

# Using Ion Beam Irradiation to Evaluate the Morphological and Chemical Characteristics of Jasmine Cultivar Rice (*Oryza Sativa L.*) Under Drought Condition

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**Abstract:** The continuous flow of water is crucial for the growth of rice, a staple meal in many nations of Asia. Hence, creating a rice genotype that is resistant to drought and low rainfall would help ensure the steady availability of rice around the world. The *Oryza sativa L.* genotype of Jasmine rice was evaluated for a number of drought-tolerance scenarios throughout this study. The genotype seeds were exposed to 350 Gy of gamma radiation before being placed into a basic Murashige and Skoog (MS) growth medium that was half strength and had been enhanced with polyethylene glycol 6000 (PEG 6000) concentrations of 20, 10, and 0%. The PEG 6000 had an impact on the lengths of the shoots and roots, weights of the dried and fresh seedlings, amount of total carbohydrates and chlorophyll, and proline build-up among other things. The average shoot length as well as the weight of the seedlings were highest among the seeds that had not been irradiated, whereas the average root length and proline concentration were highest among the seeds that had been irradiated. Additionally, the proline content increased as a result of the rise in PEG 6000 concentration. These results suggest that radiation has a beneficial effect on the ability of rice plants to withstand drought. In our opinion, the creation of a dependable genotype of rice that is drought-tolerant will enable increased food security through increased rice yield. Thus, further studies emphasising on this issue need to be in the pipeline.

**Keywords:** Rice, Drought Tolerance, Irradiation, Proline.

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## 1. Introduction

The majority of the world's populace relies on rice as their primary food supply (Fukagawa & Ziska, 2019). The irrigated and rain-fed rice farm of Asia support over 85% of the global rice production despite being frequently impacted by abiotic problems (Hollaus et al., 2022). The creation of rice varieties resistant to drought is crucial from a financial perspective for regions with insufficient irrigation systems and scant rainfall (Kadhimi et al., 2014). Many well-known genotypes were taken into consideration for the production of drought-tolerant rice plants to aid plant breeders encountering such a condition (Saad-Allah et al., 2021). The efficient methods made possible by biotechnology are less expensive and time-consuming than conventional propagation methods. Tissue culture has recently gained attention as a trustworthy and affordable solution for increasing plant life's capacity to withstand stress, which would subsequently boost agricultural yield. Plant tolerance to drought and salt in the context of biotic and abiotic stresses can be improved using mediators, like polyethylene glycol (PEG)

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and sodium chloride (NaCl), respectively (Ahmar et al., 2020). Polyethylene glycol (PEG) is a suitable osmoticum for application in a hydroponic root system due to its high molecular weight and resistance to plant tissue permeability (Robin et al., 2021). (Joshi et al., 2011) investigated how varying amounts of PEG-induced water stress affected the growth, proline accumulation, and water constituent of several rice cultivars (*Oryza sativa*). Numerous drought-tolerant plants have been found thanks to the usage of PEG to stimulate water stress (Mehmandar et al., 2023).

Crop genotypes can be changed using the method of mutation induction. This procedure makes it easier to introduce traits that do not exist during plant growth. Irradiation is typically regarded as the most efficient method of mutation induction when compared to other methods (Holme et al., 2019). As per the results of numerous studies, it is possible to generate rice plants that are resistant to drought by channelling the physiological traits that distinguish rice genotypes. However, more thorough studies are needed on the physiological system of Jasmine rice seeds in connection to drought resistance. In light of this, research on PEG and irradiation impacts on the morphological and biochemical characteristics of Jasmine rice seed genotypes should be carried out to set a standard for tolerance to droughts.

