

Identification of drought induced drastic effects on Guava (*Psidium guajava*) at seedling stage

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Abstract

The present field trial was conducted to investigate the growth performance of seedlings of guava (*Psidium guajava*) variety surahidar to investigate different drought levels at early seedling growth stage of plant development. The experiment was conducted in the field, Department of Horticulture, University College of Agriculture, University of Sargodha, Sargodha under eight treatments (Control after 2 days, 4, 6, 7, 8, 9 and 10 days) water stress to evaluate their consequence on plant growth. Data of morphological (seedling fresh and dry weight, number of leaves, leaf area, and seedling shoot/root length), physiological (photosynthesis rate, transpiration rate, water use efficiency and stomatal conductance,) and biochemical (chlorophyll contents and proline contents) parameters were recorded after 60 days of treatment applications. In case of all parameters control treatment produced maximum results, while 10 days irrigation produced minimum results. Moisture-sensitive seedlings were distinctly affected by lowest water stress and concluded the seedlings as most susceptible to water immediately after transplanting. Seedlings planted under suboptimal drought levels were coupled with slow development. Under drought stress conditions the irrigation pressure within the leaves of plants was reduced and seedlings showed wilting. The major effect of water scarcity was reduced development and growth caused by reduced photosynthesis.

Key words: Drought; Guava; Seedling; Morphological; Physiological; Biochemical

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1. Introduction

In the fruit industry of Pakistan, guava has a prominent position among fruits. It belongs to a Myrtaceae family, with about 133 genera plus more than 3,800 species along with it is among the most gregarious of fruit trees [1]. It is extensively cultivated all around the tropical and subtropical areas however guava is originated from the tropical America although in this sub-continent; guava has been in grown as early on 17th century [2]. It has been dispersed by birds, man and other animals to each or all humid areas associated with hot America along with the West Indies. Guava with a large seedy core is a berry fruit. The fruit is waxy and smooth. Guava tree carries a superficial root system with scattering branches. The elevation of tree is mostly 4-5 meters nevertheless big trees may attain a peak of 9 meters. It grows wild as bushes in the native areas of tropical America which include Mexico, Peru as well as Cuba. It is reported that guava is most significant fruit in production and area soon after mango, lemon and also

jackfruit [3]. It is cultivated in the actual farmhouse gardens through the entire state still with no or through tiny care. It has 4th position on such basis as area (63 thousands of ha) along with output (555000 tons). Involving various provinces, Punjab has contributed the main share in guava productivity in Pakistan by means of 49000 ha area and 445000.5 tons yield [4]. Guava is among the gorgeous fruit in shape, appearance, nutrition and also aroma. Guava possesses exceptional nutritional importance, medicinal properties and flavor and has a immense potential of processing into precious products. Guavas of red in colour may be used as the base of brackish products such as constituting a substitute for tomatoes, sauces, specifically for those responsive for the last tartness. Particular fruit is sweetie and used as raw or cooked. It is used to make good jam and jellies. It is often a prosperous source of vitamin C as compared to ber, citrus and also apple [5]. The most of the species marketed as 'marvellous fruit', containing vitamin A, magnesium, potassium, 4 times more vitamin C than orange (200 mg 100g⁻¹), and essential nutrients with

low calories. It has been reported that guava fruit contain soluble solid contents (SSC) ranging from 9.60-11.14%, ascorbic acid from 167.50 - 210.00 mg 100 g⁻¹, titratable acid from 0.28 - 0.38%, reducing sugars from 5.04 - 5.49%, total sugars from 7.93 - 8.90% and also acetic acid from 55.40 - 122.13 μmol kg⁻¹ [6].

Fruits contain moisture (85%), carbohydrates (11%) and protein (7%) (Samson, 1986). Guava fruits are used to make guava cheese, guava paste as well as guava jelly that's approximately marketed across the world [7]. It is additionally prepared into fruit leather and syrup concerning use on waffles, milkshakes, puddings as well as in ice cream [7]. Guava nectar as well as juice are one of many plentiful admired bottled or canned fruit beverages from the Caribbean area. Also, guavas be combined to make breakfast-food flakes with other ingredients and cornmeal [7]. Most of the guava plants in United States are cultivated only in a few favorable locations of California, Florida, and Hawaii. In recent years, the American industry demand for unique fruits, like guava, have been increasing, mainly due to increased immigration through Asia, Latin America, and other warm countries [8]. That has a long harvest time period, guava can be a potential, alternative high-value cash crop within the U.S. Throughout tropical countries, the roots, bark, leaves, and green fruit are widely-used in medicine regarding gastroenteritis, diarrhea, as well as dysentery. The guava plant is cultivated in numerous countries all around the world, including India, Brazil, South Africa, Venezuela, Cuba, the Philippines and New Zealand. It is essential in international trade along with the domestic economy in excess of 50 tropical as well as subtropical countries [9]. The present research project was planned for exploring the drought induced drastic effects on Guava at seedling stage.

