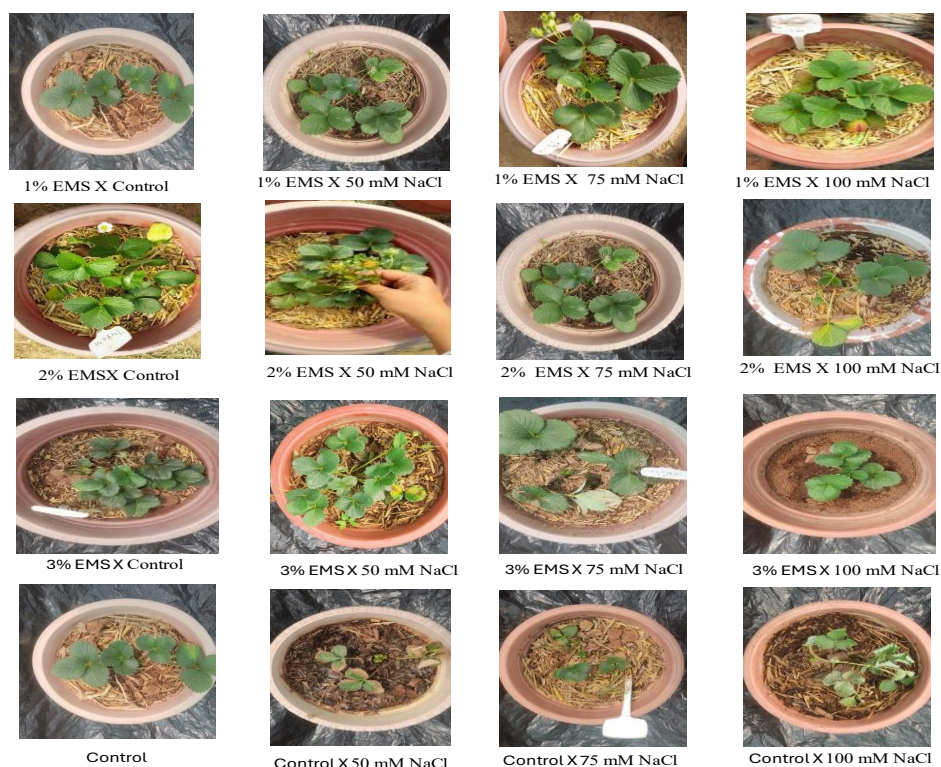
However, in 2024, leaf scorching was recorded as a minimum in runners treated with 2% EMS dose under 0 mM condition (15.84 %) and maximum leaf scorching was recorded in runners induced with 50 mM salinity condition treatments (27.32 %). The results obtained from this observation show that runners treated with 1% EMS dose have healthy leaves, instead of those treated with 3% EMS or control plants.

4.6.21 Fruit weight

The data in (Table 4.6.11) shows that fruit weight in the Chandler strawberry variety was significantly influenced by the interaction of chemical mutagen EMS treatments and varying levels of salinity stress. The pooled and individual results indicate that EMS treatment had effectively impact on fruit weight under salinity stress conditions. In the pooled analysis, the maximum fruit weight was recorded in plants treated with 1% EMS dose exposed to 50 mM salinity condition (21.85 gm) and minimum fruit weight was observed in runners exposed to 75 mM NaCl (9.97 gm). A similar trend was observed during the 2024 season, the treated runners showed maximum fruit weight was found under 75 mM NaCl (22.32 gm) and

Table 4.6.10 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on membrane stability index and leaf scorching of strawberry

Treatment	MSI 2023	MSI 2024	MSI pooled data	Leaf scorchin g 2023	Leaf scorchi ng 2024	Leaf scorchi ng pooled data	Treatment	MSI 2023	MSI 2024	MSI pooled data	Leaf scorchi ng 2023	Leaf scorchi ng 2024	Leaf scorchi ng pooled data
1% EMS X Control	87.25	76.35	81.80	15.16	15.99	15.57	Control	87.32	73.83	80.58	26.48	20.73	23.60
1% EMS X 50 mM NaCl	86.35	77.12	81.71	12.13	18.2	15.16	Control X 50 mM NaCl	77.54	79.82	78.68	23.19	27.32	25.25
1% EMS X 75 mM NaCl	86.17	78.73	82.45	13.38	20.17	16.77	Control X 75 mM NaCl	83.22	75.39	79.30	28.13	24.33	26.23
1% EMS X 100 mM NaCl	89.4	78.22	83.81	13.87	21.81	17.84	Control X 100 mM NaCl	79.25	76.37	77.79	27.86	26.65	27.25
2% EMS X Control	86.36	78.38	82.37	20.37	15.84	18.10	C.D. for Years	0.77	0.43				
2% EMS X 50 mM NaCl	87.88	83.42	85.65	18.74	17.66	18.20	C.D. for Treatment	6.69	7.70				
2% EMS X 75 mM NaCl	89.43	79.92	84.68	23.72	19.40	21.56	C.D. for Years X Treatments	2.2	1.57				
2% EMS X 100 mM NaCl	83.22	84.42	83.82	21.29	21.75	21.52							
3% EMS X Control	88.67	76.74	82.71	24.16	20.42	22.29							
3% EMS X 50 mM NaCl	86.16	79.3	82.73	24.64	21.33	22.99							
3% EMS X 75 mM NaCl	87.86	73.89	80.87	24.16	23.47	23.81							
3% EMS X 100 mM NaCl	86.44	73.27	79.85	25.53	25.19	25.36							



the minimum fruit weight was recorded in runners induced with 50 mM salinity condition (9.11 gm). However in 2023, the highest fruit weight was noted in EMS treated runners with 2 % dose exposed to 75 mM NaCl (22.65 gm) and the lowest fruit weight was recorded in plants under 100 mM NaCl 10.56 gm (10.56 gm). The maximum fruit weight was recorded in runners treated with 1% EMS dose, which tolerated salinity up to 100 mM NaCl, as compared to control runners and those treated with a 3% EMS dose.

4.6.22 Fruit Yield

The data in (Table 4.6.11) shows that fruit yield in EMS mutagen treated runners with 1% EMS and 2% EMS dose was not affected by salinity treatments in the Chandler strawberry variety. In the pooled analysis, the highest fruit yield was recorded in runners treated with a 2% EMS dose against 100 mM salinity condition (410.85 gm) and the lowest fruit yield of was observed in control runners exposed to 100 mM salinity level (176.87 gm). A similar trend was observed during 2023, maximum fruit yield was found in control runners under the 75 mM salinity level stress (415.44 gm). While the lowest yield was recorded in control plants exposed to 75 mM salinity level (181.39 gm). During the 2024 season, the highest yield was recorded in runners treated with a 1% EMS dose under 75 mM NaCl (416.10 gm) and the minimum yield was observed in control runners exposed to 100 mM NaCl (171.67 gm).

Table 4.6.11 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on fruits weight and fruit yield of strawberry cv. Chandler

Treatment	Fruit weight 2023	Fruit weight 2024	Fruit weight pooled data	Fruit yield 2023	Fruit yield 2024	Fruit yield pooled data	Treatment	Fruit weight 2023	Fruit weight 2024	Fruit weight pooled data	Fruit yield 2023	Fruit yield 2024	Fruit yield pooled data	
1% EMS X Control	19.96	18.66	19.31	382.93	368.58	375.75	Control	11.43	10.62	10.62	201.96	185.84	193.90	
1% EMS X 50 mM NaCl	21.78	21.92	21.85	291.24	299.20	295.22	Control X 50 mM NaCl	10.89	9.11	10	186.96	181.69	184.32	
1% EMS X 75 mM NaCl	17.57	22.32	19.94	415.44	385.95	400.69	Control X 75 mM NaCl	10.78	9.17	9.97	181.39	186.12	183.75	
1% EMS X 100 mM NaCl	18.18	18.09	18.13	408.33	413.38	410.85	Control X 100 mM NaCl	10.56	10.26	10.26	182.08	171.67	176.87	
2% EMS X Control	15.95	20.34	18.15	287.68	276.42	282.05	C.D. for Years	0.35	0.87					
2% EMS X 50 mM NaCl	19.03	17.72	18.38	263.98	274.33	269.15	C.D. for Treatment	3.31	5.27					
2% EMS X 75 mM NaCl	22.65	20.13	21.39	373.96	416.10	395.03	C.D. for Years X Treatments	2.16	6.63					
2% EMS X 100 mM NaCl	20.33	21.25	20.79	404.82	285.55	345.19								
3% EMS X Control	17.4	16.51	16.95	192.36	297.06	244.71								
3% EMS X 50 mM NaCl	17.12	15.37	16.24	189.73	210.14	199.93								
3% EMS X 75 mM NaCl	17.07	14.58	15.82	198.21	210.32	204.26								
3% EMS X 100 mM NaCl	16.59	12.58	14.58	196.10	221.19	208.64								

4.6.23 Malondialdehyde

The (Table 4.6.12) shows the interaction effect of different chemical mutagen EMS doses and check their tolerance against induced salinity NaCl stress conditions in Chandler variety of strawberry. In analysis, it was observed that the control runners and 3% EMS dose shows maximum malonaldehyde content instead of those treated with 1% EMS and 2% EMS dose. However, in pooled analysis results the minimum malonaldehyde content was found in runners treated with 1% EMS dose induced with 75 mM salinity level (1.53 $\mu\text{mol/g}$) and maximum malonaldehyde content was recorded in control runners induced with 100 mM NaCl (9.63 $\mu\text{mol/g}$). During 2023 malonaldehyde content was found minimum in mutagen treated runners with 2% EMS against 75 mM NaCl (1.29 $\mu\text{mol/g}$) and maximum malonaldehyde content was recorded in runners induced with 100 mM salinity level (10.6 $\mu\text{mol/g}$). A similar trend was found during 2024 malonaldehyde content was recorded as minimum in treated runners induced against 50 mM NaCl (1.12 $\mu\text{mol/g}$) and maximum malonaldehyde content was found in control runners induced with 50 mM salinity concentration (9.56 $\mu\text{mol/g}$).

4.6.24 Leaf Area

It is evident from the data of leaf area from gamma irradiation treated plant against induced salinity NaCl treatments in chandler variety of strawberry (Table 4.6.12). In chemical mutagen, the leaf area was maximum recorded in runners treated with 1% EMS and 2% EMS dose instead of control runners and 3% EMS dose-treated runners. In pooled analysis, leaf area was recorded maximum in runners treated with 2% EMS dose induced with 75 mM NaCl (25.41 mm) and minimum leaf area recorded in runners exposed 100 mm NaCl (9.09 mm). A similar trend was observed in 2024 leaf area was recorded maximum in treated runners with 2% EMS induced with 50 mM NaCl (28.10 mm) and the minimum leaf area was recorded in runners exposed to 50 mM NaCl (9.19 mm). However, during 2023 leaf area was maximum found in runners treated with 1% EMS dose against 50 mM NaCl (26.37 mm) and minimum was recorded in runners induced with 75 mM salinity condition (8.21 mm).

4.6.25 Mineral Analysis sodium

(Table 4.6.13) shows the estimated sodium content in strawberry plant treated with chemical mutagen EMS against induce salinity NaCl treatment. In the estimated sodium content analysis, plants treated with 2 % EMS doses absorbed the minimum sodium content compared to control plants and those treated with a 3 % EMS dose. In the interaction effect maximum

Table 4.6.12 Interaction effect of ethyl methane sulphonates doses under different levels of salinity on malonaldehyde content and leaf area index of strawberry cv. Chandler

Treatment	Malondialdehyde de 2023	Malondialdehyde de 2024	Malondialdehyde de pooled data	LAI 2023	LAI 2024	LAI pooled data	Treatment	Malondialdehyde 2023	Malondialdehyde 2024	Malondialdehyde de pooled data	LAI 2023	LAI 2024	LAI pooled data				
1% EMS X Control	2.96	3.86	3.41	19.64	20.23	19.93	Control	8.83	8.93	8.88	12.67	14.28	13.47				
1% EMS X 50 mM NaCl	2.43	3.58	3.01	26.37	21.42	23.89	Control X 50 mM NaCl	9.36	9.56	9.46	10.59	9.19	9.89				
1% EMS X 75 mM NaCl	1.34	1.73	1.53	24.34	21.56	22.94	Control X 75 mM NaCl	10.6	8.2	9.4	8.21	9.37	9.55				
1% EMS X 100 mM NaCl	5.43	6.03	5.73	22.78	23.12	22.95	Control X 100 mM NaCl	10	9.26	9.63	8.46	9.73	9.09				
2% EMS X Control	3.76	4.26	4.01	20.43	21.73	21.08	C.D. for Years	0.34	0.78								
2% EMS X 50 mM NaCl	2.52	1.12	1.82	20.76	28.10	24.43	C.D. for Treatment	0.97	6.39								
2% EMS X 75 mM NaCl	1.29	1.86	2.22	24.69	26.13	25.41	C.D. for Years X Treatments	0.96	6.46								
2% EMS X 100 mM NaCl	5.76	6.03	5.90	23.17	21.34	22.25											
3% EMS X Control	3.86	4.33	4.1	21.49	20.10	20.79											
3% EMS X 50 mM NaCl	2.39	3.19	2.79	15.37	17.39	16.38											
3% EMS X 75 mM NaCl	5.8	6.3	6.05	18.63	16.64	17.63											
3% EMS X 100 mM NaCl	7.03	6.8	6.91	19.76	17.49	18.62											

sodium content was recorded in runners against 100 mM NaCl (117.32 ppm) and minimum sodium was found in runners treated with 2% EMS exposed to 50 mM NaCl (19.02 ppm). However, during 2023 the estimated sodium was maximum recorded in runners against 50 mM NaCl (115.39 ppm) and minimum sodium was found in 1% EMS dose treated runners (18.49 ppm). In 2024 sodium was observed maximum in runners against 75 mM salinity level (127.17 ppm) and minimum sodium was recorded in runners treated with 3% EMS dose under 0 mM condition (16.17 ppm).