## 2. Methods And Materials

Caesium-137 gamma radiation at a dose of 350 Gy was applied to the seeds of Jasmine rice (Kadhimi et al., 2016). The seeds were then treated with an ethanol percentage of 70% for three minutes after being rinsed with distilled water three times. The items were then sterilised using a 0.1% concentration of mercury (II) chloride (HgCl<sub>2</sub>) followed by TWEEN® 20 for five minutes. The seeds were put in a flask and left to germinate for a week at 25°C after the sterilising chemicals had been removed using distilled water. The resultant seedlings were then transplanted into 15 by 2.5 cm beakers that contained either 0, 10, or 20% of PEG 6000 along with a half-strength Murashige and Skoog (MS) media. The beakers were then placed in a climatic chamber for 16:8 photoperiods of light-to-dark with a 1000lux light intensity and a temperature of 25 ± 2°C. After a germination span of one month, the length of the roots, length of the shoots, and the weight of the fresh plants were measured. The plant dry weights were calculated after the plant tissues had been dried at 72°C for a couple of days (Prajubmon et al., 2009; Wani et al., 2010). Proline concentration levels were the main subject of a 1973 investigation by (Bates et al., 1973). In order to estimate the total content of sugar; which is the concentration of carbohydrate; (Herbert et al., 1971) used the phenol sulphuric acid approach without identifying specific sugar ingredients.

### 2.1. Design of the experiment and numerical analysis

For this experiment, the completely randomised design (CRD) method was used. Gamma radiation is used in the procedure at one level and excluded at a different level. Polyethylene glycol 6000 (PEG 6000) is present in three different concentrations: 0%, 10%, and 20%. In addition, 10 samples were used in three replications. Statistical Analysis System (SAS) software (Release 9.1 for Windows) was used for the analysis of variance (ANOVA) after the normality test was conducted. The results of Duncan's multiple range (DMR) test showed considerable differences in the efficacy of the various therapies.  $P \leq 0.05$  was the point to determine which difference was the least significant.

### 3. Results

#### 3.1. Shoot Length

The average rise in shoot length for the non-irradiated seedlings is noticeably greater than for the irradiated seedlings, as shown in Figure 1.

The statistics shown in Figure 1 also support the claim that PEG 6000 treatment during seedling development decreased shoot length, whether or not gamma rays were used.

The non-irradiated plants without PEG 6000 treatment (0%) had longer shoots than the irradiated plants because of the association between irradiation and interaction. With a 20% PEG 6000 concentration, the non-irradiated plants likewise produced shoots that were longer on average than those of the irradiated plants.

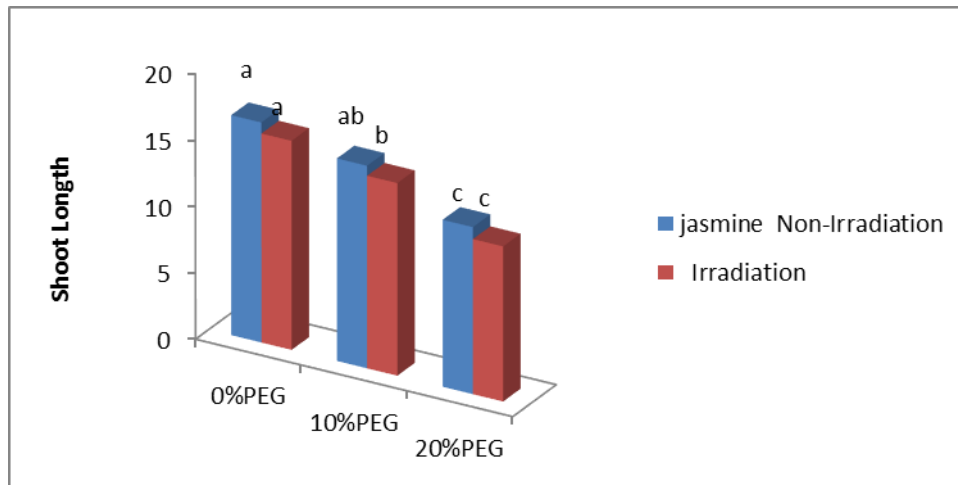


Figure 1: Average length of Jasmine plant shoots derived from non-irradiated (0 Gy) and irradiated (350 Gy) seeds treated with 20, 10, and 0% PEG 6000.

#### 3.2. Root Length

Figure 2 shows that the increase in root length for irradiated plants is higher than that of non-irradiated plants. It also shows a substantial reduction in root length at PEG 6000 concentrations of 20 and 10% than 0%.

