



RESEARCH ARTICLE

## Exogenous melatonin on Physiological and Yield Traits of Cassava (*Manihot esculenta* Crantz) under Salt Stress

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### ABSTRACT

The present investigation to evaluate the impact of melatonin on physiological, biochemical characters and yield potential of cassava under salt stress condition. The present study was carried out in cassava variety Sree Athulya with nine treatments under 120mM NaCl salt stress condition. Different treatments viz., sett treatment and foliar application of 100 ppm melatonin was done at 30 and 60 DAP of the crop growth. Control (salt stress + no melatonin) and absolute control (no stress and melatonin) also maintained for comparison purpose. The salt stress of 120 mM NaCl was imposed from day one to 120 days. Observations done on 45, 75 and 135 days after planting revealed that foliar spray of 100 ppm melatonin at 30 days after planting recorded percent increase of 33.56 in photosynthetic rate, 37.28 in stomatal conductance, 13.60 in transpiration rate and sett treatment plus foliar spray at 30 and 60 days after planting showed maximum osmotic adjustment, osmotic potential, proline (16.54 %) and soluble protein content (10.32 %). The melatonin treated plants are efficient in producing higher yield than untreated one under salt stress.

**Keywords:** Cassava, melatonin, salt stress

### INTRODUCTION

Cassava (*Manihot esculenta* Crantz) is moderately sensitive to salt stress. The varying climatic conditions like heat stress and drought causes depletion of ground water and concentration of salt in irrigation water is getting increased. During irrigation, salt deposition on the soil causes drying of leaves, reduced tuber yield and quality. It was efficient that the use of melatonin, an anti-stress compound plays a vital role in plant stress defense mechanism related to drought and salt stress. Salinity is one of the major abiotic factors limiting the crop yield and threatening food security worldwide (Sah *et al.* 2016). Salt stress to plants leads to reduced plant growth and productivity. Several factors such as unsustainable irrigation practices and deforestation causes an increase in the area of salt stress (Munns and Gilliam, 2015). Worldwide about 800 million hectares of land are affected by salt, and this accounts for about 6 % of the total land area (FAO, 2008). During irrigation, salt deposition in the soil resulting in the unproductive condition. Salinization of groundwater is becoming an increasing problem in many parts of the cassava growing areas.

The semi-arid and arid zones have the natural

cause of accumulation of salts. The soluble salts such as chlorides, sulfates, carbonates are released due to the weathering of parental rocks. The abundant cause is the release of sodium chloride and accumulation of oceanic salt by wind and rain. It was reported that 6-50 mg/kg of sodium chloride is present in rainwater. When the soils contain more than 40 mM of NaCl (EC 4 ds/m or more), it is said to be salt-affected soil (Munns and Tester, 2008). The pH of saline soils is lower than the sodic soils (less than 8.5) and have exchangeable sodium-potassium ratio of lesser than 15 (IRRI, 2011). Under salt stress, plants are prone to the production of excessive reactive oxygen species (ROS), membrane lipids or proteins peroxidation occurs, which destroys the cell membranes leading to cell death of a normal plant. Salt at high concentration causes osmotic stress with reduced water potential in plant roots. Also the uptake of water and nutrients gets affected that inhibits the growth and development of plant that results in wilting and death of plants (Liu *et al.*, 2018). Due to the rise in sea level and the groundwater getting contaminated, there is a need for the development of salt-tolerant crops.

Cassava (*Manihot esculenta* Crantz) is the most widely cultivated tuber crop in the tropics as a food crop due to the high starch content of the