4.6.26 Mineral Analysis potassium

Potassium was estimated from chemical mutagen EMS treated runners against salinity NaCl stress during two years of experiment (Table 4.6.13) pooled data shows the maximum potassium recorded in plants treated with 2% EMS dose against 50 mM NaCl (19.85 ppm) and minimum potassium was found in runners against 100 mM NaCl (9.72 ppm) decline in potassium concentration and High sodium levels can inhibit potassium uptake. A similar trend was recorded during 2024 potassium was observed maximum in runners treated with 2% EMS dose with an induced salinity concentration 75 mM NaCl (21.37 ppm) and the minimum was recorded in runners induced with in 50 mM salinity level (8.69 ppm). During 2023 potassium was recorded maximum in runners treated with 1% EMS dose (20.33 ppm) under 75 mM NaCl and minimum potassium was found in runners against 75 mM NaCl (9.13 ppm).

Table 4.6.13 Interaction effect of ethyl methane sulphonate doses under different levels of salinity on sodium and potassium ion of strawberry cv. Chandler

Treatment	Sodium Na ppm 2023	Sodium Na ppm 2024	Sodium Na ppm pooled	Potassiu m K ppm 2023	Potassiu m K ppm 2024	Potassiu m K ppm pooled	Treatment	Sodium Na ppm 2023	Sodium Na ppm 2024	Sodium Na ppm pooled	Potassi um K ppm 2023	Potassi um K ppm 2024	Potassi um K ppm pooled				
1% EMS X Control	18.49	20.67	19.58	18.43	19.33	18.88	Control	21.34	22.10	21.72	19.63	19.21	19.42				
1% EMS X 50 mM NaCl	70.31	72.10	71.20	13.67	11.87	12.77	Control X 50 mM NaCl	115.39	96.52	105.95	12.37	8.69	10.53				
1% EMS X 75 mM NaCl	72.33	71.49	71.91	20.33	18.57	19.45	Control X 75 mM NaCl	98.43	127.17	112.81	9.13	13.29	11.21				
1% EMS X 100 mM NaCl	86.34	83.19	84.76	10.76	11.39	11.07	Control X 100 mM NaCl	111.45	123.19	117.32	9.78	9.66	9.72				
2% EMS X Control	22.58	24.44	23.51	13.65	14.21	13.93	C.D. for Years	2.56	1.79								
2% EMS X 50 mM NaCl	19.76	18.28	19.02	19.37	20.34	19.85	C.D. for Treatment	7.94	3.43								
2% EMS X 75 mM NaCl	74.55	79.17	74.86	17.78	21.37	19.57	C.D. for Years X Treatments	9.87	1.73								
2% EMS X 100 mM NaCl	84.67	83.63	84.15	12.39	11.25	11.82											
3% EMS X Control	23.48	16.17	19.82	19.37	18.23	18.8											
3% EMS X 50 mM NaCl	64.71	62.54	63.62	10.67	11.43	11.05											
3% EMS X 75 mM NaCl	78.33	76.46	77.39	15.33	16.43	15.88											
3% EMS X 100 mM NaCl	91.67	89.55	90.61	10.67	9.79	10.23											

Chapter – 5

DISCUSSION



DISCUSSION

5.1 Physical mutagen gamma irradiation against salinity stress

Plant height

The height of the plant is a significant morphological parameter linked to crop growth and production. Applying gamma irradiation at a dose of 30 Gy significantly improved the salt tolerance of strawberry plants, specifically in the Winter Dawn and Chandler varieties. This dose sustained plant height across all NaCl treatments, with the maximum plant height recorded in the Winter Dawn variety under 75 mM salinity. In the Chandler variety, the mutagen-treated runners exhibited maximum plant height under 50 mM salinity conditions. This was previously confirmed by Melo *et al.* (2019) in bananas, and Salwa *et al.* (2018) who demonstrated the best results in plant height under salinity conditions. A similar overestimation was observed by Dantas *et al.* (2018) in pomegranate and Nower (2018) in strawberry. The greatest reduction in plant height was observed at a higher dose of 40 Gy. This suggests that gamma irradiation can induce beneficial mutations that improve the physiological capacity of plants to tolerate saline conditions. Gamma rays are known to induce mutations by breaking DNA strands, which can result in a range of physiological and biochemical changes that sustain a plant tolerance to abiotic stresses such as salinity (Suprasanna *et al.* 2014; Katiyar *et al.* 2022).

This is consistent with the findings of several researchers who noted that the decrease in plant growth caused by salinity is often linked to reduced plastic extensibility of cell walls in the root and leaf expansion zones (Anderson, 2024). Munns *et al.* (2006) explained that salt in soil water inhibits plant growth for two primary reasons: first, the presence of salt in the soil decreases the plant's ability to absorb water, slowing growth, a phenomenon known as the osmotic effect of salinity. Second, high salt concentrations in the transpiration stream eventually damage the cells in the transpiring leaves, further impairing growth and photosynthesis. The increased plant height observed in runners treated with the 30 Gy dose, particularly under high NaCl concentrations, aligns with earlier studies where gamma irradiation enhanced growth parameters in banana crops under salinity stress conditions (Xie *et al.* 2023; Nwokuwu, 2021). This finding underscores the potential of gamma rays to enhance stress tolerance by boosting antioxidant enzyme activities and increasing osmolyte accumulation, which together help alleviate the adverse effects of salinity (Jalil and Ansari, 2020).

Shoot fresh weight and shoot dry weight

The present study demonstrates the effects of gamma irradiation under induced salinity stress on the shoot fresh and dry weights in strawberry varieties Winter Dawn and Chandler. Both parameters are crucial indicators of plant vigour and overall health, particularly under salinity stress conditions. In the pooled analysis, the interaction between salinity levels and gamma irradiation doses produced significant differences in shoot fresh and dry weights for both varieties.

In the Winter Dawn variety, plants exposed to a 20 Gy gamma radiation dose induced with 50 mM salinity conditions exhibited the highest shoot fresh weight. This suggests that a moderate dose of gamma irradiation under salinity stress may stimulate adaptive responses in the plant, resulting in improved growth. However, in the Chandler variety, the maximum shoot fresh weight was recorded in plants treated with a 30 Gy gamma radiation dose exposed with 50 mM NaCl. This indicates that higher doses of gamma radiation might be more effective in enhancing fresh weight in Chandler compared to Winter Dawn. Similar studies in okra have shown that gamma irradiation at optimal doses can improve antioxidant activity, thereby promoting growth under salinity stress Zaka *et al.* (2004). These results align with earlier research in papaya, which demonstrated that gamma radiation can trigger physiological and molecular responses that improve a plant's ability to withstand stress conditions like salinity Arbona *et al.* (2017).

A similar pattern was observed in the shoot dry weight analysis. In the Winter Dawn variety, the maximum dry weight was recorded in runners treated with 20 Gy gamma radiation under 50 mM salinity conditions. However, in the Chandler variety, the highest shoot dry weight was measured in runners treated with 30 Gy gamma radiation under 75 mM salinity conditions. This indicates that gamma radiation can enhance dry biomass production under salinity stress, possibly by altering water retention and nutrient uptake mechanisms in the sugarcane plant Mirajkar *et al.* (2019)

Root fresh weight and dry weight

The influence of gamma radiation and salinity stress on root fresh and dry weight in strawberry varieties Winter Dawn and Chandler was assessed. Specifically, in the Winter Dawn variety, the maximum root fresh weight was observed at a 30 Gy gamma radiation dose exposed with 75 mM salinity conditions. However, in the Chandler variety, pooled analysis showed that the maximum root fresh weight and dry weight was recorded in runners treated with 20 Gy gamma

radiation under 75 mM salinity conditions. The root dry weight results further support these findings. In the Winter Dawn variety, the maximum root dry weight was observed at a 20 Gy gamma radiation dose induced with 50 mM salinity conditions. This aligns with previous research on citrus, which illustrates the beneficial effects of physical mutagens like gamma radiation on plant growth and stress resilience. Studies have shown that gamma radiation can improve various physiological and biochemical traits in plants, including root development, particularly under abiotic stress conditions such as salinity (Nawaz *et al.* 2022; Hossain *et al.* 2021). Salinity stress generally reduces growth and biomass accumulation due to osmotic and ionic imbalances, but the combination of gamma radiation and moderate salinity appears to alleviate some of these negative effects, as observed in this study.

Fruit length and diameter

The effects of gamma radiation and NaCl salinity on fruit characteristics in two strawberry varieties, Winter Dawn and Chandler, were investigated. The pooled data values for fruit length in the Winter Dawn and Chandler variety indicated that the maximum fruit length was achieved with runners treated at a 20 Gy radiation dose under 75 mM NaCl salinity condition. However, the maximum fruit diameter was recorded in both Winter Dawn and Chandler varieties, specifically in runners treated with a 30 Gy radiation dose under 50 mM salinity conditions. The study by Saadati *et al.* (2022) supports this notion, indicating that gamma irradiation can influence the growth of fruits, including improvements in length and diameter in citrus. This supports findings from other studies demonstrating the detrimental effects of salinity on fruit development Unlukara *et al.* (2010). These findings align with previous research on strawberries, which indicates that salinity can severely affect plant growth and fruit quality Kavitha *et al.* (2015). The positive effects of moderate salinity on plant growth have been shown in various crops, suggesting that balanced salinity levels can improve growth under certain conditions (Munns and Tester, 2008). Consistent with the results observed in the Winter Dawn variety, the improvement in fruit quality through gamma radiation and regulated salinity levels aligns with findings from previous studies. These findings support the use of these treatments to improve yield and quality in crops exposed to stress conditions Farooq *et al.* (2013).

Fruit weight

The pooled data demonstrate significant variation in fruit weight in the Winter Dawn and Chandler strawberry varieties under the combined influence of gamma radiation exposed to

salinity stress. In the Winter Dawn variety, the maximum fruit weight was observed in runners treated with 20 Gy of gamma radiation under 50 mM salinity conditions. However, the highest fruit weight in Chandler was recorded in plants treated with 30 Gy of gamma radiation induce with 50 mM salinity conditions. Moreover, the findings regarding Chandler variety, where the highest fruit weight was recorded at 30 Gy of gamma radiation under the same salinity conditions, are consistent with the observations made by Saadati *et al.* (2022) who reported increases in yield from mutagen treated crop. These findings suggest that gamma radiation, particularly at doses of 20 Gy or 30 Gy, can mitigate the adverse effects of salinity stress on strawberry while increasing fruit weight. A similar overestimation was observed by Kepenek *et al.* (2016) on the Strawberry crop and Alhamza *et al.* (2016) Showed the superiority of doses 20 and 50 Gy in giving the highest strawberry fruit weight under salinity stress. These findings were confirmed by (Moemen, 2021) in *Ambrosia maritima* fruit and MR, (2024) in Acid Lime. Previous studies by Shaddad *et al.* (2023) and Azzam *et al.* (2022) have shown that low doses of gamma radiation stimulate the production of growth-promoting hormones, such as auxins and cytokinins, which play a crucial role in enhancing fruit development and weight under high salinity stress condition.

Yield per plant

The combined impact of gamma radiation and NaCl salinity stress on strawberry fruit yield was significant in both varieties studied. In the Winter Dawn variety, the highest fruit yield was observed in runners treated with a 30 Gy gamma radiation dose under 50 mM salinity conditions. However, in the Chandler variety, the pooled data revealed that the maximum fruit yield occurred in runners treated with a 20 Gy gamma radiation dose under the same salinity level. This is consistent with the findings from Saadati *et al.* (2022) who observed that gamma irradiation led to increased yield in pomegranate, indicating a broader applicability of these results across different fruit-bearing plants. In addition, studies on other crops, such as the work by Nandariyah *et al.* (2020) found that gamma-ray irradiation resulted in higher fruit weights in banana plants, suggesting a direct correlation between irradiation and improved fruit yield. Lower doses of gamma radiation are known to stimulate the production of secondary metabolites and antioxidants, which can enhance plant acclimatization to salinity stress Wi *et al.* (2007). Previous research has demonstrated that high salinity negatively affects plant physiological processes, reducing growth and yield in raspberries Shahid *et al.* (2020). However, our findings suggest that gamma radiation, as a physical mutagen, can mitigate the adverse effects of salinity stress. Gamma radiation appears to induce genetic or physiological

changes that enable plants to sustain higher yields, even under challenging conditions like salinity stress Wi *et al.* (2007).

Chlorophyll content index

Chlorophyll is a crucial indicator of plant health, especially under stress conditions such as salinity. In this study, the pooled data for both Winter Dawn and Chandler strawberry varieties revealed significant differences in chlorophyll levels when exposed to various gamma radiation and NaCl salinity treatments. In Winter Dawn, the highest chlorophyll index was observed in plants treated with a 30 Gy gamma radiation dose combined with 100 mM NaCl. A similar trend was noted in the Chandler variety.

In an earlier experiment, Shereen *et al.* (2009) reported that gamma-treated seedlings exhibited the highest chlorophyll content under salinity stress conditions. Similarly, in almond Motallebi *et al.* (2005) found that gamma-irradiated cuttings demonstrated superior chlorophyll content even under saline conditions. Additionally, a study by Manjusha *et al.* (2024) on *Citrus aurantifolia* recorded best chlorophyll content under salinity stress conditions following gamma irradiation treatment. This was also confirmed by Youssef *et al.* (2023) on strawberry crop.

Sakr *et al.* (2013) observed that exposing *Dracaena* plants to 10 Gy of gamma radiation resulted in a higher chlorophyll content. Similarly, Atak *et al.* (2007) reported an increase in chlorophyll levels in *Paulownia* plants exposed to 20 Gy gamma irradiation, correlating with an enhanced chlorophyll content. Consistently, our study also demonstrated a gradual increase in chlorophyll content in irradiated plantlets at lower doses of irradiation. Castro *et al.* (2007) explained that low levels of radiation activate the enzyme system, which in turn increases chlorophyll levels. This activation likely enhances the photosynthetic rate, ultimately contributing to higher yields.