2. Material and Methods

The present study was laid out at Horticultural nursery area, University College of Agriculture, University of Sargodha, Sargodha, Pakistan during 2014-2015. The experiment was conducted for identification of drought induced drastic effects on guava at seedling stage. Young seedlings of guava variety Surahidar were collected from pattuki Lahore and were transplanted in medium size pots (14 inch diameter), filled with the field soil containing sand, silt and clay with the ratio 1:1:3 as growth medium. The sand, silt and clay was well mixed with each other. One seedling per pot was planted and pots were kept in open field conditions in nursery area in Department of Horticulture. The plants were watered according to the need of plant. The seedlings were allowed to grown under normal field conditions for 30 days. Then seedlings were transferred in the new soil. Less vigorous and diseased seedlings were replaced by the healthy ones. After 30 days of transplanting seedlings were subjected to 7 different drought levels, control, after 2 days, 4,6,7,8,9 and 10 days seedlings were irrigated. The data pertaining to various growth, physio-biochemical parameters was collected after 60 days of treatment application. Different treatments T₀- control (Watering after 2 days (1000ml/pot), T₁- after 4 days (1000ml/pot), T₂- after 6 days (1000ml/pot), T₃- after 7 days (1000ml/pot), T₄- after 8 days (1000ml/pot), T₅- after 9 days (1000ml/pot) and T₆- after 10 days (1000ml/pot) were used.

Data of different morphological parameters; shoot length (cm), root length (cm), leaves/plant, leaf area (measured in cm² with digital leaf area meter), fresh shoot weight (g), dry shoot weight (g), fresh root weight (g) and dry root weight (g). Physiological parameters; photosynthesis rate (μmol CO₂ dm⁻² S⁻¹), transpiration rate (mmol H₂O m⁻² S⁻¹), water use efficiency and stomatal conductance (mmS⁻¹) were calculated by Infra-red gas analyzer (IRGA) CI-340 Photosynthesis system. Biochemical parameter; total chlorophyll contents (mg g⁻¹) and leaf proline were determined.

Seven treatments were used and each treatment was replicated six times. Guava seedlings were grown in pots. Standard method was followed for verification of data. In described research we follow CRD Design. Data was analyzed using ANOVA under CRD in statistic 8.1 software [10].

3. Results and Discussion

3.1. Number of leaves

Leaves are known as food factory of plants. Plants contain a green pigment called chlorophyll in their mesophyll cells of leaves, which plays an essential role in carbohydrate production. The results of the experiment have significant differences among all treatments shown in Figure 1. Number of leaves increased along with irrigation water and declined gradually with decreasing amounts of irrigation water applied. With the lowest irrigation water applications among all treatments (9 and 10 days interval) number of leaves decline became most pronounced (5.667, 5.000) respectively. In contrast, the control (watering after 2 days interval) recorded a higher average number of leaves (13.167). These results are in harmony with those obtained by Hassan (1998) and Gowda (1998), which revealed that number of leaves decreased by increasing the level of water stress [11, 12]. Less number of leaves per plant with the drier treatments was also documented by Horton *et al.* (1982) and Abo-Taleb *et al.* (1998) [13,14].

3.2. Leaf Area (cm²)

Regarding leaf area data in Figure 2 showed significant difference among the treatments and results showed that the average leaf area was considerably reduced with increasing water stress. Control (2 days interval) produced maximum leaf area (5.920 cm²), while the lowest (2.085 cm²) was recorded with increasing irrigation application interval up to 10 days. Usually, increasing irrigation interval induced poor vegetative growth by decreasing number of leaves, leaf area and shoot length. These could be explained that, drought stress decreases the cytokinin transport from root to shoots and increases leaf abscisic acid. These changes in hormone balance cause reduction in enlargement and leaf expansion and shoot growth [15]. The results are in harmony with Guerfel *et al.* (2009) who accomplished that leaf area decreased distinctly with increasing drought stress [16]. Drought stress reduced plant development by disturbing different biochemical and physiological processes, such as growth parameters [17].

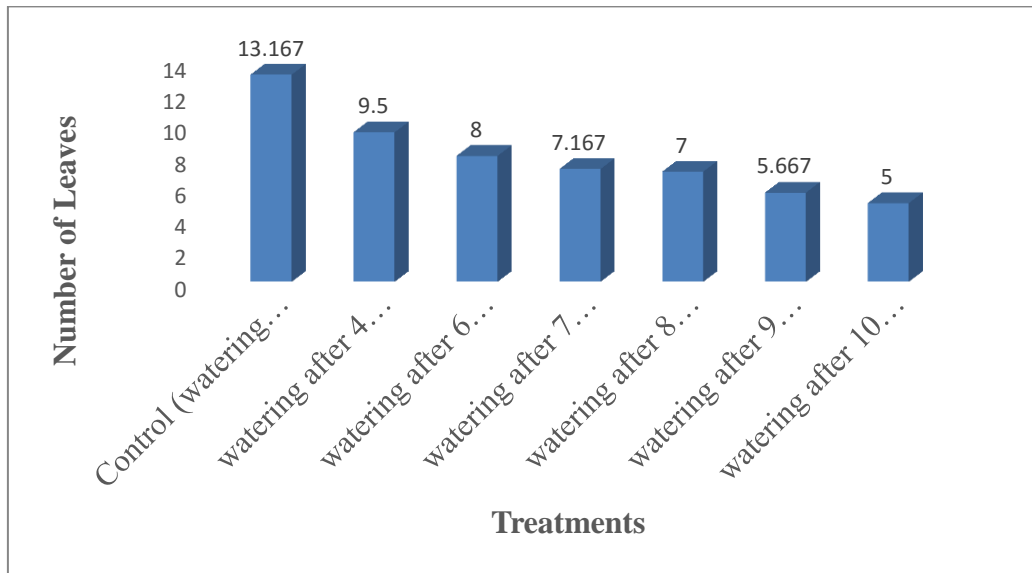


Figure 1: Effect of different water regimes treatments on number of leaves of seedlings of guava.