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roots. It has its own inherent tolerance to stressful environment, it is therefore, considered to abiotic stress-tolerant (Bull *et al.*, 2011). According to FAO, cassava is said to be moderately sensitive to salt stress (Gleadow *et al.*, 2016). Cassava cultivation is likely expanded in salt-affected soil zones to promote agriculture in unproductive lands (Carretero *et al.*, 2007) due to increased demand for cassava production (Shabala *et al.*, 2013). Cassava being the important staple crop among the tropics, has its tolerance to drought and high temperatures but its response to salt tolerance is unknown. The successful development of crops to survive under salt stress has been a long-time concern (Munns 2002). Plant growth regulators are extensively used to regulate plant growth and to enhance plant stress tolerance. Melatonin is a pleiotropic molecule and has many cellular and physiological functions in varied kingdoms (Arnao and Hernandez-Ruiz., 2015) present in plants and animals (Dubbels *et al.* 1995; Reiter *et al.* 2011; Shi *et al.* 2016). Melatonin was found to be involved in the regulation of plant growth and development, which protects plants against abiotic and biotic stresses such as salt, drought, cold, heat and heavy metal stresses (Reiter *et al.*, 2015). It was reported that the application of exogenous melatonin effectively improves salt tolerance in certain plants. In *Malus hupehensis*, the pretreatment with melatonin reduced the inhibitory effects of salt stress, such as degradation and loss of chlorophyll (Li *et al.*, 2012). Similarly, treatment with melatonin reverts this inhibition of growth such as declining net photosynthetic rate and chlorophyll content in drought and high salt stress conditions (Zhang *et al.*, 2014). Under salt stress, the application of 50–150  $\mu\text{M}$  melatonin increased the total chlorophyll content and enhanced photosynthetic capacity in cucumber (Wang *et al.*, 2016). In apple and tomato, the melatonin pretreatment increased photosynthetic efficiency (Li *et al.*, 2012; Yin *et al.*, 2019). According to Ren *et al.* (2020) maize seedlings treated with melatonin showed higher leaf area and photosynthetic activity under salt stress. Similarly, the maize seedlings showed higher transpiration rate and photosynthetic rate under drought (Qiao *et al.*, 2020; Huang *et al.*, 2019). Hwang *et al.* (2020) proved that the antioxidants present in melatonin enhanced photosynthesis. Thus exogenous melatonin exhibits a major role in ROS reduction and increases antioxidants and secondary metabolites (Shakhawat *et al.*, 2020; Shakeel *et al.*, 2020). Altaf *et al.* (2020), in tomato seedlings under salt stress, discovered the increase in gas exchange parameters when treated with melatonin. Ren *et al.* (2020) suggested that melatonin-treated plants had higher osmolyte contents, so the osmotic potential was lower compared to control. Organic osmolytes such as soluble sugars maintain osmotic adjustment

and further sucrose and fructose levels enhance melatonin-treated plants under salt stress. The present study was proposed to evaluate melatonin on physiological role related to tuber yield under salt stress in cassava.

## **MATERIAL AND METHODS**

### **Plant materials**

The cassava variety Sree Athulya, a central variety released by Central Tuber Crop Research Institute, Trivandrum was used for the study.

### **Treatments**

The study was carried out during 2019 at the Department of Crop Physiology, Tamil Nadu Agricultural University, Coimbatore. Salt stress was imposed with 120mM NaCl once in three days. from day one of planting to 120 days after planting for all the treatments except absolute control as per the method of Kalarani *et al.* (2018). The deposited salt were flushed out once in a week with normal water. The plants of absolute control were maintained with normal irrigation and without melatonin. Various treatments *viz.*, absolute Control (without salt and melatonin), control (salt+ no melatonin), sett treatment with 100 ppm melatonin, foliar spray of 100 ppm melatonin at 30 DAP, foliar spray of 100 ppm melatonin at 60 DAP, foliar spray of 100 ppm melatonin at 30 and 60 DAP, sett treatment of 100 ppm melatonin + foliar spray of 100 ppm melatonin at 30 DAP, sett treatment of 100 ppm melatonin + foliar spray of 100 ppm melatonin at 60 DAP, sett treatment of 100 ppm melatonin + foliar spray of 100 ppm melatonin at 30 and 60 DAP. The following parameters were analyzed.

### **Growth measurements**

Observations on physiological parameters were taken on 45 days, 75 days and 135 days after planting. Three replicates were taken for each treatment and the mean calculation was done for each measurement. The physiological parameters *viz.*, gas exchange parameters (photosynthetic rate, transpiration rate and stomatal conductance), osmotic potential and osmotic adjustment were taken. Tuber yield was taken during harvest.