Moreover, studies have indicated that specific doses of gamma irradiation can optimize chlorophyll content. For instance, Aly *et al.* (2019) found that a 200 Gy dose significantly increased chlorophyll a and b contents in wheat cultivars under salinity conditions. This is consistent with findings from Jan *et al.* (2023) who observed that gamma irradiation led to increased synthesis of chlorophyll and carotenoids in *cullen corylifolium* medicine plant. Furthermore, the research by Nikam *et al.* (2015) highlighted that irradiated sugarcane callus showed improved regeneration under NaCl stress, indicating that gamma irradiation can enhance salt tolerance while promoting chlorophyll synthesis. These findings align with

previous studies on strawberries, which show that gamma radiation can stimulate certain physiological processes, such as chlorophyll biosynthesis, helping plants better withstand salinity stress (Wi *et al.* 2007; Kim *et al.* 2004). Furthermore, salinity stress is known to negatively affect chlorophyll content due to oxidative damage caused by the accumulation of salt ions in plant tissues Jamil *et al.* (2011). This reduction in chlorophyll could be mitigated by the effectiveness of low doses of gamma radiation in scavenging reactive oxygen species (ROS), which otherwise damage chlorophyll pigments. The positive impact of irradiation on chlorophyll has been linked to the stabilization of the enzyme's active site and the enhancement of photosynthetic processes, as noted by Zabalza *et al.* (2006). Additionally, Matile *et al.* (1999) emphasized a critical period of interconversion between chlorophyll a and chlorophyll b, particularly under stress conditions. Variations in the timing of chlorophyll synthesis in palisade and spongy mesophyll cells, along with the biosynthetic pathways of chlorophyll pigments, contribute to the observed chlorophyll variegation in irradiated plants.

Number of leaves

The number of leaves on a plant is a key indicator of its overall health and ability to perform photosynthesis. This study analysed the effects of gamma radiation and salinity stress on leaf production in two strawberry varieties, Winter Dawn and Chandler. In the Winter Dawn variety, the pooled data revealed that the combination of 30 Gy gamma radiation and 50 mM NaCl salinity resulted in the highest number of leaves. This suggests that this treatment may have triggered stress tolerance mechanisms in the plants, enhancing their leaf development under salinity stress. This finding is consistent with previous research on banana crops, which showed that high salinity levels inhibit plant growth by disrupting water uptake and causing cellular damage (Munns and Tester, 2008).

Similarly, in the Chandler variety, experiments conducted over two years revealed a comparable trend. The maximum number of leaves was recorded in plants treated with 20 Gy gamma radiation and 50 mM NaCl salinity. Increased leaf production is crucial for maintaining the plant's photosynthetic area by producing more functional leaves. Leaf growth and expansion are essential developmental processes for establishing photosynthetic capacity and supporting overall plant growth Volkenburgh *et al.* (1999). Therefore, leaf development and physiological traits are valuable criteria for selecting cultivars with better tolerance to stress (Alamgir and Ali, 2006).

Reduced leaf growth, commonly associated with increased soil salinity, is primarily due to the osmotic effects of salt around the roots. This reduces water uptake, causing leaf cells to lose water. Although this reduction in cell volume and turgor is temporary, it inhibits cell elongation and division, resulting in smaller leaves (Bartels and Sunkar, 2005; Munns and Tester, 2008). Furthermore, salt accumulation in leaves has been linked to a decline in photosynthesis, metabolic damage, and eventual leaf death (Yeo and Flowers, 1986). Our results align with the findings of Jala *et al.* (2011) who reported that increased gamma radiation concentrations reduced leaf number in the Toremia fourmieri plant. The production and expansion of leaves are crucial for establishing photosynthetic capacity and promoting plant growth Volkenburgh *et al.* (1999). The reduction in leaf growth caused by increased soil salinity is mainly attributed to the osmotic effects of salt around the roots, which limits water uptake and leads to water loss in leaf cells. While this reduction in cell volume and turgor is temporary, it leads to slower leaf development, smaller leaf size, and decreased cell division over time.

Number of bud, flowers and fruits

This study analysed the effects of gamma irradiation on bud, flowers and fruits production in two strawberry varieties, Winter Dawn and Chandler, under salinity stress. In winter Dawn and chandler variety, the maximum number of buds, flowers and fruit was recorded in runners treated with a 30 Gy gamma irradiation dose exposed to 75 mM salinity condition. Gamma irradiation appears to mitigate the negative effects of salinity stress on number of buds, flowers and fruit production in both Winter Dawn and Chandler.

In a study by Zafar *et al.* (2022) it was reported that gamma rays induced genetic variability in tomato germplasm, leading to significant increases in the number of flower clusters, flowers, and fruits per plant compared to untreated controls. This aligns with findings from (Ulukapi, 2021) which demonstrated that low-dose gamma irradiation under salt stress conditions positively influenced the stability of protein synthesis and enhanced flowering and fruiting in Phaseolus vulgaris. El-Beltagi *et al.* (2013) demonstrated that gamma irradiation significantly improved growth parameters in cowpea plants subjected to salt stress, leading to an increase in flower production compared to controls plants. Similarly Ezzat *et al.* (2019) Indicated that gamma radiation could induce genetic variation in cowpea plants, leading to an increase in the number of flowers per plant. Similarly, Geng *et al.* (2019) reported that pretreatment with high doses of gamma irradiation on sweet osmanthus seeds enhanced the seedlings' tolerance to salinity stress. The findings indicated that seedlings from irradiated seeds not only survived

better under saline conditions but also exhibited improved growth characteristics, which included an increase in flowering.

Moreover, the study by Moghaddam *et al.* (2011) highlighted that low doses of gamma irradiation could stimulate growth by altering hormonal signaling pathways in plants, which may be particularly beneficial under stress conditions. This hormonal modulation could explain the increased number of reproductive structures, such as buds and flowers, observed in plants subjected to gamma irradiation. Similarly, research by Qi *et al.* (2014) indicated that low-dose gamma irradiation could activate RNA or protein synthesis, further supporting the notion that irradiation can enhance growth and development in plants.

Gamma irradiation acts as a physical mutagen that can stimulate plant growth and development, particularly at low doses. Research has shown that low doses of gamma rays can enhance enzymatic activities and promote cell division, particularly in meristematic tissues, which are crucial for root and shoot growth (Almukhtar and Bajlan, 2022). This stimulation can lead to increased biomass and improved reproductive traits, such as the number of flowers and fruits produced. For instance, studies have indicated that gamma irradiation can activate genes associated with growth and stress responses, which may contribute to enhanced flowering and fruiting under adverse conditions (Qi *et al.* 2014; Purwanto *et al.* 2004).

Days to first bud, flower and fruit initiations

In this study, we observed significant variation in bud, flower, and fruit initiation times across different treatments in both the Winter Dawn and Chandler strawberry varieties. The pooled analysis revealed that runners treated with a 30 Gy gamma radiation dose exhibited earlier bud initiation under 75 mM salinity condition. El-Beltagi *et al.* (2013) reported significant improvements in growth parameters of cowpea plants subjected to gamma irradiation under salt stress conditions. Their findings suggest that gamma irradiation can enhance growth traits, which may correlate with earlier reproductive phases such as bud and flower initiation.

These findings are consistent with previous studies, which have shown that gamma irradiation in mango can enhance growth parameters, such as early bud initiation, by inducing genetic changes that promote faster development (Ahloowalia and Maluszynski, 2001; Shu, 2009).

In contrast, salinity stress is known to hinder plant growth, leading to delayed flowering and fruiting, as reported in strawberry crops (Munns and Tester, 2008; Zorb *et al.* 2019). Gamma radiation can influence physiological processes related to bud development, potentially through

the enhancement of antioxidant activity or the modulation of stress-related gene expression Jan *et al.* (2012). Studies suggest that gamma radiation, when applied at optimal doses, can trigger adaptive responses in plants, potentially boosting growth rates and accelerating early developmental stages, which is crucial for plants in saline environments Ali *et al.* (2015).

Furthermore, gamma radiation can support physiological processes in plants, helping them better tolerate stress such as salinity, which typically delays fruit development (Li *et al.* 2021; Sharma *et al.* 2022). Additionally, it has been suggested that gamma irradiation can improve overall plant tolerance to stress by inducing mutations that promote growth and development under adverse conditions Khan *et al.* (2020).

Physiological parameter

Membrane stability index

The Membrane Stability Index (MSI) is an important parameter for evaluating the physiological response of plants to salinity stress, as it reflects the integrity and functionality of cell membranes under stress conditions. High salinity can cause ion imbalance and oxidative damage, disrupting membrane stability, leading to cell leakage and reduced cellular function Yadav *et al.* (2022). In this experiment, the highest MSI was observed in runners treated with 20 Gy gamma irradiation under a 50 mM salinity level. Similarly reported by Abdoun *et al.* (2022) Chickpea cultivar shows increasing membrane stability under salt stress in gamma irradiation treated seedlings. A similar overestimation was observed by Geng *et al.* (2019) sweet osmanthus increase membrane stability under saline condition. A similar study on salinity conditions showed that gamma-irradiated in salinity condition sweet osmanthus seedlings exhibited improved MSI, reduced oxidative damage, and increased antioxidant enzyme activities, including superoxide dismutase (SOD) and catalase (CAT) Geng *et al.* (2019). These findings align with previous research indicating the positive effects of gamma irradiation on enhancing plant stress tolerance by stabilizing cell membranes and reducing oxidative damage under various stress conditions (Abello *et al.* 2020; Muley *et al.* 2019). Sairam *et al.* (2005) suggested that salinity-induced cell membrane damage is associated with the presence of reactive oxygen species (ROS). Similarly, Upadhaya *et al.* (1990) identified increased ion leakage as an indicator of membrane damage caused by lipid peroxidation and ROS. Typically, the plasma membrane is protected from desiccation-induced damage by membrane-compatible solutes such as sugars and amino acids. Consistent with this, Surendar *et al.* (2013) observed minimal electrolyte leakage and the lowest reduction in MSI in salt-

resistant banana cultivars compared to the control. Salinity stress often leads to an increase in ion concentration, which can disturb cellular balance and damage membranes. A higher MSI indicates stronger membrane integrity, suggesting that the plant can effectively withstand salinity stress by preserving cellular stability and function. This resilience is vital for plant survival and productivity under saline conditions, making MSI a key measure for assessing salinity tolerance in plant species Ali *et al.* (2021).

Leaf Relative water content

In this study, the leaf relative water content (LRWC) of strawberry plants was evaluated under the combined influence of gamma radiation and salinity stress, using two varieties: Winter Dawn and Chandler. For the Winter Dawn variety, the highest LRWC was observed in runners treated with a 30 Gy gamma irradiation dose combined with 75 mM salinity. In contrast, the Chandler variety exhibited the maximum LRWC in runners treated with a 20 Gy gamma irradiation dose and 50 mM NaCl exposure.

Salinity stress induces cellular dehydration in plants by disrupting water balance within the cells. High salt concentrations in the soil lead to water loss and cell shrinkage, ultimately reducing leaf LRWC as cells lose their ability to retain water Karlidag *et al.* (2010). A reduction in LRWC in stressed plants often correlates with decreased plant vigor (Halder and Burrage, 2003). Variations in the ability of mutant lines to retain water or regulate water loss through stomata may be linked to their relative water content. This suggests differences in the ability of the studied varieties to accumulate water and adapt osmotically, helping them maintain tissue turgor and support physiological activities. Ahmed *et al.* (2020) noted that higher LRWC enables plants to sustain turgidity, though it may result in lower biomass and yield under stress. Suriya-arunroj *et al.* (2004) highlighted RWC as an effective measure of salt stress tolerance in rice. Unlike plant water potential, RWC estimation more accurately reflects the balance between water absorbed by the plant and water lost through transpiration. (Oszyszka, 2024) observed that even a 5% decline in RWC could cause a 40-50% reduction in photosynthesis. (David, 2002) suggested that decreases in LRWC can negatively impact stomatal conductance, photosynthesis, and transpiration.

Membrane integrity plays a crucial role in salinity tolerance Zhang *et al.* (2024) found that salt-tolerant varieties maintain better membrane integrity than susceptible ones. Stress-induced cellular membrane dysfunction is characterized by increased permeability and ion leakage, which can be easily quantified through electrolyte efflux Rachappanavar *et al.* (2024). Additionally, Sairam *et al.* (2005) suggested that reactive oxygen species (ROS) contribute to

membrane damage caused by salinity in plants. any factor leading to membrane damage increases permeability and ion leakage. concluded that membrane integrity is better preserved in salt-tolerant varieties compared to susceptible ones.

Electrolyte leakage

Electrolyte leakage estimation is commonly used as an indicator of membrane damage in plants, especially under stress conditions like salinity, as it reflects the integrity of the cell membrane and its ability to maintain homeostasis. Under salinity stress, excessive salt accumulation can disrupt cellular processes, leading to increased membrane permeability and higher electrolyte leakage Sharma *et al.* (2020).

For the Winter Dawn variety, the lowest electrolyte leakage was observed in runners treated with a 30 Gy gamma irradiation dose under 50 mM salinity conditions. In the Chandler variety, the lowest electrolyte leakage was recorded in runners treated with a 20 Gy gamma irradiation dose under 100 mM salinity conditions. Similarly, reduced electrolyte leakage under saline conditions has been reported in pomegranate by Saadati *et al.* (2022). A comparable observation was made by Nikam *et al.* (2014), where gamma irradiation as a mutagenic treatment effectively reduced electrolyte leakage in sugarcane under saline conditions.

These results suggest that moderate levels of gamma radiation help sustain the plant's ability to maintain cell membrane integrity under stress. This finding aligns with Munns and Tester (2008), who noted that high salinity disrupts cellular processes, leading to increased ion leakage, which is indicative of cell membrane damage. Salinity is known to impose osmotic stress, triggering physiological responses that can compromise plant health Zhang *et al.* (2015). Studies on strawberries have shown that salt stress increases membrane permeability and subsequent leakage, signalling membrane damage and cell death. This can lead to reduced fruit quality and shelf life. The leakage of electrolytes, such as potassium (K^+) and calcium (Ca^{2+}), disrupts normal cellular processes and nutrient balance within the fruit. Consequently, it can result in physiological disorders, such as reduced sugar accumulation, impaired flavour development, and changes in fruit texture Karlidag *et al.* (2009). Electrolyte leakage is generally considered an indirect measure of plant cell membrane damage, with increased electrical conductivity highlighting greater membrane permeability and electrolyte leakage Chauhan *et al.* (2006).