The association between PEG 6000 and irradiation resulted in irradiated plants treated with a 20% PEG 6000 concentration having longer roots on average than non-irradiated plants treated with the same PEG 6000 concentration. The data show that both genotypes respond to PEG 6000 stress similarly, regardless of whether they receive irradiated or non-irradiated therapy. As PEG 6000 content increases, both plant height and root length are reduced.

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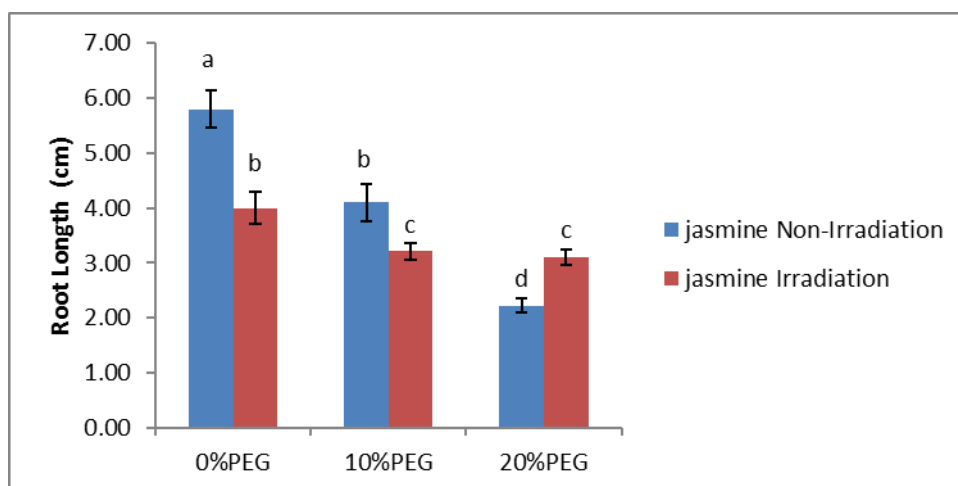


Figure 2: Average length of Jasmine plants root derived from non-irradiated (0 Gy) and irradiated (350 Gy) seeds treated with 20, 10, and 0% PEG 6000.

### 3.3. Fresh Weight

As depicted in Figure 3, the increase in average fresh weight for the non-irradiated JASMINE plants is bigger than it is for the irradiated JASMINE plants.

The collected data indicates that both irradiated and non-irradiated plants respond similarly to PEG 6000 stress. Any increase in PEG 6000 concentration results in a reduction in fresh weight.

For non-irradiated plants, the relationship between PEG 6000 and irradiation resulted in the highest weight of fresh plants at PEG 6000 0%, but a lower fresh weight was seen for the irradiated plants treated with the same PEG 6000 treatment. The irradiated and non-irradiated plants exposed to PEG 6000 concentrations of 10% and 20%, however, showed no discernible differences.

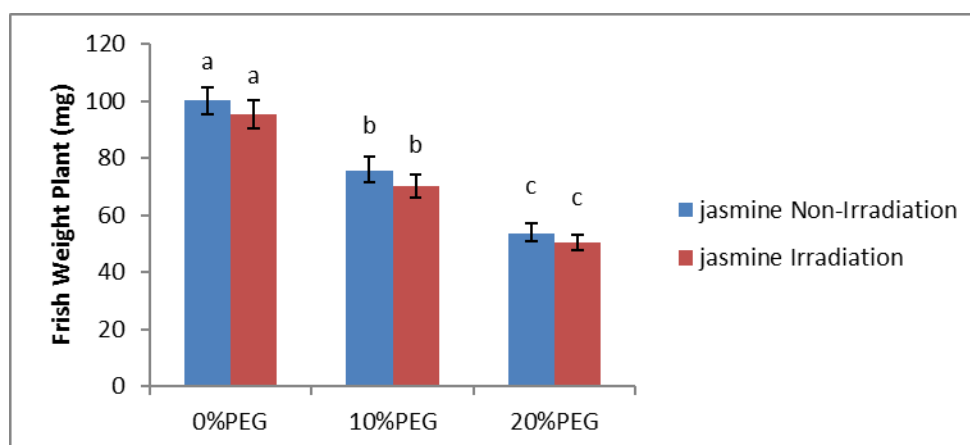


Figure 3: Average weight of fresh Jasmine plants derived from non-irradiated (0 Gy) and irradiated (350 Gy) seeds treated with 20, 10, and 0% PEG 6000.