### **Leaf gas exchange parameters**

Gas exchange parameters *viz.*, photosynthetic rate, transpiration rate and stomatal conductance were recorded using an advanced portable photosynthesis system (LI-6400 XT, LicorInc, Nebraska, USA). The readings were recorded from 10.00 am to 12.00 noon on a clear sunny day when the photosynthetically active radiation was more than 1000  $\mu\text{mol photons m}^{-2}\text{s}^{-1}$  also which avoids effects of photo-inhibition. A fully expanded leaf from the top was clamped inside the leaf chamber and

held perpendicular to incident light and computed values were recorded. The instrument maintained a constant CO<sub>2</sub> flux to leaf chamber, which was maintained at ambient concentration. Relative humidity was maintained at a steady level equal to the ambient relative humidity to simulate a condition similar to ambient air. The photosynthetic rate expressed as μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>, stomatal conductance expressed as mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup> and transpiration rate expressed as mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>.

#### **Osmotic potential and osmotic adjustment**

The penultimate fully expanded leaf on the main stem was cut, wrapped in a plastic bag and soaked in water in the refrigerator for 24 hours to rehydrate the tissue. The rehydrated leaf was placed in aluminium foil, frozen with liquid nitrogen for 30 seconds to stop the physiological function of its cells and stored in a -80 °C freezer. The sap was collected by squeezing the leaf sample with a sterile syringe and the osmolality (mmol kg<sup>-1</sup>) of the expressed sap was determined using a vapor pressure osmometer (Vapro Model 5520 Wescor Inc., Logan, UT, USA). Osmotic potential (ψ<sub>s</sub>) was calculated as ψ<sub>π</sub> = -c RT, where c is concentration, R is the universal gas constant (0.0832) and T is the temperature in degrees Kelvin (310° K). The following conversion equation was used to compute osmotic potential (inMPa).

$$\text{Osmotic potential} = \frac{[(\text{Osmolality mmol kg}^{-1}) (0.0832) (310)]}{10000}$$

Osmotic adjustment was calculated as the difference between the turgid potential in normal watered treatment and stress treatment (Babu *et al.*, 1999).

#### **Proline content**

The proline content was estimated by acid ninhydrin protocol given by Bates *et al.* (1973) and expressed in mg g<sup>-1</sup>. The leaf sample (0.5g) was homogenized with 10 ml of 3 per cent sulphosalicylic acid and centrifuged at 3000 rpm for 10 minutes. Two ml of the supernatant was taken and 2 ml of glacial acetic acid, 2 ml of orthophosphoric acid and 2 ml of acid ninhydrin mixture were added. The contents were allowed to react at 100 °C for 1 hour and then it is incubated on ice for 10 minutes to terminate the reaction. The reaction mixture was mixed vigorously with 4 ml toluene for 15 to 20 seconds in separating funnel. The chromophore containing toluene aspired from the aqueous phase, warmed to room temperature and optical density was read at 520 nm.

#### **Soluble protein content**

Soluble protein content in the leaf was estimated at 660 nm by using Folin Ciocalteu reagent by following the procedure described by Lowry *et al.* (1950). 250 mg of leaf sample was macerated with 10 ml of phosphate buffer and the content was centrifuged at 3000 rpm for about 10 minutes. The supernatant was collected and made up to 25 ml. One ml of the supernatant and 5 ml of alkaline copper tartarate reagent were mixed with 0.5 ml of folin ciocalteu reagent and the OD value was measured at 660 nm in the spectrophotometer. The soluble protein content was expressed as mg g<sup>-1</sup> fresh weight by using bovine serum albumin as the standard.

#### **Tuber yield per plant**

The weight of all marketable tubers per plant was recorded in each replication was added and an average yield per plant was worked out and expressed in kg per plant.

#### **Statistical analysis**

The data collected from this experiment on various parameters were statistically analyzed in Completely Randomized Design (CRD) as suggested by Gomez and Gomez (1992) with three replicates. The treatment differences were analyzed using DMRT. The critical difference (CD) was computed at five per cent probability and were furnished and standard error was calculated.

## **RESULTS AND DISCUSSION**

### **Physiological characters**

#### **Gas exchange parameters**

Irrespective of the treatments, photosynthetic rate was increased from 45 DAP to 135 DAP. The photosynthetic rate decreased under stress condition. The rate of photosynthesis shows a significant difference between control and other treatments. The results showed that the salt-treated plants have a declined net photosynthetic rate in all three stages (13.59, 15.19 and 16.13 μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>). Among the treatments, sett treatment plus foliar spray of melatonin at 30 DAP and 60 DAP was recorded the highest photosynthetic rate under salt stress conditions 21.81, 19.08 and 18.76 μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup> at 45, 75 and 135 DAP (Table. 1) followed by foliar application of melatonin only at 30 DAP. Photosynthesis, physico-chemical process which by utilizing light energy forms organic compounds for plant growth, development and production (Barnawal *et al.*, 2017). Melatonin preserves chlorophyll and improves the efficiency of photosynthesis under stress conditions (Jiang *et al.*, 2016; Li *et al.*, 2018). According to Zhang *et al.* (2014) melatonin protects chlorophyll and delays leaf senescence, furthers

maintains photosynthetic rates. These findings support the present investigation.