Proline content

Proline estimation under salinity stress is widely recognized as an indicator of plant response to osmotic challenges. Under salinity stress, plants tend to accumulate proline as a defence mechanism to stabilize cellular structures, scavenge reactive oxygen species, and maintain osmotic balance. As an osmoprotectant, proline helps buffer the cellular environment, preventing damage to essential cellular components such as proteins and membranes. Its accumulation is often associated with improved stress tolerance, as it aids in maintaining cell turgor and preventing dehydration (Ashraf, 2004; Yoshida *et al.* 1997).

In the present study, the highest proline content was observed in the Winter Dawn and Chandler strawberry runners treated with a 30 Gy dose of gamma irradiation under 50 mM NaCl salinity conditions. The ability of plants to withstand salinity stress largely depends on their capacity to sustain osmotic balance, often achieved through the accumulation of organic or inorganic compounds. Research by (Bybordi, 2012; Krupa-Mańkiewicz *et al.* (2015) has shown that elevated proline levels in plant tissues are a robust indicator of exposure to stress factors. The accumulation of osmoprotectants like proline is well-documented as a primary defence mechanism, helping to regulate osmotic pressure within cells (Desingh and Kanagaraj, 2007; Koca *et al.* 2007; Veeranagamallaiah *et al.* 2007). In various plant species, the ability to accumulate proline in response to environmental stress has been closely linked to increased stress tolerance (Maggio *et al.* 2002; Claussen, 2005).

In this study, proline levels significantly increased across all mutant lines subjected to varying levels of salt stress. For mutant lines treated with 0 mM NaCl, proline remained relatively low at different days after treatment (DAT). The observed variations in proline content across mutant lines, along with its increase as salinity levels rose, are consistent with findings in citrus Amal *et al.* (2008), pistachio Karimi *et al.* (2009), and mango Srivastav *et al.* (2010). Excessive proline accumulation can occur under intense environmental stress, resulting from increased biosynthesis, reduced oxidation, limited protein incorporation, or release due to proteolysis. Studies indicate that salinity-tolerant plants tend to accumulate more proline than susceptible varieties. Additionally, proline helps mitigate the toxic effects of NaCl, particularly by stabilizing DNA helical structure during replication. Increased proline levels with higher gamma irradiation concentrations have also been noted in studies on Iraqi dates (Auda and Al-WanDawni, 1980), Akshatha *et al.* (2013), Pigeon Pea Chandra *et al.* (2014) Desairao *et al.* (2014) Sugarcane Nikam *et al.* (2015).

Leaf scorching

In the Winter Dawn variety of strawberry, the use of physical mutagens such as gamma radiation has been shown to significantly reduce the damaging effects of salinity, particularly leaf scorching. In the present study, the application of 30 Gy of gamma radiation combined with a 75 mM salinity condition resulted in the lowest leaf scorching. Similarly, in the Chandler variety of strawberry, physical mutagenesis using gamma radiation also demonstrated a significant reduction in leaf scorching under salinity stress.

In the pooled analysis, plants treated with 20 Gy of gamma radiation and exposed to 75 mM NaCl showed minimal leaf scorching. Salinity stress in fruit crops, including strawberries, often leads to symptoms like leaf scorching, where the leaf edges and tips develop brown or necrotic spots. This response is primarily due to the accumulation of high levels of sodium and chloride in the leaves, which disrupts water uptake, impairs photosynthesis, and damages cellular structures through osmotic and oxidative stress Singh *et al.* (2018).

Leaf scorching in fruit crops under salinity stress is caused by the accumulation of salt ions, particularly sodium (Na^+) and chloride (Cl^-), in the leaf tissues. When these ions build up, they disrupt cellular function, leading to dehydration and cellular damage Yadav *et al.* (2011). Under saline conditions, salt injury often manifests as leaf burn and defoliation, which are linked to the buildup of harmful salt concentrations in leaf cells. The reaction to these toxic effects varies depending on the rootstock used Yesiloglu *et al.* (2015).

Leaf area

The application of gamma irradiation at a 30 Gy dose on strawberry runners significantly helps sustain leaf area, particularly under high salinity 100 mM NaCl conditions, as observed in both the Winter Dawn and Chandler varieties of *Fragaria ananassa*. Gamma irradiation is known to cause mutagenic changes that alter cellular functions, leading to sustained osmotic adjustment, proline accumulation, and antioxidative responses in plants under stress (Ahloowalia *et al.* 2004; Siddiqui *et al.* 2016). These changes may contribute to a larger leaf area, as irradiated plants are better able to manage oxidative stress induced by salinity, thereby supporting leaf expansion Asadi *et al.* (2019).

In strawberries, where leaf area is directly correlated with photosynthetic capacity and overall yield potential, an increase in leaf area under stress conditions due to gamma irradiation suggests improved resilience and productivity Kumar *et al.* (2022). The reduction in leaf area observed under higher salt levels is a general response in many plants, including bananas, and has been described in several genotypes evaluated by different researchers (Gomes *et al.* 2004;

Silva *et al.* 2009; Ferraz *et al.* 2008; Willadino *et al.* 2011). This decrease in leaf area may also be due to restricted CO₂ fixation, which lowers the net assimilation rate and increases reactive oxygen species (ROS) production Miller *et al.* (2009).

The decrease in leaf area is also linked to the energy diversion required to maintain and activate metabolic activities related to salinity tolerance. These include the synthesis of organic solutes, maintenance of membrane integrity, ion extrusion, and the regulation of ion transport and distribution within various organs and cell (Munns and Tester, 2010).

Mineral analysis potassium K⁺

The results indicating that runners treated with a 30 Gy gamma irradiation dose and exposed to 50 mM NaCl exhibited the highest potassium (K⁺) levels, while the lowest potassium levels were recorded in runners under 100 mM NaCl, highlight the complex interaction between ionizing radiation and salinity stress in plant physiology. This phenomenon can be explained by the physiological mechanisms that enhance potassium uptake and its role in mitigating salt stress.

Gamma irradiation has been shown to improve the growth and nutrient uptake of plants, particularly under stress conditions. Also (Guseva, 2022) reported that gamma radiation can enhance the biological potential of seeds and plants, leading to increased potassium ion and sustained growth. This finding aligns with (Kapoor and Yadav, 2022) who noted that exogenous potassium significantly reduced malondialdehyde content, suggesting a protective role against oxidative stress induced by salinity. Therefore, the improved potassium levels in the irradiated runners may be attributed to enhanced root architecture and ion transport mechanisms that facilitate K⁺ absorption, particularly under saline conditions.

Additionally, the interaction between potassium and sodium ions is crucial for understanding plant responses to salinity. Chen *et al.* (2022) emphasized that salt-tolerant plants preferentially absorb K⁺ over Na⁺, thus maintaining a higher K⁺/Na⁺ ratio, which is essential for cellular homeostasis and stress tolerance. This is relevant in the context of the observed results, as the irradiated runners likely exhibited improved K⁺ absorption capabilities, which would help counteract the harmful effects of high Na⁺ concentrations in the soil. The physiological basis for these observations is also linked to potassium's role in regulating osmotic balance and enzyme activities that mitigate oxidative stress. For instance, Chen *et al.* (2022) noted that potassium supports the activity of antioxidant enzymes, which are crucial for scavenging reactive oxygen species (ROS) generated under salt stress. The increase in K⁺ levels in the

irradiated runners may have contributed to a stronger antioxidant response, reducing oxidative damage and promoting overall plant health and stress tolerance.

Mineral analysis sodium Na⁺

The results of the mineral analysis indicate that sodium (Na⁺) content in plants is significantly influenced by salinity treatments. Specifically, the highest sodium content was observed in plants treated with 100 mM salinity, while the lowest was recorded in runners treated with a 30 Gy gamma irradiation dose under 50 mM salinity conditions. This finding supports the physiological understanding that salinity stress typically leads to increased sodium accumulation in plant tissues, which can adversely affect plant growth and development by disrupting ion homeostasis and metabolic processes (Aly *et al.* 2019; El-Beltagi *et al.* 2013; Kumar *et al.* 2017).

Salinity stress induces both osmotic and ionic stress in plants, triggering physiological responses such as the accumulation of compatible solutes like proline and the activation of stress-responsive pathways (Aly *et al.* 2019; Kumar *et al.* 2017). The increase in sodium content under high salinity conditions is a well-documented phenomenon, as sodium can interfere with potassium uptake, which is essential for various cellular functions, including enzyme activation and maintaining osmotic balance Frary *et al.* (2011). Conversely, the reduction in sodium content observed in gamma-irradiated runners under moderate salinity suggests a potential adaptive mechanism. This suggests that gamma irradiation may help the plant manage salt stress more effectively. Studies have shown that low doses of gamma irradiation can stimulate growth and enhance stress tolerance by modulating physiological and biochemical pathways (Qi *et al.* 2014; Geng *et al.* 2019).

Furthermore, gamma irradiation has been shown to induce changes in gene expression that improve the plant's stress tolerance mechanisms. For instance, it can upregulate genes involved in antioxidant defense and osmotic adjustment, helping to mitigate the adverse effects of salinity (El-Fiki *et al.* 2016; Abd-Elhameed *et al.* 2022). The interaction between gamma irradiation and salinity stress highlights a complex physiological response, where irradiation may enhance the plant's resilience to ionic stress by promoting the synthesis of protective compounds and improving ion homeostasis (El-Beltagi *et al.* 2013; Kumar *et al.* 2017).

5.2 Chemical mutagen EMS against salinity stress

Plant height

The pooled data in this study highlights the significant impact of EMS treatment combined with salinity stress on plant height in two strawberry varieties, Winter Dawn and Chandler. In the Winter Dawn variety, the maximum plant height was observed when plants were treated with a combination of 2% EMS dose against 50 mM NaCl as well as in the Chandler variety, the highest plant height was recorded in runners treated with 1% EMS dose against 75 mM NaCl, indicating that this EMS doses promote growth even under saline conditions. Similar findings were reported by Abbas *et al.* (2018) the EMS dose of 2% shows maximum plant height under salinity NaCl concentration in strawberry crop. This is also confirmed in the literature of several workers Karimi *et al.* (2009) in pistachio, Refai *et al.* (2016), Kishk *et al.* (2016), Harty *et al.* (2018) and (Palaniappam and Yerriswamy, 1997) in banana. This might be reflected with the views of several author who stated reduction induced by salinity in growth of plants have often been related to measured decreases in the plastic extensibility of cell walls in root and leaf expansion zones (Neumann, 1993; 1995). EMS induces point mutations in the DNA by alkylating the guanine base, leading to mispairing during DNA replication. This process creates genetic variability that can lead to phenotypic changes, including height variations in plants Nair *et al.* (2019). Mutations induced by EMS can alter the levels of plant hormones like auxins, gibberellins, and cytokinins, which play crucial roles in cell elongation and growth. For instance, changes in gibberellin biosynthesis can lead to increased plant height under saline conditions by promoting cell elongation and division or by activating specific genes related to stress tolerance, such as those involved in ionic balance, osmoprotection, and cell wall stability. (Zhou *et al.* 2022; Luan *et al.* 2007; Sikder *et al.* 2013).

Chlorophyll

In the current study on the Winter Dawn and Chandler varieties of strawberries, we analysed the effects of different doses of the chemical mutagen EMS and varying salinity stress levels on plant chlorophyll content. Chlorophyll content is an essential parameter as it is closely related to the plant photosynthetic efficiency and overall growth. The chlorophyll content was recorded as the maximum in winter Dawn and Chandler variety runners treated with 2% EMS dose against the 75 mM and 100 mM salinity conditions. In strawberry plants, induced salinity stress from NaCl can cause changes in chlorophyll content due to osmotic and ionic stress. High salinity disrupts cellular water potential, leading to dehydration and oxidative stress,

which in turn impairs chlorophyll synthesis and accelerates its degradation. NaCl exposure causes an accumulation of sodium (Na^+) and chloride (Cl^-) in plant tissues, disrupting ion homeostasis and affecting enzyme activities that are essential for chlorophyll biosynthesis. Moreover, salinity stress triggers the production of reactive oxygen species (ROS), which can damage chlorophyll molecules and the photosynthetic apparatus, reducing chlorophyll content and, subsequently, photosynthetic efficiency (Parida and Das, 2005; Ashraf and Harris, 2013).

One study on EMS treated tomatoes found that mutant lines maintained significantly higher chlorophyll levels under salt stress than wild type lines, with enhanced activities of antioxidant enzymes that help reduce oxidative damage to chlorophyll molecules Ahmad *et al.* (2020). Similar effects have been observed in strawberries, where EMS-mutated lines showed not only improved chlorophyll retention but also overall increased biomass in saline conditions Jbir *et al.* (2019). A similar overestimation was observed by Al-Salihy *et al.* (2018) and Abd EL-Moneim *et al.* (2021) in Strawberry.

These findings align with other studies that suggest EMS can help improve plant stress tolerance by inducing beneficial mutations. For instance, research by Ghafoor *et al.* (2022) found that EMS treatment in papaya crop resulted in increased chlorophyll content and better stress adaptation, similar to our observations. Additionally, Ahmad *et al.* (2021) reported that combining EMS mutagen treatments against salinity stress could help plants maintain higher chlorophyll levels, supporting their growth under adverse conditions.

Number of bud, flower and fruit

The application of the chemical mutagen ethyl methane sulfonate (EMS) has demonstrated a notable impact on the reproductive traits of strawberry plants, particularly influencing the number of buds, flowers, and fruits. Studies on both the Winter Dawn and Chandler varieties indicate that EMS, especially at a 2 % concentration combined with 50 mM NaCl salinity stress, can significantly increase these reproductive outputs. The mechanism behind this enhancement appears linked to EMS potential to disrupt genes essential for flowering, fruit setting, and fruit development (Yogendra and Ramanjini, 2013). This disruption likely induces genetic changes that promote a greater resilience to salinity stress, allowing for improved reproductive success. EMS is known to cause point mutations, which can lead to the expression of favourable traits, such as an increased number of buds and flowers and fruits (Roychowdhury and Tah, 2011). Salinity stress typically impairs plant growth and reproductive development by disturbing ion balance and water uptake, which adversely affects the plant's

physiological health. However, EMS-treated plants seem to tolerate better under such conditions, likely due to genetic modifications that enhance their physiological responses and stress management Yatta El Djouzi *et al.* (2023). These findings align with previous research that has reported similar improvements in plant growth and yield under salinity stress following EMS treatment. For instance, studies on other crops like tomatoes and wheat have demonstrated that EMS can boost flowering under salinity tolerance and improve reproductive development despite stress (Mustafa *et al.*, 2021; Serra, 2009). These studies highlight the potential of EMS as a tool for enhancing crop performance under challenging environmental conditions.