### 3.4. Dry Weight

The average dry weights of the irradiated and non-irradiated plants are similar, as shown in Figure 4, with no obvious differences.

Additionally, Figure 4 shows that at PEG 6000 concentrations of 20 and 10%, the weight loss by dry plants is higher than at 0%.

Because of the relationship between PEG 6000 treatment and irradiation, dry non-irradiated Jasmine plants with 0% PEG 6000 weighed more than irradiated plants with a same PEG 6000 treatment. The amassed data shows that at a 20% PEG 6000 treatment concentration, irradiated Jasmine plants provide more dry weight than non-irradiated plants.

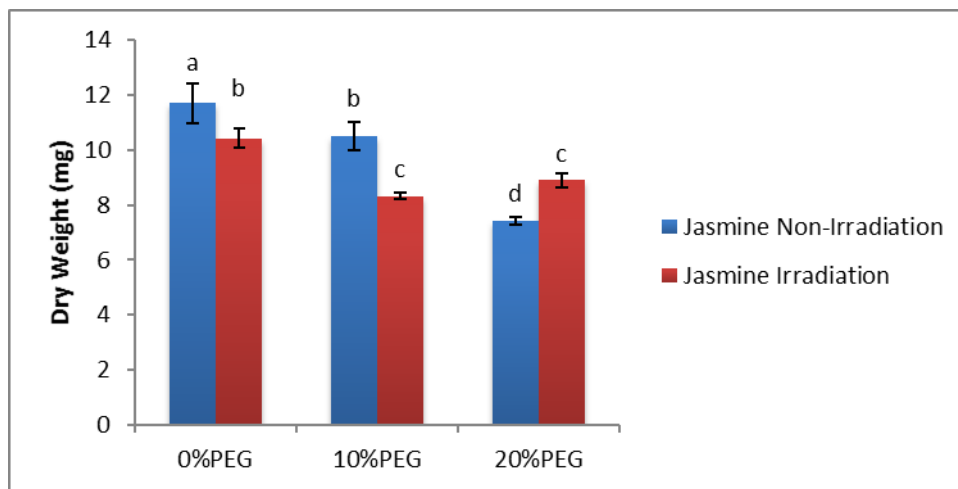


Figure 4: Average weight of dry Jasmine plants derived from non-irradiated (0 Gy) and irradiated (350 Gy) seeds treated with 20, 10, and 0% PEG 6000.

### 3.5. Overall Chlorophyll Composition

Figure 5 shows that regardless of the PEG 6000 concentration used, there were no notable differences between the total chlorophyll in irradiated and non-irradiated plants.

In contrast to a 0% PEG 6000 treatment, at PEG 6000 treatment concentrations of 20 and 10% (Figure 5), the overall chlorophyll composition significantly decreased in genotypes that had been exposed to radiation and those that had not.

At a PEG 6000 concentration of 20%, the relationship between PEG 6000 treatment and irradiation resulted in irradiated plants having a higher total chlorophyll content than non-irradiated plants.

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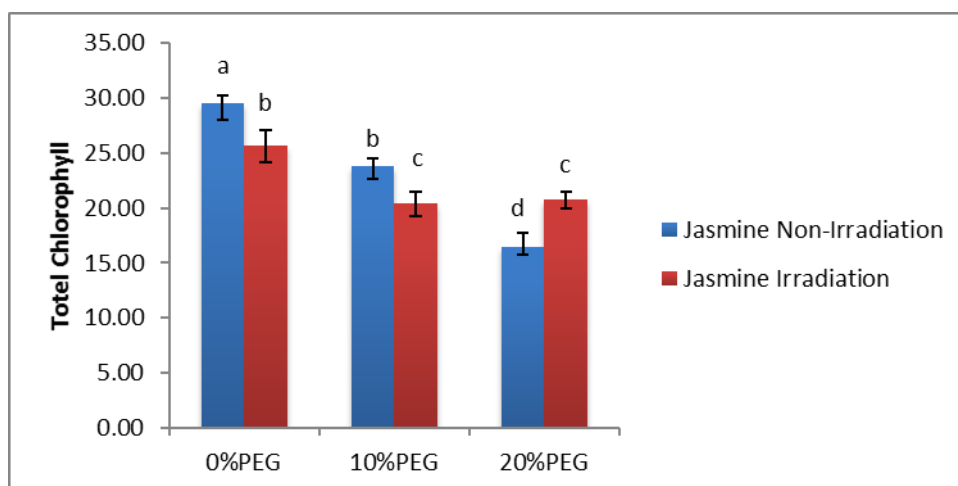