Similar to photosynthetic efficiency, the transpiration rate and stomatal conductance show a significant difference. Initially, under salt stress conditions there was a high transpiration rate in

control than all other treatments. Later transpiration rate increased in melatonin-treated plants. Under salt stress conditions, maximum rate of transpiration and stomatal conductance was observed in foliar spray of 100 ppm melatonin at 30 DAP in all the three stages which is on par with sett treatment plus

foliar application of 100 ppm melatonin at 30 and 60 DAP (Table. 2). Salt plus melatonin treatment showed higher stomatal conductance than plants in salt alone, but it was less compared to absolute control plants affected by salt stress with melatonin.

**Table 1 . Effect of melatonin on Photosynthetic rate ( $\mu\text{mol CO}_2 \text{ m}^{-2}\text{s}^{-1}$ ), transpiration rate ( $\text{mmol H}_2\text{O m}^{-2}\text{s}^{-1}$ ) of cassava under salt stress condition**

| Treatments |  | Photosynthetic rate<br>( $\mu\text{mol CO}_2 \text{ m}^{-2}\text{s}^{-1}$ ) |               |               |              | Transpiration rate<br>( $\text{mmol H}_2\text{O m}^{-2}\text{s}^{-1}$ ) |               |               |             |
|------------|--|---|---------------|---------------|--------------|---|---------------|---------------|-------------|
|            |  | 45 DAP  | 75 DAP        | 135 DAP       | Mean         | 45 DAP  | 75 DAP        | 135 DAP       | Mean        |
| T1         | Absolute Control (without salt and melatonin)                                      | 23.08   | 25.14         | 27.03         | 25.08        | 8.10  | 8.74          | 9.06          | 8.63        |
| T2         | Control (with salt)  | 13.59   | 15.19         | 16.13         | 14.27        | 9.14  | 6.74          | 6.03          | 7.30        |
| T3         | Sett treatment with 100 ppm melatonin  | 14.76   | 16.83         | 17.97         | 16.52        | 7.30  | 7.40          | 7.88          | 7.52        |
| T4         | Foliar spray of 100 ppm melatonin at 30 days after planting                        | 19.20   | 21.74         | 23.51         | 21.48        | 8.03  | 8.35          | 8.97          | 8.45        |
| T5         | Foliar spray of 100 ppm melatonin at 60 days after planting                        | 14.18   | 14.87         | 14.35         | 14.80        | 7.52  | 7.63          | 7.91          | 7.68        |
| T6         | Foliar spray of 100 ppm melatonin at 30 and at 60 days after planting              | 16.81   | 14.01         | 13.83         | 14.88        | 7.72  | 7.97          | 8.46          | 8.05        |
| T7         | Sett treatment+ Foliar spray of 100 ppm melatonin at 30 days after planting        | 17.94   | 15.28         | 15.02         | 16.08        | 7.68  | 7.93          | 8.42          | 8.01        |
| T8         | Sett treatment+ Foliar spray of 100 ppm melatonin at 60 days after planting        | 16.33   | 16.17         | 15.81         | 16.10        | 7.37  | 7.52          | 7.97          | 7.62        |
| T9         | Sett treatment+ Foliar spray of 100 ppm melatonin at 30 and 60 days after planting | 21.81   | 19.08         | 18.76         | 19.88        | 7.94  | 8.22          | 8.71          | 8.29        |
|            | <b>Mean</b>  | <b>17.52</b>  | <b>17.59</b>  | <b>18.27</b>  | <b>17.75</b> | <b>7.87</b>   | <b>7.83</b>   | <b>8.16</b>   | <b>7.95</b> |
|            | <b>SEd</b>   | <b>0.46</b>   | <b>0.47</b>   | <b>0.49</b>   |              | <b>0.20</b>   | <b>0.20</b>   | <b>0.21</b>   |             |
|            | <b>CD (0.05)</b>   | <b>0.96**</b>   | <b>0.98**</b> | <b>1.03**</b> |              | <b>0.43**</b>   | <b>0.44**</b> | <b>0.45**</b> |             |