Fruit weight

Although, due to alterations in processes such as cell division, cell expansion or other growth related mechanisms, certain mutants could be noticed with decreased fruit weight or increased fruit weight in segregating progeny (Jankowicz-Cieslak and Till, 2015; Krasileva *et al.* 2017). In the present investigation shows the two year results of Winter Dawn and Chandler variety shows that the runners treated with 1% EMS dose under 50 mM NaCl stress shows maximum fruit weight. According to research by Arora *et al.* (2019), the application of EMS in the strawberry cultivar Albion significantly influenced fruit characteristics under saline stress. They reported that a 2% EMS dose notably increased fruit weight and quality under induced salinity stress conditions, which aligns with the findings of our pooled analysis. Similar observations were reported by Abbas *et al.* (2018) in the strawberry cultivar *Festival*, Urias-Salazar *et al.* (2023) in *Capsicum*, and Chen *et al.* (2018) in *Cucurbita moschata*.

Moreover, the physiological response to EMS can enhance the plant metabolic activities, including photosynthesis and nutrient assimilation, which are crucial for fruit development (Al-Babili and Bouwmeester, 2015). The increase in fruit weight in response to EMS treatment can be attributed to the enhanced physiological processes stimulated by the mutagen. EMS is known to induce genetic variability, which can lead to improved growth and fruit development characteristics in plants Hussain *et al.* (2019).

Fruit yield per plant

The investigation into the fruit yield per plant of the Winter Dawn and Chandler varieties, particularly in relation to the application of a 2% Ethyl Methane Sulfonate (EMS) dose induced with 100 mM NaCl, reveals significant findings consistent with previous studies. The

application of EMS as a mutagen has been shown to enhance various yield attributes across different crops. For instance, (Singh, 2021) reported that lower concentrations of mutagens, including EMS, positively influenced yield attributes such as overall fruit yield per plant in brinjal genotypes under salinity stress. This suggests that similar methodologies could be beneficial in optimizing fruit yield in the Winter Dawn and Chandler varieties.

Moreover, the relationship between mutagen application and plant yield is further supported by the findings of Gandhi *et al.* (2014), who noted that EMS treatment resulted in a spectrum of mutations that positively affected quantitative traits, including the total number of fruits per plant. This aligns with the observed maximum fruit yield in the treated runners of the varieties, indicating that the application of EMS can be a viable strategy for enhancing fruit production. In addition to mutagenesis, the influence of salinity on plant yield has been documented extensively. Bezerra *et al.* (2020) highlighted that salinity levels significantly affect the number of productive branches and the overall yield of passion fruit, suggesting that the interaction between EMS and NaCl could similarly impact fruit yield in the studied varieties. Furthermore, the positive correlation between the number of fruits per plant and yield, as noted in various studies, reinforces the potential for increased fruit yield through targeted mutagenesis and salinity management (Kumar and Patil, 2020; Gillani *et al.* 2019; Yatung *et al.* 2015). The findings from these studies collectively support the assertion that the combination of a 2% EMS dose expose with 100 mM NaCl treatment can lead to enhanced fruit yield per plant in the Winter Dawn and Chandler varieties.

Days to bud, flower and fruit

The data shows that the application of the chemical mutagen EMS, with salinity stress, significantly impacts on the days of bud, flower and fruit initiation in both the Winter Dawn and Chandler strawberry variety. In both the variety early bud, flower and initiation was recorded in runners treated with 2% EMS induced with 50 mM NaCl. The interaction between chemical mutagens, such as ethyl methanesulfonate (EMS), and stress as salinity has been shown to significantly influence the physiological processes in various plant species, including strawberries. Specifically, the combination of 2 % EMS induced with 50 mM salinity conditions resulted in earlier initiation of these reproductive phases compared to control runners. Salinity stress, on the other hand, poses a significant challenge to plant growth and development, particularly in sensitive crops like strawberries. Excessive NaCl can disrupt physiological processes, leading to reduced chlorophyll content and impaired photosynthesis,

which ultimately affects growth and yield (Aras & Esitken, 2019). The physiological response to salinity includes alterations in cell membrane integrity and nutrient uptake, which can be exacerbated by the presence of mutagens (Koc, 2015). However, the application of EMS appears to mitigate some of these adverse effects by promoting earlier reproductive development, potentially allowing plants to complete their life cycle before the full impact of salinity stress is realized. Chemical mutagens like EMS are known for their ability to induce mutations through alkylation of DNA, primarily affecting guanine bases, which can lead to base substitutions and transitions in the genetic material (Jency and Ravikesavan, 2022; Hwang *et al.* 2014). This mutagenic action can enhance genetic variability, which is crucial for plant breeding and adaptation to stress conditions. Studies have demonstrated that EMS induces higher mutation rates than physical mutagens, providing a more efficient and specific means of generating genetic diversity (Mohammed and Ibrahim, 2017; Tharek *et al.* 2021). The mutagenic effects of EMS are particularly beneficial in breeding programs aimed at developing salt-tolerant cultivars, as they can lead to desirable traits such as improved growth and reproductive success under saline conditions Turkoglu *et al.* (2023).

Fruit length and diameter

The findings regarding the effects of ethyl methanesulfonate (EMS) and salinity stress on the fruit characteristics of the Winter Dawn and Chandler strawberry varieties reveal significant insights into plant physiology and stress tolerance mechanisms. In the Winter Dawn variety, the application of a 2% EMS dose in conjunction with 75 mM salinity condition resulted in enhanced fruit length and diameter, while the Chandler variety exhibited maximum fruit dimensions with a 1% EMS treatment under 100 mM NaCl. In the Chandler variety, the optimal fruit dimensions observed with a lower EMS concentration 1% at a higher salinity level 100 Mm salinity level suggest a threshold effect where lower mutagen doses may be more effective in certain genotypes. This aligns with findings in other crops, where moderate salinity stress can enhance fruit quality traits by promoting the accumulation of beneficial metabolites while minimizing the detrimental effects of excessive salt Oliveira *et al.* (2022). The physiological responses to salinity, such as osmotic adjustment and ion homeostasis, are critical for maintaining cellular functions and promoting fruit growth under stress conditions Wang, (2023).

The physiological basis for the observed improvements in fruit characteristics can be attributed to the role of EMS in inducing genetic variability, which may enhance the plant ability to adapt

to saline conditions. EMS is known to induce point mutations that can lead to beneficial traits, such as improved stress tolerance (Kumar & Kumari, 2021). The enhanced fruit size in response to EMS treatment under salinity stress may be linked to improved metabolic processes, including increased photosynthetic efficiency and enhanced antioxidant activity Kalaji *et al.* (2016).

Moreover, the interaction between salinity and EMS treatment likely influences hormonal regulation within the plants. Auxins, particularly indole-3-acetic acid (IAA), play a crucial role in regulating growth and development under stress conditions. Increased auxin levels have been associated with enhanced tolerance to salinity, as they can modulate root architecture and promote nutrient uptake, which is vital for fruit development Jia *et al.* (2019). The differential response of the two strawberry varieties to EMS and salinity may reflect their unique genetic backgrounds and inherent physiological mechanisms that govern stress responses.

Shoot fresh weight and dry weight

In this observation, pooled data for the Winter Dawn variety indicated that treating runners with a 2% EMS dose induced with 50 mM salinity condition resulted in the maximum shoot fresh weight. Additionally, the shoot dry weight in the Winter Dawn variety significantly improved in runners treated with 2% EMS dose-treated runners induced against 75 mM salinity level. For the Chandler variety, pooled data also revealed a significant improvement in shoot fresh weight with EMS treatment. The maximum shoot fresh weight was recorded in plants treated with a 2% EMS dose against 50 mM NaCl. Shoot dry weight followed a similar trend in the Chandler variety, with the highest was observed in the 2% EMS dose against the 50 mM NaCl treatment. The physiological mechanisms underlying these observations can be attributed to the interplay between salt stress and the plant's antioxidant defence systems. Nahar *et al.* (2016) elucidated that salt stress leads to ionic toxicity, which disrupts critical physiological processes and enhances the production of reactive oxygen species (ROS). This oxidative stress can be countered by the activation of antioxidant enzymes, which are essential for maintaining cellular homeostasis under saline conditions. The increase in shoot fresh weight observed in the current study may be a result of improved nutrient uptake and enhanced photosynthetic efficiency Feng *et al.* (2017) who found that salt stress induces ion imbalances and oxidative stress but can be mitigated through appropriate treatments. Furthermore, the findings regarding the Chandler variety's response to a 2% EMS dose against 50 mM NaCl corroborate earlier research that indicates specific salinity thresholds can optimize growth responses in various

plant species. For example, (Xia and Mattson, 2022) reported that certain concentrations of NaCl can favourably influence the shoot/root ratio, thereby enhancing overall biomass accumulation. This suggests that moderate salinity levels may stimulate growth processes rather than inhibit them, particularly when combined with growth-promoting treatments.

Root fresh weight and dry weight

The use of the chemical mutagen EMS (ethyl methane sulfonate) has shown significant promise in enhancing the tolerance of strawberry plants, specifically the Winter Dawn and Chandler varieties, to salinity stress induced by NaCl. The physiological effects of ethyl methanesulfonate (EMS) as a chemical mutagen in conjunction with salinity stress, particularly sodium chloride (NaCl), have been investigated in various plant species, revealing significant impacts on root fresh weight and dry weight. In the winter Dawn and Chandler varieties, it was observed that treatment with 2% EMS under 75 mM NaCl conditions resulted in maximum root fresh weight and dry weight. This phenomenon can be attributed to several physiological mechanisms that enhance plant resilience to salinity stress. Salinity stress is known to adversely affect plant growth by disrupting water uptake and ion balance, leading to reduced fresh and dry weights of roots and shoots. For instance, studies have shown that NaCl stress can significantly decrease root and shoot lengths, as well as overall biomass production in various species, including *Glycine max* and *Tagetes spp* Koksai *et al.* (2016). The physiological response to salinity involves alterations in metabolic pathways, including the generation of reactive oxygen species (ROS), which can lead to oxidative stress Dhillon *et al.* (2023). However, the application of EMS appears to mitigate some of these adverse effects by enhancing the plant adaptive responses.

The role of auxins, particularly indole-3-acetic acid (IAA), is crucial in regulating plant growth and stress responses. Auxins have been implicated in the modulation of root architecture and the enhancement of salinity tolerance (Jia *et al.* 2019; Korver *et al.* 2019). The EMS treatment may induce mutations that enhance the expression of auxin-related genes, thereby improving root development and function under saline conditions. This is supported by findings that demonstrate improved root architecture and physiological processes in plants subjected to salinity stress when treated with growth regulators Ma *et al.* (2017). Moreover, the interaction between EMS treatment and salinity stress may also involve the modulation of polyamine metabolism. Research indicates that effective microorganisms (EMS) can reduce the activities of diamine oxidase and polyamine oxidase in salt-stressed plants, suggesting that EMS may

similarly enhance polyamine levels, which are known to play a role in stress tolerance Kalaji *et al.* (2016). This modulation can lead to improved detoxification of ROS and enhanced growth metrics, including root fresh and dry weights.

Number of leaves

The interaction between ethyl methanesulfonate (EMS) treatment and salinity stress, particularly regarding the number of leaves produced by different plant varieties, has been a subject of interest in plant physiology. In the context of the Winter Dawn and Chandler varieties, it has been observed that EMS treatment enhances the number of leaves under NaCl stress conditions. Specifically, the Winter Dawn variety exhibited the highest leaf count when treated with 2% EMS in conjunction with 50 mM NaCl stress, while the Chandler variety showed optimal results with 1% EMS under similar salinity conditions. The physiological basis for the increased leaf production in EMS-treated plants under salinity stress can be attributed to several biochemical pathways. For instance, the application of EMS has been linked to enhanced nitrogen metabolism, which is crucial for leaf development. Studies have shown that salinity stress often leads to oxidative damage in plants, and the modulation of antioxidant systems is vital for mitigating such effects (Song *et al.* 2014; ElSayed *et al.* 2022). The presence of EMS appears to bolster these antioxidant defenses, thereby promoting healthier leaf growth even in the face of salinity stress. Moreover, the role of melatonin in enhancing plant tolerance to salinity stress has been documented, indicating that it can improve physiological parameters such as photosynthesis and antioxidant activity (Varghese *et al.* 2019; Ke *et al.* 2018). This suggests that EMS may similarly influence the synthesis of protective compounds that facilitate growth under adverse conditions. The induction of key enzymes involved in nitrogen metabolism, such as nitrate reductase, has been shown to be beneficial in maintaining leaf growth under salinity stress (Idrees *et al.*, 2010). This aligns with findings that indicate a positive correlation between enzyme activity and leaf number in stressed plants.

Additionally, the modulation of osmotic balance through the accumulation of compatible solutes like proline and glycine betaine has been reported to play a significant role in plant adaptation to salinity (Ghorbani *et al.* 2023; Ahmad *et al.* 2014). These compounds help in maintaining cell turgor and mitigating the detrimental effects of high salinity, thereby supporting leaf development. The enhancement of such osmoprotectants in EMS-treated plants could explain the observed increase in leaf number under NaCl stress.

Leaf relative content

The application of ethyl methanesulfonate (EMS) as a chemical mutagen has been shown to enhance the leaf relative water content (LRWC) of strawberry plants under salinity stress, particularly in varieties such as Winter Dawn and Chandler. In the Winter Dawn variety, an increase in LRWC was observed in plants treated with 2% EMS when subjected 50 mM salinity conditions, while the Chandler variety also exhibited improved LRWC with 1% EMS treatment induced with 50 mM salinity level. This suggests that EMS treatment may aid in water retention and enhance the physiological resilience of strawberry plants against salinity-induced stress Moneim *et al.* (2021).