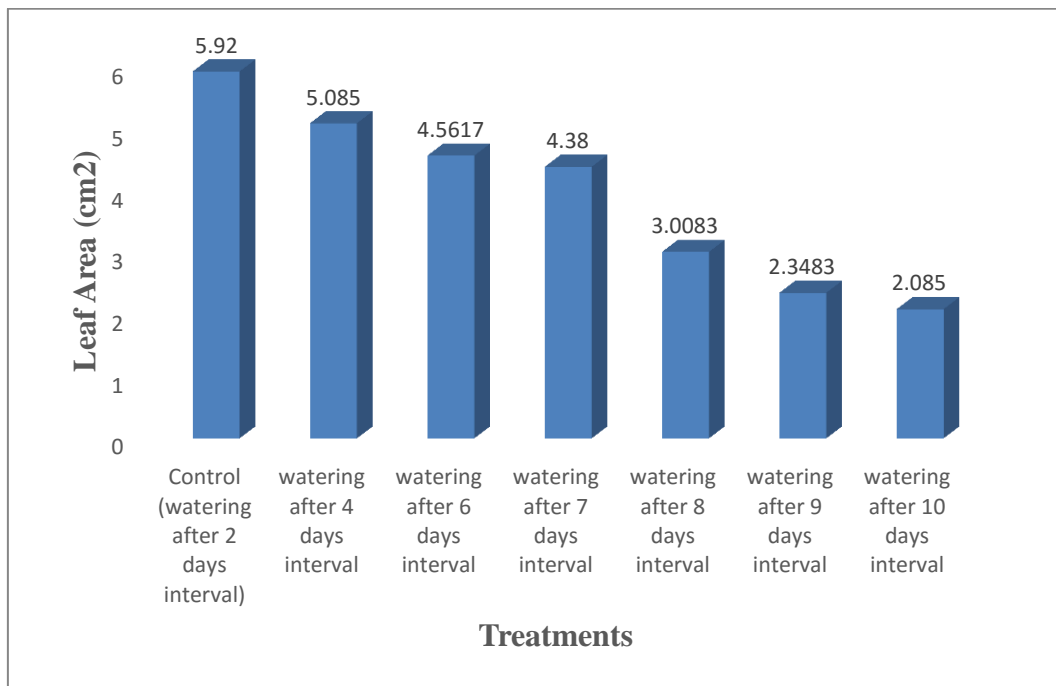


Figure 2 : Effect of different water regimes treatments on leaf Area (cm²) of seedlings of guava

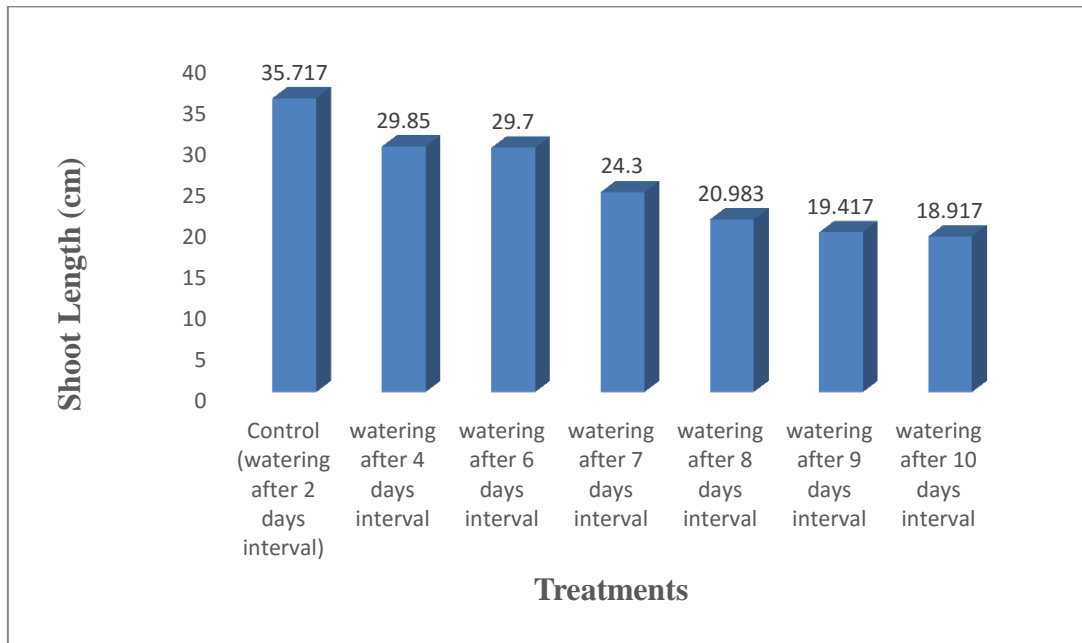


Figure 3: Effect of different water regimes treatments on Shoot length (cm) of seedlings of guava.