NS- Non significant \*- Significant \*\*- Highly significant

45 DAP: 15 days after 1st spray, 75 DAP: 15 days after 2nd spray, 135 DAP: Recovery stage

It was concluded that stomatal conductance holds larger stomatal opening (Zhang *et al.*, 2020). Chen *et al.* (2018) reported that the exogenous melatonin mitigates from salt stress and decreases the reduction in stomatal conductance in maize. Zhang *et al.* (2019) observed that water deficit stress on photosynthesis could be reduced by melatonin in soybean leaves which enhances stomatal conductance, transpiration rate and maintains normal photosynthetic rate for normal growth and development. Wang *et al.* (2013) and Weeda *et al.* (2014) reported that melatonin activates CAB gene, which is associated in chlorophyll biosynthesis and reduced PAO gene, which degrades chlorophyll. This might be the reason that melatonin increases the photosynthetic rate. The mechanism for an increase in transpiration and assimilation rate might be the down-regulation of ABA synthetic gene and up-regulation of ABA catabolic genes by melatonin.

As a result ABA level gets reduced in stress-induced plant and the stomata remains open Hasan *et al.* (2015). The earlier findings collaborated well with the present study.

#### **Osmotic potential (-Mpa) and osmotic adjustment (Mpa)**

The osmotic potential and osmotic adjustment were observed in control plants and stressed plants. The relationship between osmotic potential and osmotic adjustment is inverse under stress conditions. It was observed that data on osmotic potential and osmotic adjustment shows the effect of melatonin compared to control (Figure. 1). Sett treatment plus foliar application of 100 ppm melatonin at 30 DAP and 60 DAP indicates more osmotic adjustment and less osmotic potential in all the three stages 0.85 (Mpa) and -2.06 (-Mpa) at 45 DAP, 0.94 (Mpa) and -1.75 (-Mpa) at 75 DAP, 0.88

(Mpa) and -1.47 (-Mpa ) at 135 DAP respectively followed by melatonin spray only at 30 DAP. Absolute control shows the maximum reduced potential -1.57 (-Mpa), -0.81 (-Mpa ) and -0.59 (-Mpa ) at 35 DAP,

75 DAP and 135 DAP respectively. The effective strategy for plants to resist salt-stimulated osmotic stress is an osmotic adjustment (Yin et al., 2013). Chen et al. (2014) suggested a decrease in osmotic

potential and an increase in osmotic adjustment has been observed in melatonin-treated plants under salt stress conditions. Under salt and drought stress conditions, the effect of melatonin in maintaining water status was reported by Chen et al. (2018) and Su et al. (2019). These earlier findings confirm the present study.

**Table 2. Effect of melatonin on stomatal conductance ( $\text{mmol H}_2\text{O m}^{-2}\text{s}^{-1}$ ) of cassava under salt stress condition**

| Treatments  | Stomatal conductance ( $\text{mmol H}_2\text{O m}^{-2}\text{s}^{-1}$ ) |               |               |             |
|---|--|---------------|---------------|-------------|
|   | 45 DAP   | 75 DAP        | 135 DAP       | Mean        |
| T1 Absolute Control (without salt and melatonin)                                      | 0.48   | 0.82          | 0.94          | 0.74        |
| T2 Control (with salt)  | 0.32   | 0.36          | 0.45          | 0.37        |
| T3 Sett treatment with 100 ppm melatonin  | 0.31   | 0.34          | 0.50          | 0.38        |
| T4 Foliar spray of 100 ppm melatonin at 30 days after planting                        | 0.42   | 0.53          | 0.84          | 0.59        |
| T5 Foliar spray of 100 ppm melatonin at 60 days after planting                        | 0.31   | 0.32          | 0.52          | 0.38        |
| T6 Foliar spray of 100 ppm melatonin at 30 and at 60 days after planting              | 0.35   | 0.40          | 0.64          | 0.46        |
| T7 Sett treatment+ Foliar spray of 100 ppm melatonin at 30 days after planting        | 0.36   | 0.41          | 0.62          | 0.46        |
| T8 Sett treatment+ Foliar spray of 100 ppm melatonin at 60 days after planting        | 0.33   | 0.38          | 0.56          | 0.42        |
| T9 Sett treatment+ Foliar spray of 100 ppm melatonin at 30 and 60 days after planting | 0.38   | 0.44          | 0.73          | 0.51        |
| <b>Mean</b>   | <b>0.36</b>  | <b>0.44</b>   | <b>0.64</b>   | <b>0.48</b> |
| <b>SEd</b>  | <b>0.18</b>  | <b>0.19</b>   | <b>0.20</b>   |             |
| <b>CD (0.05)</b>  | <b>0.38**</b>  | <b>0.40**</b> | <b>0.43**</b> |             |