Figure 5: Overall chlorophyll composition of Jasmine plants derived from non-irradiated (0 Gy) and irradiated (350 Gy) seeds treated with 20, 10, and 0% PEG 6000.

### 3.6. Proline Concentration

As seen in Figure 6, irradiated plants experience a larger average proline concentration rise than plants that had not been irradiated. Additionally, compared to PEG 6000 concentrations of 0%, the rise in proline concentration is more substantial at 10% and 20%.

Higher proline concentrations were obtained for the irradiated plants at 0% PEG 6000 than non-irradiated plants at the same PEG 6000 concentration, thanks to the relationship between irradiation and PEG 6000 treatment. From the data, it can be inferred that irradiated plants treated with 20% PEG 6000 had higher proline concentrations than non-irradiated plants treated with the same amount of PEG 6000.

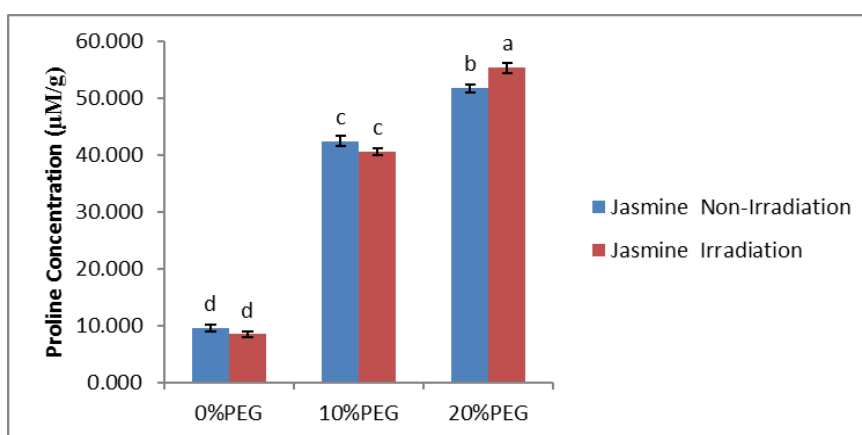


Figure 6: Average proline concentration ( $\mu\text{M/g}$ ) of Jasmine plants derived from non-irradiated (0 Gy) and irradiated (350 Gy) seeds treated with 20, 10, and 0% PEG 6000.

### 3.7. Carbohydrate Concentration

Figure 7 illustrates that the average rise in carbohydrate concentration is greater for non-irradiated plants compared to irradiated plants. Additionally, Figure 7 demonstrates that both the non-irradiated

and irradiated plants' carbohydrate concentration levels significantly decreased at PEG 6000 concentrations of 10% and 20% compared to the PEG 6000 of 0%.

Because of the relationship between irradiation and PEG 6000 concentration, non-irradiated plants treated with 0% PEG 6000 had a higher level of carbohydrate concentration than irradiated plants treated with the same amount of PEG 6000. According to the data, irradiated plants treated with 10% and 20% PEG 6000 concentrations register higher carbohydrate concentration levels than non-irradiated plants treated with the same PEG 6000 concentrations.