The mechanism behind this improvement can be attributed to the mutagenic effects of EMS, which induce genetic variations that may enhance stress tolerance traits in plants. EMS is recognized for its ability to induce a high frequency of mutations, particularly GC to AT transitions, which can lead to beneficial phenotypic changes (Turkoglu *et al.* 2023; Mohd-Yusoff *et al.* 2015). Such mutations can potentially enhance the physiological mechanisms that regulate water retention and osmotic balance in plants, thereby improving their ability to cope with salinity stress Dalin *et al.* (2022). Moreover, studies have indicated that EMS treatment can lead to morphological variations that are advantageous under stress conditions. For instance, EMS-treated strawberry plants have shown improvements in leaf thickness and overall plant vigor, which are critical factors in maintaining higher LRWC during periods of salinity stress Dalin *et al.* (2022). The enhanced LRWC observed in EMS-treated plants can be linked to improved stomatal regulation and increased root biomass, which are essential for water uptake and retention under saline conditions Moneim *et al.* (2021).

Electrolyte leakage

In this study, the effect of EMS (ethyl methanesulfonate) treatments on electrolyte leakage in strawberry plants under salinity stress was evaluated in two different strawberry varieties, Winter Dawn and Chandler. Electrolyte leakage is a common indicator of membrane damage in plants, often used to assess their response to environmental stresses such as salinity Hasanuzzaman *et al.* (2020). When plants are exposed to high levels of salinity, cellular membranes can be damaged, leading to the leakage of ions, which in turn affects overall plant health. The pooled analysis across both years revealed that the 2% EMS exposed with 100 mM NaCl resulted in the lowest electrolyte leakage in winter Dawn variety and chandler variety. These results were further supported by Khayyat *et al.* (2009) in Strawberry and Arefian *et al.* (2014) in chickpea. The application of EMS has been associated with enhanced stability of cell

membranes in some studies, but the specific mechanisms and effects can vary widely among different plant species and conditions Talaat *et al.* (2014).

This suggests that the mutagen treatment helped the plants better tolerate salinity, potentially by stabilizing their cellular membranes. This finding further reinforces the idea that EMS treatment can mitigate the harmful effects of salinity on strawberry plants by protecting their cellular structures (Narimani and Seyed Sharifi, 2023). For instance, studies on strawberry have shown that under salt stress, genotypes with stable electrolyte leakage levels were better at tolerating salinity. Specifically, low electrolyte leakage in treated plants has been correlated with improved membrane integrity, which supports enhanced ion homeostasis and osmotic balance under saline conditions Khayyat *et al.* (2009). These findings on reduced electrolyte leakage in strawberry treated with EMS under salt stress align with research that suggests EMS treatments enhance salinity tolerance by improving cellular membrane stability. In a study involving EMS-treated ‘Festival’ strawberry, plants showed increased resilience to NaCl stress, maintaining better growth and physiological function than controls plants. This stability, in turn, is associated with reduced membrane damage and electrolyte leakage under high salinity conditions Abbas *et al.* (2018).

Proline content

In this study, the impact of chemical mutagen EMS on the proline content of strawberry varieties Winter Dawn and Chandler was evaluated under induced salinity stress caused by NaCl. Proline accumulation is a well-known physiological response of plants to abiotic stresses, such as salinity, as it plays a crucial role in osmotic adjustment and protecting plant cells from oxidative damage (Ashraf and Foolad, 2007).

For the Winter Dawn and Chandler variety the proline content was estimated maximum in runners treated with 1% EMS induced against 50 mM salinity condition. The application of ethyl methanesulfonate (EMS) as a mutagen has been shown to enhance proline accumulation in Rice, Wheat and Tomato under salinity stress, which is crucial for plant adaptation to osmotic stress. Proline serves as an osmoprotectant, stabilizing proteins and cellular structures, and mitigating the detrimental effects of salinity on plant physiology (Abdelaziz *et al.* 2018; Ebeid *et al.* 2022). Additionally, the research by Thiam *et al.* (2013) indicated that EMS-induced mutations in cowpea cultivars resulted in varying levels of proline accumulation under salt stress, suggesting that EMS can be effectively utilized to enhance proline levels and improve salinity tolerance in plants. This is corroborated by findings from Wei *et al.* (2019)

which showed that EMS treatment in *Arabidopsis* resulted in increased proline levels, highlighting the potential of EMS in breeding programs aimed at enhancing abiotic stress tolerance. Conducted a study on the impact of EMS-induced mutagenesis on strawberries, reporting that EMS treatments significantly improved proline content under salt stress conditions. This accumulation was associated with improved water retention and reduced oxidative stress, highlighting the role of EMS in enhancing stress resilience in strawberries (Raza and Alam, 2022).

Membrane stability index

The Membrane Stability Index (MSI) is a crucial indicator of plant tolerance under stress conditions, particularly salinity stress. In this study, we estimate the MSI in two strawberry varieties Winter Dawn and Chandler both treated with different doses of the chemical mutagen EMS (ethyl methanesulfonate) and exposed to varying levels of NaCl induced salinity stress.

For the Winter Dawn variety, the results showed a significant improvement in the Membrane Stability Index (MSI) in plants treated with a 2% EMS dose under 50 mM induced salinity stress. A similar trend was observed in the Chandler variety. This finding is consistent with previous studies by Abd El-Moneim *et al.* (2021) in strawberries, Abbas *et al.* (2018) in the Strawberry Festival cultivar, Arefian *et al.* (2014) in chickpea, and Meena *et al.* (2023) in sugarcane, all of which reported improvements in membrane stability index in plants subjected to EMS treatment under salinity stress conditions. Salinity stress is known to compromise membrane integrity, leading to increased permeability and subsequent cellular damage. For instance, Janmohammadi *et al.* (2012) reported that elevated NaCl concentrations result in decreased MSI which are indicative of compromised membrane stability and increased reactive oxygen species (ROS) accumulation in sensitive plant species Janmohammadi *et al.* (2012). This aligns with findings by Ali *et al.* (2022) who noted that salt stress significantly reduces overall growth and leaf gas exchange, which can be linked to membrane stability issues. This indicates that the mutagen-treated plants had a stronger membrane stability, reflecting their enhanced tolerance to salinity stress. These findings align with previous research that shows EMS-induced mutations can enhance plant resilience to abiotic stresses by stabilizing cell membranes, thus reducing damage caused by stressors such as salinity Hossain *et al.* (2006). EMS-treated plants likely have an enhanced ability to maintain ion homeostasis and membrane integrity, key factors in coping with salinity stress Ks *et al.* (2018).

Leaf scorching

The interaction between chemical mutagens, specifically ethyl methanesulfonate (EMS), and salinity stress, particularly sodium chloride (NaCl), has garnered attention in plant physiology due to its implications for enhancing stress tolerance. Recent findings indicate that EMS treatment can significantly mitigate leaf scorching in various plant varieties under saline conditions. For instance, the Winter Dawn variety exhibited minimal leaf scorching when treated with a 2% EMS dose in conjunction with 75 mM NaCl, while the Chandler variety showed similar resilience with a 1% EMS dose under 100 mM NaCl stress. This suggests that EMS-induced mutations may enhance physiological mechanisms that tolerance to salinity stress. Salinity stress is known to disrupt ion homeostasis, primarily by increasing sodium ion (Na⁺) concentrations, which can lead to physiological damage, including leaf scorching Guan *et al.* (2013). However, the application of EMS appears to facilitate adaptive responses that help maintain ion balance. For example, plants induced to salinity stress often exhibit a decrease in the potassium/sodium (K⁺/Na⁺) ratio, which is critical for maintaining cellular functions Gengmao *et al.* (2015). The ability of EMS-treated plants to sustain a higher K⁺/Na⁺ ratio under saline conditions may be linked to enhanced uptake and compartmentalization of potassium ions, thereby alleviating the toxic effects of sodium accumulation (Guan *et al.* 2013; Rivero *et al.* 2013). Moreover, the physiological responses to salinity stress can include alterations in metabolic pathways that enhance the production of protective compounds. Research indicates that salinity stress can stimulate the synthesis of secondary metabolites and protective enzymes, which play vital roles in stress mitigation Gengmao *et al.* (2015). The presence of these compounds in EMS-treated plants may contribute to their improved tolerance by enhancing cellular protection against oxidative stress and maintaining membrane integrity under high salinity conditions (Ahmadi and Sour, 2020). Additionally, the physiological adaptations observed in EMS-treated plants may involve changes in gene expression related to stress response pathways. For instance, studies have shown that salinity stress can induce specific gene expression profiles that help plants tolerate with ioni imbalances and osmotic stress (Jang *et al.* 2023; Ma *et al.* 2016). The mutagenic effects of EMS could potentially enhance the expression of these stress-responsive genes, leading to improved physiological resilience against salinity-induced leaf scorching.

Mineral analysis potassium K⁺

The interaction between ethyl methanesulfonate (EMS) chemical mutagen and induce salinity stress, particularly sodium chloride (NaCl), has been understanding plant physiological responses. Studies have shown that EMS treatment can enhance the resilience of plants under

saline conditions by modulating physiological parameters such as potassium (K). In the present findings, pooled data show that the maximum potassium ion was recorded in plants treated with a 2% EMS dose exposed to 50 mM NaCl. The minimum potassium ion was found in subjected to 100 mM NaCl. Moreover, the physiological implications of potassium levels under salinity stress are significant. Khoshbakht *et al.* (2018) highlighted that salinity stress typically leads to a reduction in potassium uptake due to the competitive inhibition of sodium ions, which can severely impact plant health. This is corroborated by the findings of Nikam *et al.* (2015) who emphasized that the accumulation of proline, a common response to salinity, is often accompanied by changes in potassium and sodium ion concentrations, affecting overall plant tolerance. In a similar Kamyab *et al.* (2016) demonstrated that maintaining a higher K⁺/Na⁺ ratio is crucial for enhancing salinity tolerance in plants, suggesting that EMS treatment may help in achieving this balance. Moneim *et al.* (2021) reported that EMS treatment improved salt stress tolerance in strawberry plants, which may also be relevant for understanding potassium retention under saline conditions. Furthermore, the role of potassium in physiological processes under salinity stress cannot be overstated. Studies have indicated that potassium can mitigate the adverse effects of salinity by improving ion homeostasis and enhancing photosynthetic efficiency Abbasi *et al.* (2015). For instance, Abbasi *et al.* (2015) found that potassium application significantly improved the growth and physiological parameters of maize hybrids under saline conditions, reinforcing the idea that potassium plays a vital role in plant resilience to salinity.

Mineral analysis sodium Na⁺

Research indicates that the application of EMS can modulate the physiological responses of plants under saline conditions. For instance, the present study has shown that runners exposed to high salinity (100 mM NaCl) exhibit maximum sodium content, while runners treated with EMS at a concentration of 2% under 50 mM NaCl conditions show reduced sodium content. This suggests that EMS treatment may enhance the plant's ability to manage sodium accumulation, potentially through mechanisms that involve increased tolerance to osmotic stress or improved ion homeostasis.

The mutagenic effects of EMS are characterized by a high frequency of mutations with relatively low lethality, making it a preferred choice for inducing genetic variability in plant breeding programs Hwang *et al.* (2014). The physiological implications of these mutations can be profound, as they may lead to enhanced traits such as salt tolerance. For example, EMS-

induced mutations have been linked to improved germination rates and seedling vigour under saline conditions, which are critical for the survival of plants in saline environments (Shamshad, 2023). However, the interaction of EMS with salinity stress may also influence the cytological aspects of plant development. Studies have demonstrated that EMS can induce chromosomal aberrations, which may affect the plant ability to tolerate with salinity stress (Mohammed and Ibrahim, 2017; Alam *et al.* 2022). The induction of such chromosomal changes could lead to variations in traits associated with salt tolerance.

Leaf area

The impact of salinity stress on leaf area in plants has been extensively studied, revealing significant variations in physiological responses among different species and cultivars. In the context of the winter Dawn and Chandler varieties, the application of 2% EMS dose has shown promising results in enhancing leaf area under NaCl-induced salinity stress. Specifically, the maximum Leaf Area Index (LAI) was recorded in runners treated with a 2% EMS dose against 75 mM NaCl, while a marked reduction in LAI was observed at 100 mM salinity levels. This phenomenon aligns with findings that indicate salinity stress adversely affects plant growth, particularly leaf area, which is one of the first indicators of stress in plants Corrado *et al.* (2021). The leaf area has consistently been reported to exhibit a positive correlation with net photosynthetic activity, as the expansion of the leaf is associated with an increase in the number and width of grana as well as a higher level of chlorophyll grana stacking Flore *et al.* (1985). In this study, the reduction in leaf area due to salinity aligns closely with previous findings on the adverse effects of salinity on leaf area and leaf numbers reported by Romero-Aranda *et al.* (1998) in citrus and Dong *et al.* (2007) in cucumber. The results also agree with Behera *et al.* (2012) in *Asteracantha longifolia*. The decline in leaf area observed at higher salt levels is a common response in bananas and has been extensively showed in various genotypes by several researchers (Gomes *et al.* 2004; Silva *et al.* 2009; Ferraz *et al.* 2008; Willadino *et al.* 2011). This decrease in leaf area may also be attributed to the restricted fixation of CO₂, which reduces the net assimilation rate and increases the production of reactive oxygen species (ROS) (Miller *et al.* 2009). Additionally, the decrease in leaf area can be explained by the diversion of energy needed for maintaining and activating metabolic processes associated with salinity tolerance, such as the synthesis of organic solutes, maintenance of membrane integrity, ion extrusion, and the regulation of ion transport and distribution within various organs and cells (Munns and Tester, 2010).

Salinity stress leads to physiological changes that can severely impair plant growth. For instance, it is well-documented that high salinity levels result in reduced leaf turgor pressure and impaired photosynthesis due to stomatal closure El-HenDawny *et al.* (2019). The reduction in leaf area can be attributed to both direct effects of ion toxicity and indirect effects on physiological processes such as nutrient uptake and water relations Ibrahim *et al.* (2016). In the case of the winter Dawn and Chandler varieties, the enhanced leaf area observed with EMS treatment may be linked to improved osmotic balance and reduced sodium ion accumulation, which are critical for maintaining cellular functions under stress Hamani *et al.* (2021). The ability of plants to maintain higher rates of leaf production than leaf senescence under saline conditions is crucial for sustaining leaf area and overall plant health Posada *et al.* (2011). This is particularly relevant in the context of the Chandler variety, which has shown resilience to salinity stress, suggesting a potential genetic basis for tolerance that could be exploited in breeding programs (Elhakem, 2020).