NS- Non significant \*- Significant \*\*- Highly significant

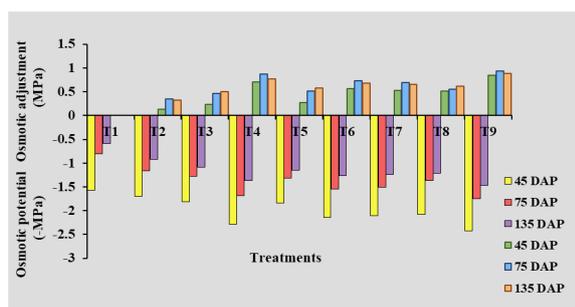
45 DAP: 15 days after 1st spray, 75 DAP: 15 days after 2nd spray, 135 DAP: Recovery stage

## BIOCHEMICAL PARAMETERS

### Proline content

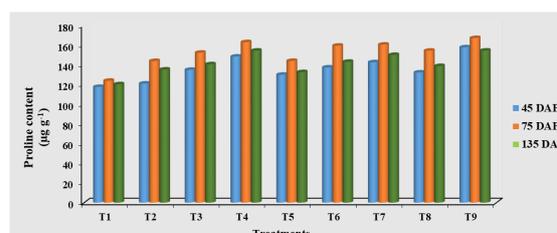
Proline content showed a significant increase under salt stress (Figure. 2). Among the melatonin treatments, sett treatment plus foliar spray of 100 ppm melatonin had maximum proline content of 157.62, 167.01 and 154.31  $\mu\text{g g}^{-1}$  at 45, 75 and 135 DAP respectively, followed by foliar spray alone at 30 DAP (148.23, 162.84 and 154.27  $\mu\text{g g}^{-1}$ ). 16.54 per cent increase in proline content was observed in the best melatonin-treated plants than the salt-stressed plant.

**Figure 1. Effect of melatonin on osmotic potential (-Mpa) and osmotic adjustment (Mpa) of cassava under salt stress condition**



Proline is an amino acid that has an adaptive role in osmotic adjustment (Ashraf and Harris 2004). Melatonin under abiotic stress up-regulates the expression of proline synthesis gene *P5CS1* and down-regulates *PDH1* gene (Aghdam et al., 2019). Proline is an important osmoprotectant in plants, also functions as chaperons involved in protecting protein integrity. Mansour and Ali. (2017) explained the role of proline in quenching singlet oxygen. Godoy et al. (2021) reported the mechanism of proline in abiotic stress tolerance.

**Figure 2. Effect of melatonin on proline content ( $\mu\text{g g}^{-1}$ ) of cassava under salt stress condition**



The salt-induced tomato seedling maintained enhanced proline content when treated with melatonin (Manzer et al., 2019). Similar results of

increased proline content was observed in pistachio leaves treated with melatonin under salt stress (Kamiab, 2020). These earlier findings support the present investigation.

**Table 3. Effect of melatonin on Tuber yield plant<sup>-1</sup> of cassava under salt stress condition**

| Treatments  | Tuber yield (kg plant <sup>-1</sup> ) |
|---|---------------------------------------|
| T1 Absolute Control (without salt and melatonin)                                      | 5.21                                  |
| T2 Control (with salt)  | 2.13                                  |
| T3 Sett treatment with 100 ppm melatonin  | 2.56                                  |
| T4 Foliar spray of 100 ppm melatonin at 30 days after planting                        | 4.01                                  |
| T5 Foliar spray of 100 ppm melatonin at 60 days after planting                        | 2.48                                  |
| T6 Foliar spray of 100 ppm melatonin at 30 and at 60 days after planting              | 3.05                                  |
| T7 Sett treatment+ Foliar spray of 100 ppm melatonin at 30 days after planting        | 3.12                                  |
| T8 Sett treatment+ Foliar spray of 100 ppm melatonin at 60 days after planting        | 2.73                                  |
| T9 Sett treatment+ Foliar spray of 100 ppm melatonin at 30 and 60 days after planting | 3.95                                  |
| <b>Mean</b>   | <b>3.25</b>                           |
| <b>SEd</b>  | <b>0.08</b>                           |
| <b>CD (0.05)</b>  | <b>0.17**</b>                         |