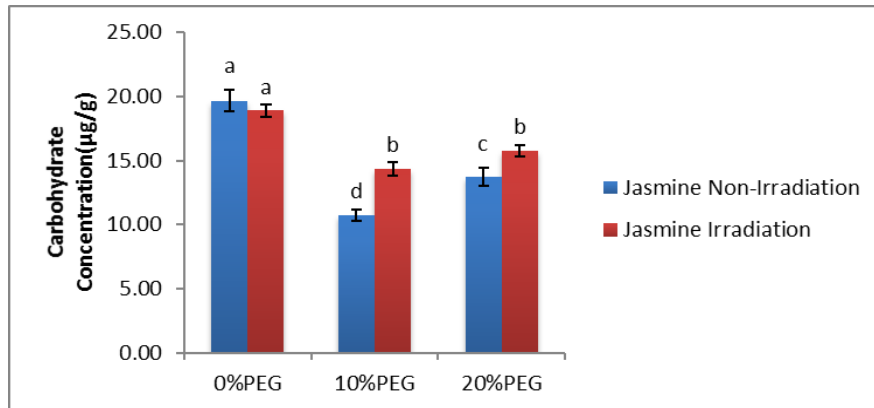


Figure 7: Average carbohydrate concentration ( $\mu\text{g/g}$ ) of Jasmine plants derived from from non-irradiated (0 Gy) and irradiated (350 Gy) seeds treated with 20, 10, and 0% PEG 6000.

## 4. Discussions

### 4.1. Shoot length, root length, fresh weight, and dry weight

Plants that grow rice are particularly vulnerable to water-induced stress (El-Okkiah et al., 2022). The crop growth stage and the time of water stress dictate how much of a rice production shortfall there will be. In contrast to the non-irradiated Jasmine plants, which delivered higher volumes of dry and fresh weights as well as root and shoot lengths at low PEG 6000 concentrations, the irradiated Jasmine plants delivered smaller volumes of fresh weights and larger proportions of dry weights along with larger volumes of root and shoot lengths at high PEG 6000 concentrations (Figures 1, 2, 3, and 4) (Sharma et al., 2022). Nearly all the genotypes that were tolerant also amassed more matter that was dry compared to the genotypes that were susceptible. The water content of the culture medium reduced when PEG 6000 is added (Kacem et al., 2017). Due to the influence of osmosis pressure on the accumulation of soluble substances within the cells, the decrease in plant fresh weight with an increase in PEG 6000 concentration can be explained (Wahab et al., 2022). The increase in PEG 6000 concentration causes a significant decrease in shoot length when compared to the control. This conclusion is consistent with the one reached by (Mahpara et al., 2022).

Numerous abiotic and biotic factors have an impact on the cultivation and growth of plants (Kumari et al., 2022). Water scarcity is one of the abiotic stress factors that is thought to be the most important. As per our observation, physiological traits or a drought tolerance response are responsible for the reduction in the length of a shoot when the PEG 6000 content rises. The results noted by (Castañeda & González, 2021), who also hypothesised that the reduction in leaf size is probably a response to a

drought environment, concur with this observation. In addition to the constrained leaf growth, they observed that the prospect of a drought also inhibits leaf gas exchange.

Irradiated Jasmine plants can continue to grow long roots while having a higher PEG 6000 concentration, as seen in Figure 2. According to (Guo et al., 2021), the increase in root length can be viewed as a reaction to a scenario brought on by drought. The process of photosynthesis is not hampered by a stop in leaf growth (Huang et al., 2018). According to (Kumari et al., 2022), the branching structure and increased biomass of roots help plants tolerate drought by allowing them to acquire water in deeper soil layers and transport it to the plant's upper portions for photosynthesis. The impacts of drought on the plant's physiological cycle prevent plant cell division. Genotypes that had not been irradiated recorded the greatest values for shoot and root length as well as dry and fresh weight at the PEG 6000 concentrations of 10 and 0%, whereas the irradiated genotypes recorded the highest values at a 20% concentration of PEG 6000. From a physiological perspective, gamma radiation has an impact because it causes the accumulation of phenolic compounds and chlorophyll pigments during the hydrolysis of water. It also modifies the anti-oxidative system, which affects the dry and fresh weight as well as root and shoot length (Merewitz et al., 2011).