Chapter – 6

Summary and Conclusion



The study titled “Deciphering the effectiveness of induced mutation for salinity tolerance on Strawberry (*Fragaria x ananassa* Duch.) cv. Chandler and Winter Dawn” was conducted at Lovely Professional University in Jalandhar, Punjab in two seasons 2022-2023 and 2023-2024. The two years pooled findings of the research are categorized as follows:

5.1 Standardization of gamma irradiation and EMS LD₅₀ dose in Chandler and Winter Dawn

1. The LD₅₀ value in gamma irradiation determined based on lethality was found to be 40 Gy with the highest mortality recorded at 40 Gy in the Winter Dawn Chandler variety.
2. For EMS LD₅₀ value is determined based on lethality observed at the 3% EMS dose with the highest mortality recorded at the 3% EMS dose.

5.2 Gamma irradiation doses against induced salinity stress conditions in Winter Dawn variety

1. The maximum plant height (28.37 cm) was recorded in runners treated with a 30 Gy dose exposed to a 75 mM NaCl salinity level. The plant height was significantly lowest (18.93 cm) in control runners induced with a 100 mM salinity condition.
2. Chlorophyll content index was recorded as maximum (58.18) in runners treated with a 20 Gy dose when exposed to 50 mM NaCl and minimum chlorophyll content index (51.73) was recorded runners exposed to 75 mM salinity level.
3. The number of buds was recorded as maximum in 30 Gy gamma irradiation dose and exposed to 75 mM NaCl (28.16) and minimum buds (22.86) were found under runners against 100 mM NaCl.
4. The number of flowers was recorded as maximum (31.83) in 30 Gy gamma irradiation treated runners induced with 75 mM salinity level and a minimum number of flowers was found (24.23) under the 100 mM salinity condition.
5. The highest numbers of fruits (33.33) were obtained in runners treated with 30 Gy mutagen dose under 50 mM salinity level. However, the minimum number of fruits was recorded (24.25) in control runners induced with 100 mM Salinity condition.
6. The lowest number of days (48.33) taken to first bud initiation was achieved in strawberry runners treated with 30 Gy mutagen dose under 0 mM salinity condition. In runners induced with 75 mM salinity level (67.16).

7. After bud initiation early flower initiation at (56) days was recorded in 30 Gy mutagen dose induced with 50 mM salinity stress. In control runners, late flower initiation (75.16) was recorded under 75 mM salinity level.
8. Fruit initiation was recorded earlier at (79.83) days in 30 Gy gamma irradiation dose-treated runners under 100 mM salinity level. However, the late fruit initiation was recorded at (93) days in runners induced with 100 mM salinity condition.
9. The maximum fruit length (43.97 mm) was recorded in 30 Gy mutagen dose-treated runners under 100 mM salinity level and the minimum fruit length (32.12 mm) was recorded in runners against 75 mM salinity level.
10. The diameter of fruit was maximum recorded (33.47 mm) in a 30 Gy gamma irradiation dose treated runners induced with 50 mM salinity condition. While the minimum fruit diameter (25.22 mm) was recorded in runners induced with 75 mM salinity level.
11. Fruit weight was recorded as maximum (13.32 gm) in 30 Gy mutagen dose treated runners with 50 mM salinity condition. The minimum was found (10.17 gm) under the 100 mM salinity level.
12. The fruit yield was recorded as maximum (350.33 gm) in mutagen treated runners with a 30 Gy dose induced with a 50 mM salinity level. While minimum fruit yield (257.16 gm) was recorded under 100 mM salinity level.
13. In Gamma Irradiation dose 20 Gy showed maximum shoot fresh weight (14.53 gm) under the 50 mM salinity level. However, the minimum shoot fresh weight (10.18 gm) was recorded in runners against 100 mM salinity stress.
14. Shoot Dry Weight was maximum recorded (3.51 gm) in runners treated with 20 Gy gamma irradiation dose induced with 50 mM salinity level. The minimum shoot dry weight (1.98 gm) was recorded in runners induced with 75 mM NaCl salinity condition.
15. The root fresh weight was maximum recorded (14.59 gm) in treated runners with 30 Gy dose exposed to 75 mM NaCl. Minimum root fresh (10.57 gm) was recorded in runners induced with 100 mM NaCl.
16. Root dry weight was recorded as maximum (4.27 gm) with a 20 Gy dose in mutagen treated runners induced with 75 mM NaCl stress and minimum root dry weight recorded (2.80 gm) found in runners against 75 mM salinity level.
17. The number of leaves maximum recorded (11.83) in 30 Gy gamma irradiation dose induced against 50 mM NaCl. While the minimum leaves were found (8) in runners exposed to 100 mM salinity condition.

18. Leaf relative water content was maximum recorded (81.69 %) in 30 Gy gamma irradiation dose induced with 75 mM salinity stress conditions. However minimum LRWC recorded (72.37 %) in intreated runners induced with 100 mM salinity level.
19. Electrolyte leakage was recorded lowest (24.56 %) with a 30 Gy dose exposed to 50 mM salinity level and the highest was recorded (34.17 %) in plants exposed to 100 mM salinity condition.
20. The proline content was maximum found (3.82 $\mu\text{mol/gm}$) in runners treated with 30 Gy dose exposed to 50 mM salinity level and minimum proline was found (2.54 $\mu\text{mol/g}$) in runners induced with 75 mM salinity condition.
21. The Membrane Stability Index was recorded as maximum (80.99 %) in mutagen dose 20 Gy exposed to 50 mM salinity level, as well as minimum proline was found (74.4 %) in runners under 100 mM salinity level.
22. Leaf scorching was found highest (22.98 %) in runners induced with 100 mM salinity level, However, minimum leaf scorching was recorded in runners treated with 30 Gy dose (13.94%) under 75 mM salinity condition.
23. Malonaldehyde content was recorded maximum (9.99 $\mu\text{mol/g}$) in runners treated with 20 Gy dose of gamma radiation under 100 mM NaCl stress, while the minimum malonaldehyde content (2.05 $\mu\text{mol/g}$) was found in runners exposed to 100 mM NaCl stress.
24. Mineral Analysis maximum sodium Na^+ content was recorded in runners induced with 100 mM NaCl (99.45 ppm), while the minimum Sodium Na^+ content was recorded (24.51 ppm) in runners treated with 30 Gy mutagen dose against 50 mM NaCl.
25. In mineral analysis potassium K^+ was recorded as maximum (27.76 ppm) in runners treated with 30 Gy gamma irradiation dose against 50 mM salinity level, minimum potassium K^+ recorded (12.15 ppm) in runners induced with 100 mM salinity level.

5.3 Chemical mutagen doses against induced salinity stress conditions in Winter Dawn variety

1. Chemical mutagen EMS treated runners with 2 % EMS dose against 50 mM NaCl showed maximum plant height (24.82 cm) while the minimum plant height was found (20.15 cm) in runners exposed to 100 mM salinity condition.
2. In 2% EMS mutagen treated runners chlorophyll content index was recorded it was found maximum (55.74) under 75 mM salinity level. Minimum Chlorophyll content index value recorded (51.8) in runners under 100 mM salinity stress.

3. The number of buds was maximum recorded (26.16) in 2% EMS mutagen dose treated runners exposed with 50 mM salinity level, minimum buds recorded (20.5) in runners induced with 100 mM salinity condition.
4. Flower count was the maximum recorded (27.22) in chemical mutagen 2% EMS dose induced with 50 mM salinity level. However, the minimum number of flowers (20.5) was recorded in plants exposed to 100 mM NaCl.
5. Chemical mutagen 2% EMS dose showed maximum number of fruits (29) under 50 mM salinity level, while minimum fruit was recorded (21.82) in runners induced with 75 mM salinity level.
6. Early bud initiation was recorded at (30.88) days in runners treated with 2% EMS dose exposed to 50 mM NaCl and late initiation (50.83) was recorded in runners treated with 100 mM salinity condition.
7. Flower initiation was recorded early at (54.78) 2% EMS dose under 50 mM NaCl, However, late flower initiation was found (63.66) in runners under 100 mM salinity level.
8. EMS mutagen dose at 2% EMS shows early fruit initiation at (74.66) induced with 50 mM salinity condition, while the late flower found at (89.16) runners induced with 100 mM salinity conditions.
9. Maximum fruit length (89.46 mm) was recorded in 2% EMS dose under 75 mM induced salinity stress. However minimum fruit length (74.66 mm) was recorded in runners induced with 100 mM salinity level.
10. In chemical mutagen 2% EMS induced with 75 mM salinity condition shows maximum diameter (30.49 mm) while the minimum was recorded (24.16 mm) at runners exposed to 100 mM salinity level.
11. The weight of fruit was the highest recorded (12.32 gm) in 2% EMS dose under 50 mM NaCl stress condition as well as the lowest weight (9.29 gm) found in runners induced with 100 mM NaCl salinity stress.
12. Fruit yield was found to be maximum (350.66 gm) in mutagen treated runners with 2% EMS dose and exposed to 50 mM NaCl stress. The minimum yield was found (256.66 gm) under runners induced with 100 mM salinity condition.
13. Shoot fresh weight maximum recorded (12.90 gm) in 2% EMS dose induced with 75 mM salinity condition also the minimum weight (8.38 gm) recorded in runners exposed to 100 mM salinity level.

14. The dry weight of the shoot recorded a maximum (3.37 gm) in runners treated with a 2% EMS dose under 75 mM NaCl salinity. However, the minimum weight (1.89 gm) was found in runners with 75 mM NaCl stress.
15. Root fresh weight recorded maximum (14.35 gm) in 2% EMS dose combined with a 75 mM NaCl salinity condition. The minimum root fresh weight recorded (9.63 gm) in runners exposed to 100 mM salinity condition.
16. The weight of root recorded maximum (3.42 gm) was observed in runners treated with a 2% EMS dose and while the lowest root dry weight was recorded under 100 mM salinity condition (1.13 gm).
17. The leaves number was recorded as maximum (12.76) in runners treated with a 2% EMS dose under 50 mM NaCl (12.76) while the minimum number of leaves was observed in runners exposed to 100 mM salinity (8.83).
18. The highest LRWC was recorded in plants treated with a 2% EMS dose combined with 50 mM NaCl (80.96%), while the lowest LRWC was observed in runners exposed to 100 mM NaCl (73.21%).
19. The minimum electrolyte leakage was recorded in plants treated with 2% EMS and exposed to 100 mM NaCl (25.71%), However, the maximum electrolyte leakage was observed in plants exposed to 100 mM NaCl (36.23%).
20. Proline content was recorded as maximum (4.22 $\mu\text{mol/g}$) in runners treated with a 1% EMS dose exposed to 50 mM NaCl, while the minimum proline content was observed in runners exposed to 100 mM NaCl (2.48 $\mu\text{mol/g}$).
21. The minimum malondialdehyde content was recorded in runners treated with a 2% EMS dose exposed to 100 mM NaCl (1.87 nmol/g), while the maximum was observed in runners subjected to 100 mM salinity level (10.68 nmol/g).
22. Maximum Leaf Area Index was observed in runners treated with a 2% EMS dose induced with 75 mM NaCl (21.46 mm), while the minimum LAI was recorded in runners exposed to 100 mM induced salinity stress (10.68 mm).

5.4 Gamma irradiation doses against induced salinity stress conditions in Chandler variety

1. Maximum plant height was recorded in runners treated with 30 Gy of gamma radiation against 50 mM NaCl (23.13 cm), However, the minimum plant height was observed in plants exposed to 100 mM NaCl (17.6 cm).

2. Chlorophyll content index was recorded in the 30 Gy gamma irradiation dose treated runners exposed with 50 mM NaCl (59.8). The minimum chlorophyll content index was observed in runners induced with 100 mM NaCl (45.8).
3. The number of buds was maximum recorded (24.36) in runners treated with a 30 Gy dose exposed to 50 mM NaCl. While the minimum was found in runners exposed to 100 mM salinity level (17.33).
4. Flowers were recorded maximum in plants treated with a 30 Gy dose exposed to 50 mM salinity condition (27.33), Conversely, the minimum flower count was observed in plants exposed to 100 mM salinity level (19.87).
5. Maximum number of fruits was recorded in 30 Gy gamma irradiation treated runners induced with 50 mM salinity level (28.66), However, the minimum was observed in the runners exposed to 100 mM salinity condition (21.14).
6. Early bud initiation was recorded in the treatment of 30 Gy dose exposed with 75 mM NaCl (39.33), while late bud initiation occurred in 100 mM highest salinity level (59.39).
7. Runners treated with a 30 Gy dose induced to 50 mM NaCl exhibited early flower initiation (50.48), whereas runners exposed to 100 mM salinity conditions showed delayed flower initiation (70.5).
8. Earliest fruit initiation was recorded in plants treated with a 30 Gy dose exposed to 50 mM NaCl (79), while the late fruit initiation was recorded in plants exposed to 100 mM salinity condition (99.39).
9. Early bud initiation was recorded in the treatment of 30 Gy dose exposed with 75 mM NaCl (39.33), while late bud initiation occurred in 100 mM highest salinity level (59.39).
10. The early flower initiation recorded in runners treated with a 30 Gy dose induced to 50 mM salinity stress (50.48), whereas runners exposed to 100 mM salinity conditions showed delayed flower initiation (70.5).
11. Gamma Irradiation-treated runners, induced with varying levels of salinity, showed the earliest fruit initiation was recorded in plants treated with a 30 Gy dose exposed to 50 mM NaCl (79), while the late fruit initiation was recorded in plants exposed to 100 mM salinity condition (99.39).
12. Maximum fruit length was observed in runners treated with a 30 Gy dose exposed to salinity stress 75 mM level (39.33 mm). The minimum fruit length was recorded in runners exposed to the highest salinity stress 100 Mm salinity level (29.82 mm).