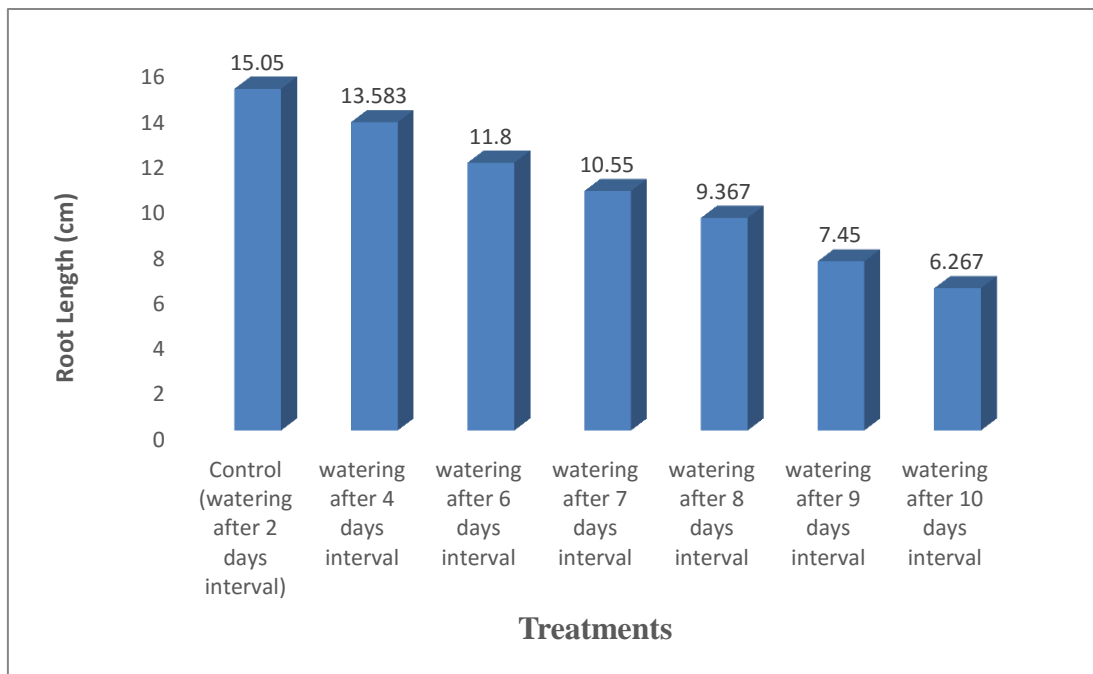


Figure 4: Effect of different water regimes treatments on root length (cm) of seedlings of guava.

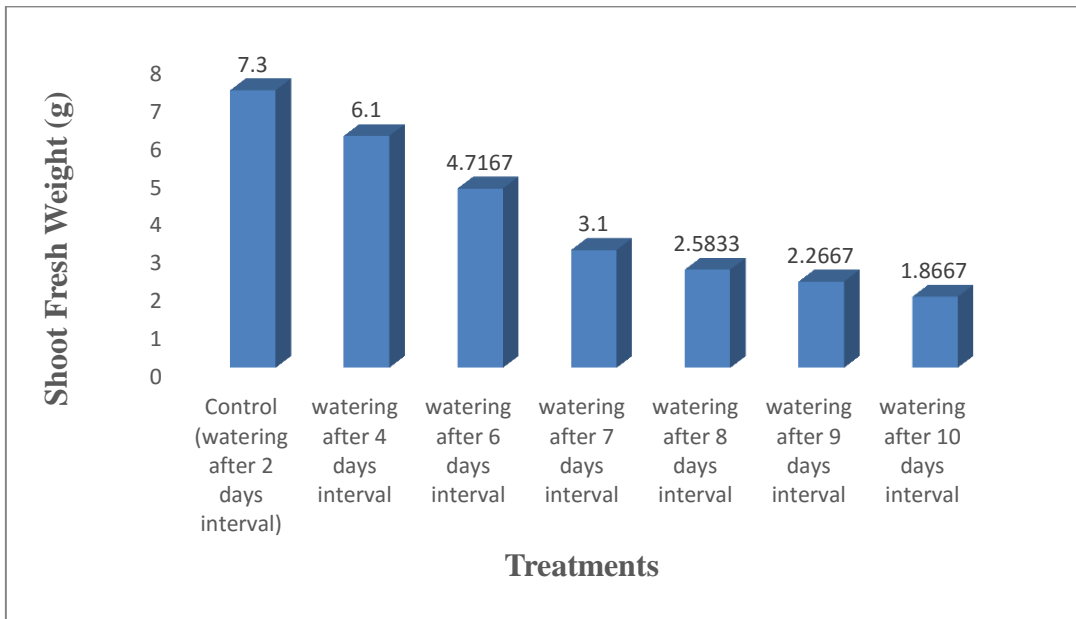


Figure 5: Effect of different water regimes treatments on shoot fresh Weight (g) of seedlings of guava.