NS- Non significant \*- Significant \*\*- Highly significant

### Soluble protein content

Soluble protein content has been observed in absolute control plants and salt-stressed plants (Figure. 3). A significant increase of soluble protein content was reported in melatonin-treated cassava plants under salt stress. Leaf soluble protein assessment imparts the photosynthesis efficiency and production of assimilates. Soluble protein content declined in salt stress condition. Results indicate that the application of melatonin as sett treatment plus foliar spray at 30 and 60 DAP increased the soluble protein content of 13.10, 14.64 and 15.87 mg g<sup>-1</sup> of fresh weight followed by foliar spray of melatonin at 30 DAP alone (12.03, 13.74 and 14.07 mg g<sup>-1</sup>) at 45, 75 and 135 DAP respectively. An increase per cent of 33.98 was observed in the best melatonin treatment over the salt-stressed plants. In line with our results the pretreated tomato seeds exposed to salt stress condition had enhanced soluble protein content (Altaf *et al.*, 2020). Similarly, Yin *et al.* (2019) reported increased leaf soluble protein content on exogenous application of melatonin at low concentration.

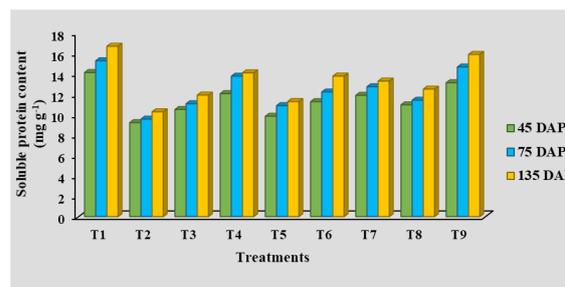
### Yield trait

#### Tuber yield (kg/plant)

The data on yield potential is presented in (Table. 3). The application of melatonin has an improved

effect on the tuber yield of plants compared to the control plants under salt stress condition. Apart from absolute control (5.21 kg plant<sup>-1</sup>), foliar spray of 100 ppm melatonin at 30 DAP (4.01 kg plant<sup>-1</sup>) recorded maximum yield (3.95 kg plant<sup>-1</sup>), which is on par with sett treatment plus foliar spray of 100 ppm melatonin at 30 DAP and 60 DAP. Among the various treatments, stressed plant (control) recorded the least yield (2.13 kg plant<sup>-1</sup>).

**Figure 3. Effect of melatonin on soluble protein content (mg g<sup>-1</sup>) of cassava under salt stress condition**



Beyon and Back (2014) explained that melatonin might alter plant characters such as seedling growth, senescence and yield. The increase in yield and nutritional quality of crops has been observed in plants that are introduced with melatonin biosynthetic genes (Nawaz *et al.*, 2016). Similarly, foliar application 100 mM melatonin in moringa under drought was known to increase growth rate, yield and yield components (Sadak *et al.*, 2020). Pre-soaking treatment of wheat seed in 100 μM improved grain yield and other yield components (Ye *et al.*, 2020). When exogenous melatonin was treated with soybean seeds, the higher grain yield *viz.*, number of pods, number of grains per pod and total pod weight were observed (Zhang *et al.*, 2019). Earlier studies of moringa, wheat, and soybean confirmed the present investigation.

### CONCLUSION

The application of 100 ppm melatonin as sett treatment combined with foliar spray at 30 and 60 DAP resulted in higher photosynthetic rate, stomatal conductance, transpiration rate, osmotic potential, osmotic adjustment, proline and soluble protein content of cassava plants under salt stress. The findings of this study suggest the positive effect of melatonin in alleviating salt stress. Hence the exogenous melatonin application in cassava exhibited a better performance in physiological and biochemical traits associated with improved yield potential.

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