13. The maximum fruit diameter was recorded in plants treated with a 30 Gy dose exposed to 50 mM NaCl (32.94 mm). Minimum fruit diameter was observed in runners exposed to 100 mM salinity condition (22.37 mm).
14. Highest fruit weight of (11.51gm) was observed in the treatment combination of 30 Gy gamma irradiation induced with 50 mM salinity conditions. While the lowest fruit weight of (8.51gm) was recorded in 100 mM NaCl.
15. The highest fruit yield was recorded in plants treated with a 30 Gy dose induced with 50 mM salinity level (380.66 gm). However, the lowest yield was observed in runners exposed to 100 mM highest salinity condition (252.83 gm).
16. Shoot fresh weight maximum recorded in runners treated with a 30 Gy dose under 50 mM NaCl achieved the highest shoot fresh weight (12.21 gm). While the lowest was recorded in plants under 100 mM NaCl (7.22 gm).
17. The Dry weight of shoot maximum recorded in runners treated with a 30 Gy dose exposed to 75 mM NaCl (3.52 gm). While the minimum shoot dry weight was observed in without mutagen-treated runners against 100 mM NaCl (1.85 gm).
18. Maximum root fresh weight was recorded in plants treated with a 20 Gy dose exposed to 75 mM NaCl (13.29 gm). The minimum root fresh weight was observed in plants exposed to 100 mM NaCl (10.66 gm).
19. The maximum root dry weight in the 20 Gy mutagen dose treated runners against 75 mM salinity condition (4.35 gm). However, minimum root dry weight was recorded in runners against 50 mM salinity level (2.71 gm).
20. Maximum number of leaves was observed in plants treated with a combination of 20 Gy gamma irradiation treated runners against 50 mM NaCl (10.33). In contrast, the minimum number of leaves was recorded in plants exposed to 100 mM salinity level (5.96).
21. LRWC of was recorded maximum in runners treated with a combination of 20 Gy gamma irradiation against 50 mM NaCl (84.75%), The minimum LRW was observed in plants exposed to 100 mM NaCl (66.04%).
22. The lowest electrolyte leakage was observed in plants treated with 20 Gy dose exposed to 100 mM NaCl (26.31%), Highest electrolyte leakage was recorded in plants exposed to 100 mM NaCl (34.21%).
23. Proline content was maximum recorded, in runners treated with mutagen 30 Gy dose exposed to 50 mM NaCl (3.29 $\mu\text{mol/g}$) The lowest proline content was recorded in runners exposed to 100 mM salinity level (1.78 $\mu\text{mol/g}$).

24. Maximum Membrane Stability Index was observed in the treatment of 20 Gy dose against 50 mM NaCl (84.37 %), The minimum MSI of was recorded in the without mutagen treated runners against 100 mM NaCl treatment (79.10 %).
25. Minimum leaf scorching was recorded in the combination of 20 Gy dose against 75 mM NaCl (12.28 %). The maximum scorching was observed in the 100 mM NaCl treatment (44.76 %).
26. Minimum malonaldehyde content was recorded in 20 Gy dose treated runners against 75 mM NaCl (1.50 $\mu\text{mol/g}$) and maximum malonaldehyde content was found under runners induced with 100 mM salinity condition (11.59 $\mu\text{mol/g}$).
27. The maximum leaf area was recorded in runners treated with a 30 Gy dose against 100 mM NaCl (24.79 mm), while the minimum leaf area was recorded in runners exposed to 100 mM NaCl (10.84 mm).

5.4 Chemical mutagen doses against induced salinity stress conditions in Chandler variety

1. The maximum plant height of (20.96 cm) was recorded in the treatment combination of 1% EMS dose exposed with 75 mM NaCl, while the minimum height of (16.53 cm) was observed in runners induced with 75 mM NaCl stress.
2. Highest chlorophyll content index was recorded at (65.35) in plants treated with 2% EMS and exposed to 100 mM salinity stress, while the lowest was observed (40.72) in without mutagen-treated plants exposed to 100 mM salinity level.
3. Number of buds recorded maximum (24.36) in plants treated with a 2% EMS dose exposed to 50 mM NaCl, while the minimum was noted at (17.33) in runners exposed to 100 mM induced salinity stress.
4. In chemical mutagen EMS treated runners flowers were maximum found (27.66) in 1% EMS dose induced with 50 mM salinity level. However, minimum flower occurred (21.57) in runners exposed with 100 mM NaCl stress.
5. The highest number of fruits was recorded (31.46) in plants treated with a 2% EMS dose exposed to 50 mM NaCl, while the lowest fruits (23.83) was observed in runners induced to 100 mM salinity level.
6. Early bud initiation was recorded at (48.97) days in runners treated with 2% EMS against 50 mM NaCl, while the late bud initiation was observed at (61) days recorded in without mutagen treated runners induced with 100 mM salinity stress.

7. In chemical mutagen treated runners flower initiation early recorded at (62.70) in 2% EMS induced with 50 mM NaCl, while late flower initiation was observed at (78.83) days in 100 mM salinity condition.
8. Fruit imitation was recorded early at (90.21) day in 2% EMS dose against 50 mM NaCl, while late fruit initiation was noted at (104.39) days in runners under the 100 mM salinity stress.
9. Length of fruit was recorded maximum (41.15 mm) 1 % EMS induced with 100 mM salinity stress. However, minimum fruit length was recorded at (28.88 mm) in the without mutagen treated runners exposed with 100 mM salinity stress.
10. Fruit diameter recorded maximum (29.64 mm) in chemical mutagen 1% EMS treated runners induced with 100 mM salinity level. However, the minimum fruit diameter was noted at (24.83 mm) in runners exposed to 100 mM salinity level.
11. Fresh weight of shoot recorded maximum (14.73 gm) in mutagen 1% EMS dose treated runners against 50 mM salinity level. While the minimum weight was found (11.21 gm) in runners induced with 100 mM NaCl salinity dose.
12. The chemical mutagen treated runners with 2 % EMS dose induced with 50 mM salinity level showed maximum shoot dry weight (3.76 gm), However, minimum shoot dry weight (2.29 gm) in the runners induced with 100 mM salinity level.
13. The maximum root fresh weight (16.59 gm) was observed in plants treated with 2% EMS exposed to 100 mM NaCl, while the minimum root fresh weight of (12.39 gm) was recorded in runners exposed to 100 mM salinity condition.
14. Root dry weight recorded maximum (4.25 gm) was observed in the treatment with 2% EMS dose induced with 100 mM NaCl, while the minimum root dry weight (2.89 gm) was recorded in runners induced with 100 mM salinity stress.
15. Leaves was found maximum (13.5) in EMS mutagen treated runners with 1% dose induced with 50 mM salinity concentration. While the minimum leaves (7.66) recorded in runners exposed to 100 mM salinity stress.
16. The leaf relative water content was recorded maximum (86.63 %) was recorded in runners treated with 2% EMS dose induced with 50 mM NaCl, while the minimum LRWC of (77.26 %) was observed in plants exposed to 100 mM salinity level.
17. Electrolyte leakage minimum was observed (17.85 %) in plants treated with 2% EMS combined with 50 mM NaCl, while the maximum electrolyte leakage (41.12 %), was recorded in the runners exposed to 75 mM NaCl.

18. The proline content was recorded maximum (3.94 $\mu\text{mol/g}$) in runners treated with 1% EMS dose induced with 75 mM NaCl, while the minimum was (1.50 $\mu\text{mol/g}$) in the runners exposed to 100 mM salinity level.
19. Membrane stability index recorded maximum (85.65 %) in runners treated with 2% EMS exposed to 50 mM salinity level, while the minimum MSI of (77.79 %) was recorded in runners exposed to 100 mM salinity concentration.
20. In contrast, leaf scorching found maximum (27.25 %) runners induced with 100 mM salinity stress. However minimum leaf scorching (15.75 %) found in runners with 0 mM NaCl.
21. Fruit weight was found maximum (21.85 gm) in 1% EMS dose induced with 50 mM salinity level. While the minimum fruit weight recorded (9.97 gm) was observed in runners exposed to 75 mM salinity condition.
22. Yield of fruit was recorded maximum (410.85 gm) in EMS treated runners with 1 % dose induced with 100 mM salinity condition, Minimum yield observed (176.87 gm) in runners induced with 100 mM stress.
23. Malonaldehyde content was found maximum (9.63 %) in untreated runners induced with 100 mM salinity stress. Minimum malonaldehyde content was recorded (1.53 %) in runners treated with 1% EMS dose against 75 mM salinity stress.
24. In mineral analysis sodium was found minimum (19.02 ppm) in runners treated with 2% EMS dose exposed with 50 mM salinity level. However, maximum sodium content was recorded (117.32 ppm) in runners against 100 mM salinity condition.
25. Potassium was found maximum (19.85 ppm) in plant treated with 2% EMS dose under 50 mM salinity concentration. Minimum potassium was found (9.72 ppm) in runners against 100 mM salinity level.

The varietal response towards salinity after physical mutagen treatments

- In the Winter Dawn and Chandler varieties, malondialdehyde content was estimated. Runners treated with 20 Gy dose of the mutagen exhibited the best malondialdehyde content under 100 mM high salinity conditions in the Winter Dawn variety. However, runners of the Chandler variety treated with the same mutagen dose showed the best malondialdehyde content under 50 mM salinity conditions. Regarding varietal response, Winter Dawn demonstrated positive result in malondialdehyde condition.
- The electrolyte leakage was recorded in Winter Dawn and Chandler varieties. In the winter Dawn variety 30 Gy mutagen dose treated runners exposed to 50 mM NaCl

showed the best results in electrolyte leakage. However, in the Chandler variety mutagen dose 20 Gy treated runners tolerate the highest salinity level 100 mM and showed the best results in electrolyte leakage as compared to the Winter Dawn variety

- Proline content was recorded in Winter Dawn and Chandel varieties it was observed best in 30 Gy mutagen dose treated runners induced with 50 mM salinity condition.
- In the Winter Dawn and Chandler variety membrane stability index was recorded best in mutagen treated runners with a 20 Gy dose exposed to a 50 mM salinity level
- Plant height was recorded as the best in the Winter Dawn variety when runners treated with a 30 Gy mutagen dose were exposed to high salinity conditions 75 mM. However, in the Chandler variety runners treated with the same mutagen dose showed the best plant height under 50 mM salinity conditions. Regarding varietal response, Winter Dawn exhibited the best plant height, even under high salinity conditions.
- The chlorophyll content was recorded best in winter Dawn variety runners treated with 30 Gy dose under the highest salinity condition 100 mM. However, in the Chandler variety same mutagen dose treated runners showed the lowest chlorophyll as compared to the Winter Dawn variety under 50 mM salinity level
- In Winter Dawn and Chandler variety mutagen treated runners with 30 Gy dose against the highest 100 mM salinity condition showed the best leaf area index.

The varietal response towards salinity after chemical mutagen EMS treatments

- Plant height in the Winter Dawn variety was recorded as best in runners treated with 2% EMS induced with 50 mM salinity stress. However, in the Chandler variety, the best plant height was observed in runners treated with 1% EMS dose under 75 mM salinity stress. These findings indicate that the Chandler variety demonstrated better tolerance to salinity stress, resulting in best plant height compared to the Winter Dawn variety.
- The malondialdehyde content in the Winter Dawn variety was found best in runners treated with 2% EMS dose exposed to 100 mM NaCl. However, the best malondialdehyde content in the Chandler variety was observed in runners treated with 1% EMS dose under 75 mM salinity conditions. Regarding varietal response, the Winter Dawn variety exhibited the highest malondialdehyde content, even under high salinity levels.

- EMS-treated runners of the Winter Dawn variety showed positive results in electrolyte leakage 2% EMS dose induced with 100 mM NaCl. However, in the Chandler variety best electrolyte leakage results were recorded in runners treated 1% EMS dose induced with 50 mM salinity condition. In electrolyte leakage, the winter variety showed the best result tolerating a high salinity level as compared to the Chandler variety.
- In EMS doses the 1% EMS dose treated runners of Winter Dawn and Chandler variety tolerate salinity stress in Winter Dawn it tolerates salinity 50 mM NaCl showed positive results in proline content. However, the highest proline content was recorded in the Chandler variety in runners treated with 1% EMS dose exposed with 75 mM NaCl.
- Proline content was estimated in both the Winter Dawn and Chandler varieties. In the Winter Dawn variety, the highest proline content was observed in runners treated with 1% EMS dose under 50 mM salinity conditions. In the Chandler variety, the highest proline content was recorded in runners treated with the same EMS dose under 75 mM salinity conditions. Overall, the Chandler variety exhibited higher proline content than the Winter Dawn variety under a high 75 mM salinity condition.
- The membrane stability index (MSI) recorded the highest values in runners treated with a 2% EMS dose under 50 mM salinity conditions. Similarly, in the Chandler variety, the best MSI was observed in runners treated with a 1% EMS dose and exposed to 50 mM salinity. In comparison, both varieties demonstrated optimal MSI under these conditions.
- Chlorophyll content index were found to be best in the Winter Dawn variety runners treated with 2% EMS dose under 75 mM NaCl salinity. However, in the Chandler variety, the best Chlorophyll content index value was observed with 2% EMS dose under 100 mM salinity conditions. These findings indicate that the Chandler variety exhibited superior chlorophyll levels even under high salinity conditions compared to the Winter Dawn variety.
- Leaf area index was recorded best in Winter Dawn and Chandler variety runners treated with 2% EMS dose exposed in 75 mM salinity condition.

Conclusion

It can be finally concluded that lower doses of ^{60}Co gamma rays, specifically at 30 Gy and 20 Gy were effective in tolerating salinity stress up to 100 mM NaCl and proved most effective for mutagenesis in the Winter Dawn and Chandler strawberry varieties. Similarly, in chemical mutagen doses, EMS 1% and EMS 2% showed positive effects against salinity stress and were also most effective for mutagenesis in these varieties. Runners treated with these physical and chemical mutagen doses demonstrated positive results in physiological observations, such as reduced malondialdehyde content, electrolyte leakage, leaf scorching, and improved shoot and root fresh and dry weights. Additionally, they exhibited increased membrane stability index, proline content, and leaf relative water content. In morphological observations, the treatments resulted in maximum leaf area, number of leaves, chlorophyll content index, fruit length and fruit diameter and plant height. The study also emphasized the significance of conducting dose determination studies prior to embarking on large-scale experiments, considering the variations in radiation sensitivity observed among different cultivars.

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