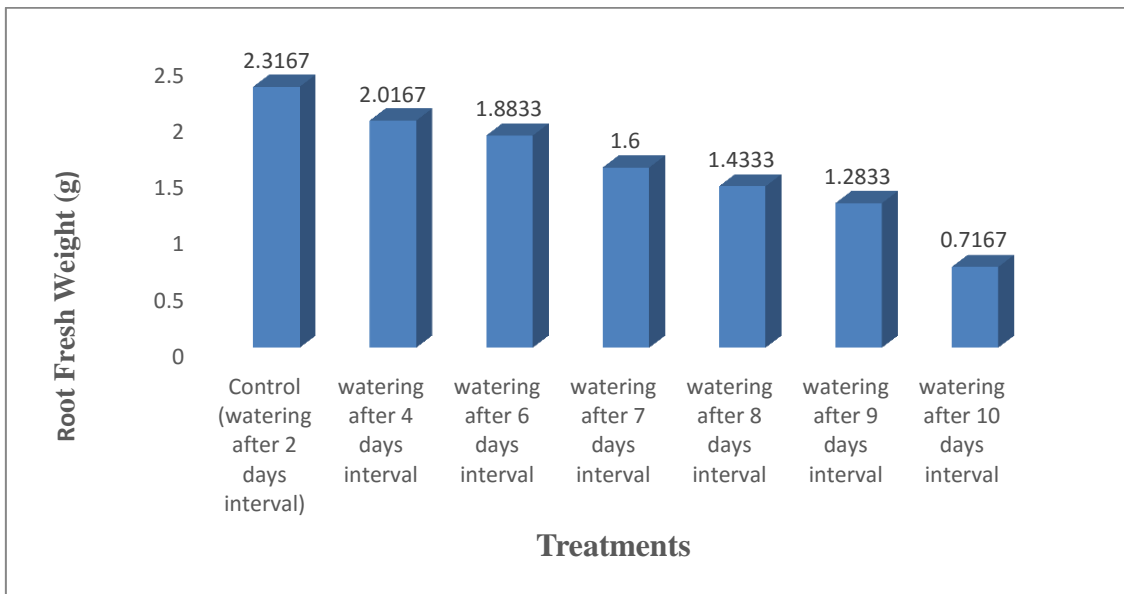


Figure 6: Effect of different water regimes treatments on root fresh Weight (g) of seedlings of guava.

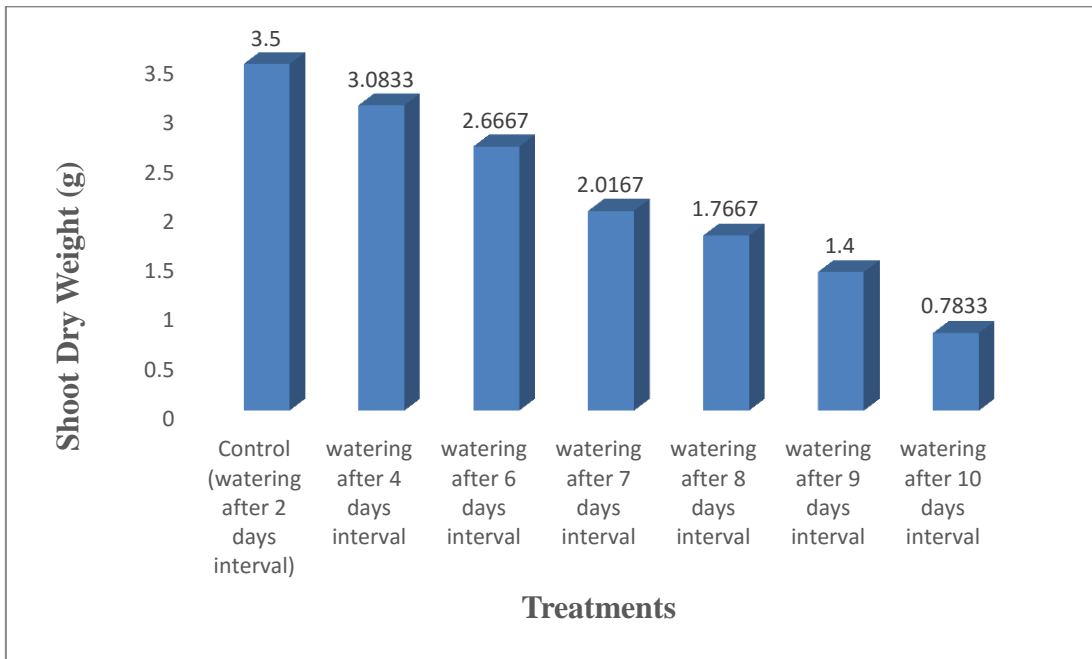


Figure 7: Effect of different water regimes treatments on shoot dry weight (g) of seedlings of guava.