#### **4.2. Total chlorophyll, proline, and carbohydrate content**

Through lipid peroxidation, nitrogen regression, and chlorophyll impairment, drought can hasten biological deterioration (Swain et al., 2014). The stomatal and mesophyll activities are inhibited by drought stress, which also hampers photosynthesis (Nadal-Sala et al., 2021). It also has an impact on the content level of chlorophyll (Mehmandar et al., 2023). The amount of chlorophyll in the leaves controls the rate of photosynthesis. The decreased chlorophyll values ascribed to stress from droughts is associated with the oxidative stress brought on by the breakdown of chlorophyll and the photooxidation of pigment (Kayoumu et al., 2023). Lack of water has a negative effect on chlorophyll levels because drought affects how plants function physiologically. According to (Seleiman et al., 2021), the accumulation of water stress decreased the water content of wheat leaves, which led to a subpar harvest. According to (Wang et al., 2018), the impacts of drought decrease the chlorophyll content of sunflower plants and lower cotton plant yields.

The non-irradiated plants seemed to have higher chlorophyll, proline, and carbohydrate contents at PEG 6000 doses of 0% and 10%. Notably, the proline, chlorophyll, and carbohydrate levels were found to be higher in the irradiated plants at a 20% PEG 6000 concentration. According to (Hasanuzzaman et al., 2020), gamma radiation plays a key part in the enhancement of genetic variability. Plant tissues and cells are stimulated by gamma rays to undergo genetic, cytological, physiological, morphogenetic, and biochemical changes (Bidabadi & Jain, 2020). It should be noted that the function of the enzyme may change if extremely high doses of gamma radiation; ranging between 2 to 20 kGy; are applied to the seed prior to sowing (Hong et al., 2022). Ionising radiation is what gamma rays essentially are, and they interact with molecules or atoms to create free activists in cells. The elimination of photosynthetic pigments can result from high radiation exposures and the ensuing weakening of in photosynthetic capability (Duarte et al., 2023). By changing the ability to photosynthesize and the activity of the antioxidant enzymes, gamma radiation can increase the capacity to withstand stress (Kiong et al., 2008). An increase in proline content prompted by gamma radiation has been shown in a research by (Kiani et al., 2022) to help boost osmotic capacity. Numerous metabolic processes that are triggered by drought stress can increase a plant's ability to withstand unfavourable climatic conditions (Oguz et al., 2022).



When plants are under water stress, molecules can build up in their tissues, improving osmotic performance and reducing tissue damage. When there is a water stress, the non-protein amino acid, proline, accumulates in plant tissues. According to (Hosseinifard et al., 2022), an increase in the concentration of proline under stressful circumstances can act as a nitrogen basin or osmoticum and a desiccation-protector. Figures 5 to 7 show the changes brought on by the accumulation of amino acids during osmotic stress. According to (Hosseinifard et al., 2022), the increased amount of proline found in leaves during salt-induced stress can be attributed to either protein synthesis or stress-triggered protein degradation. The accumulation of solutes in rice plants under drought stress increases their capacity to withstand drought, according to (Shim et al., 2023). An increase in amino acid content improves drought resistance by preserving optimum water conditions and guaranteeing the availability of amino acids for the synthesis of proteins during a drought. Following a treatment with a PEG 6000 concentration of 10%, a sizable decrease in the concentration of carbohydrate was seen (Figure 7). To increase stress tolerance, the cells of a plant rely heavily on carbohydrates for energy (Jeandet et al., 2022). It was found that the rise in carbohydrate content is more prominent for the irradiated plants compared to the non-irradiated plants at PEG 6000 concentrations of 10% and 20%. This increase in carbohydrate concentration can be attributed to many sugars dissolving at a time of water constraint. As noted by (Guo et al., 2021), drought situations cause the intensity of carbohydrate concentration to increase. These observations are in line with that of (Kayoumu et al., 2023).

## 5. Conclusion

The findings of this present study indicate that the genotypes that are non-irradiated had the highest values at a concentration of PEG 6000 that was 0%, whereas the irradiated genotypes rendered the highest root and shoot lengths, chlorophyll and proline contents, dry and fresh weights along with the highest carbohydrate concentration level at PEG 6000 concentrations of 10% and 20%. Thus, it is clear that using radiation increases plants' ability to withstand drought circumstances.

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