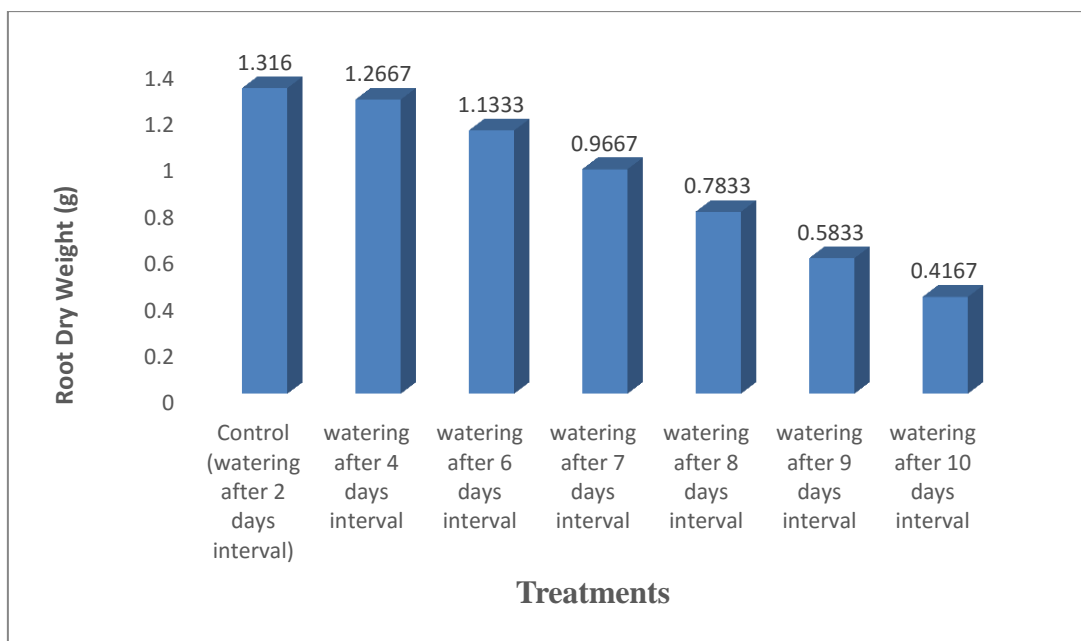


Figure 8: Effect of different water regimes treatments on root dry weight (g) of seedlings of guava.

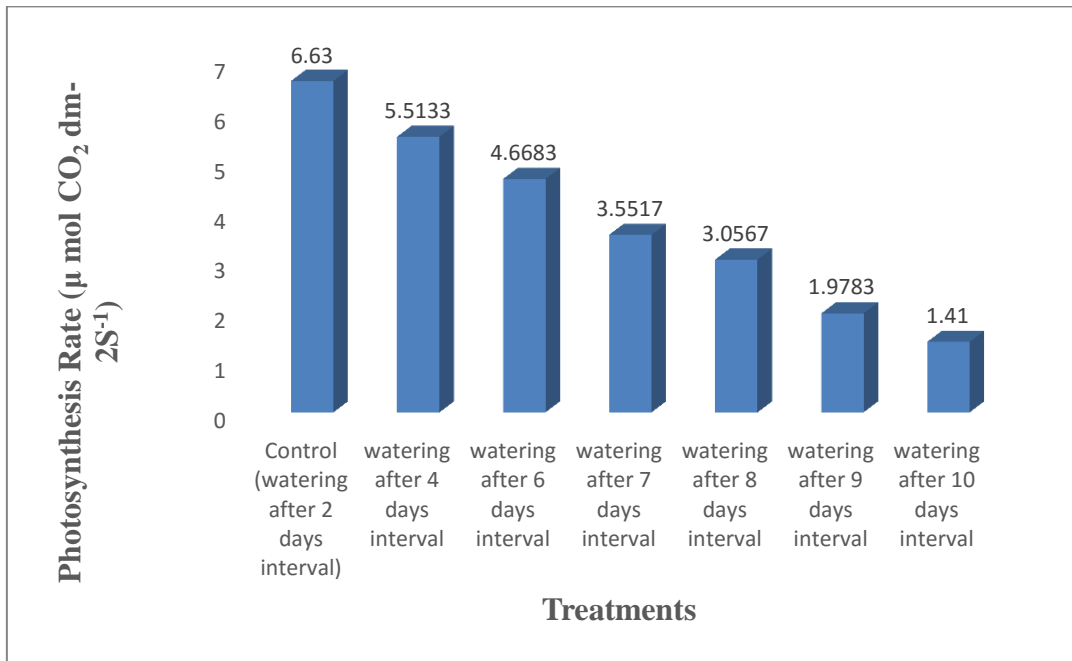


Figure 9: Effect of different water regimes treatments on photosynthesis rate ($\mu \text{ mol CO}_2 \text{ dm}^{-2} \text{ S}^{-1}$) of seedlings of guava.

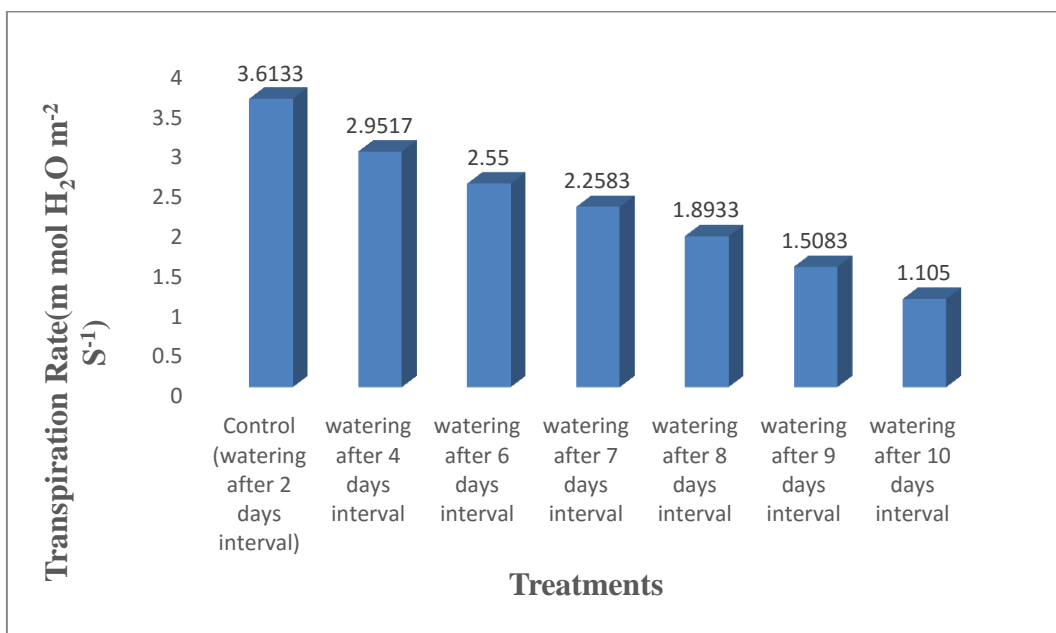


Figure 10: Effect of different water regimes treatments on transpiration rate ($\text{m mol H}_2\text{O m}^{-2} \text{ S}^{-1}$) of seedlings of guava.

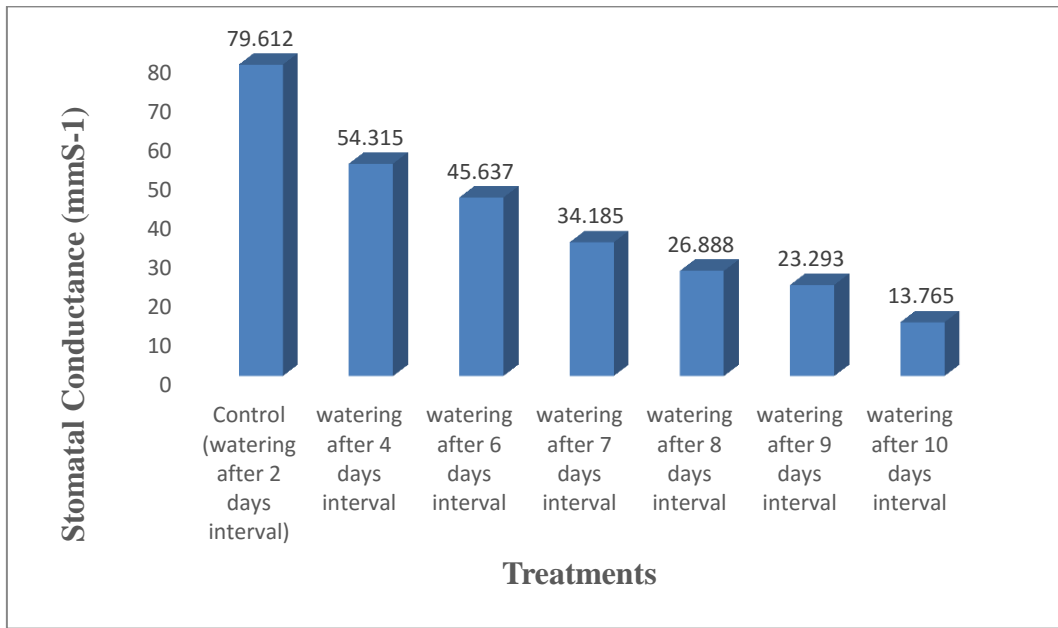


Figure 11: Effect of different water regimes treatments on stomatal conductance (mmS⁻¹) of seedlings of guava.

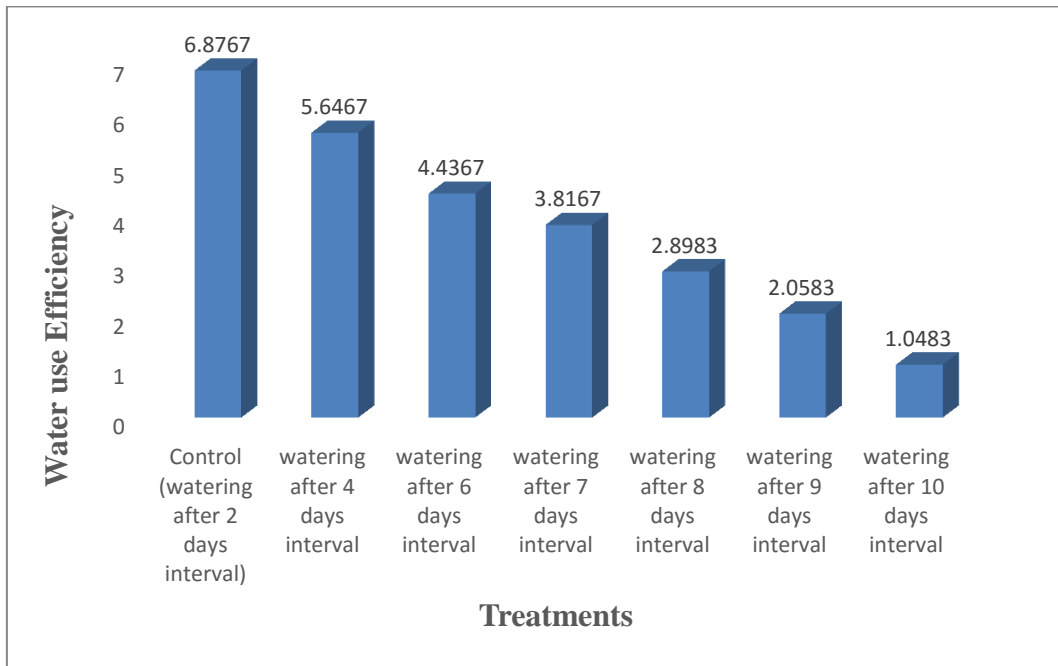


Figure 12: Effect of different water regimes treatments on water use efficiency of seedlings of guava.

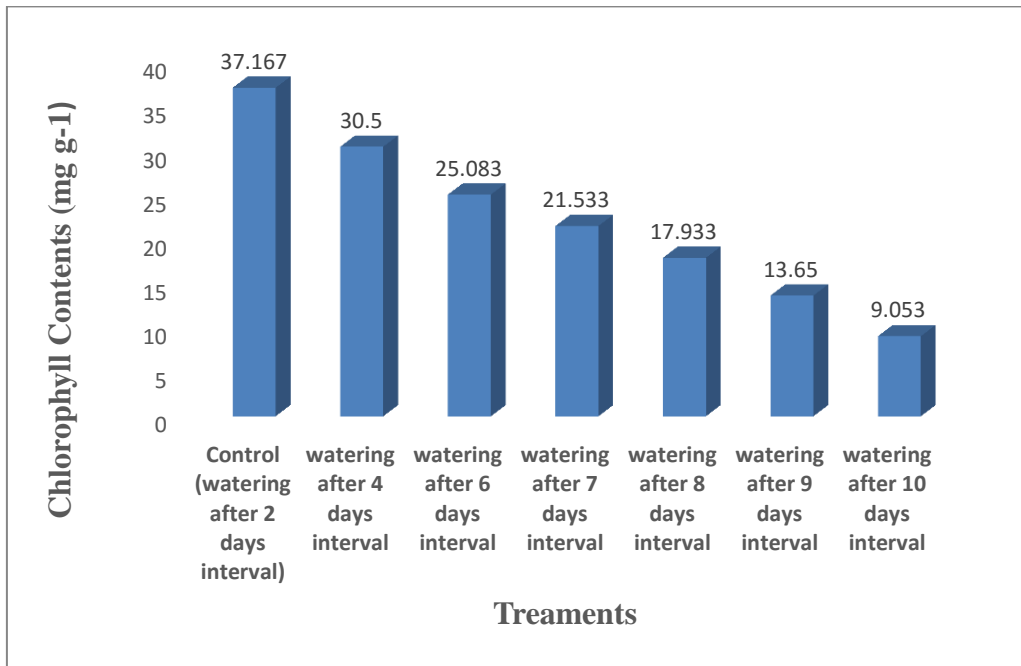


Figure 13: Effect of different water regimes treatments on chlorophyll contents (mg g⁻¹) of seedlings of guava.

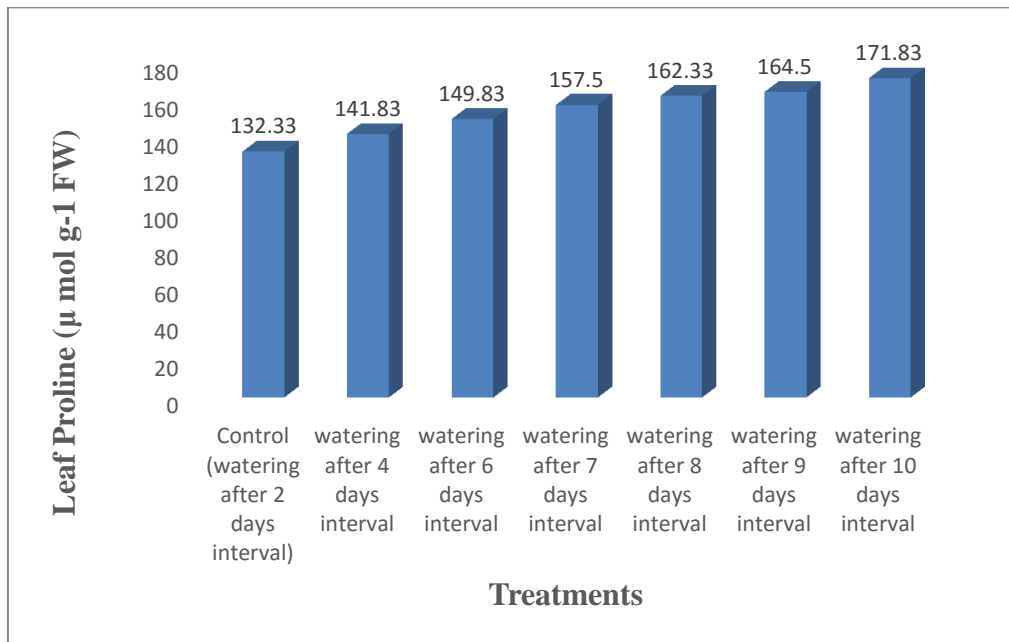


Figure 14: Effect of different water regimes treatments on leaf proline (µ mol g⁻¹ FW) of seedlings of guava.

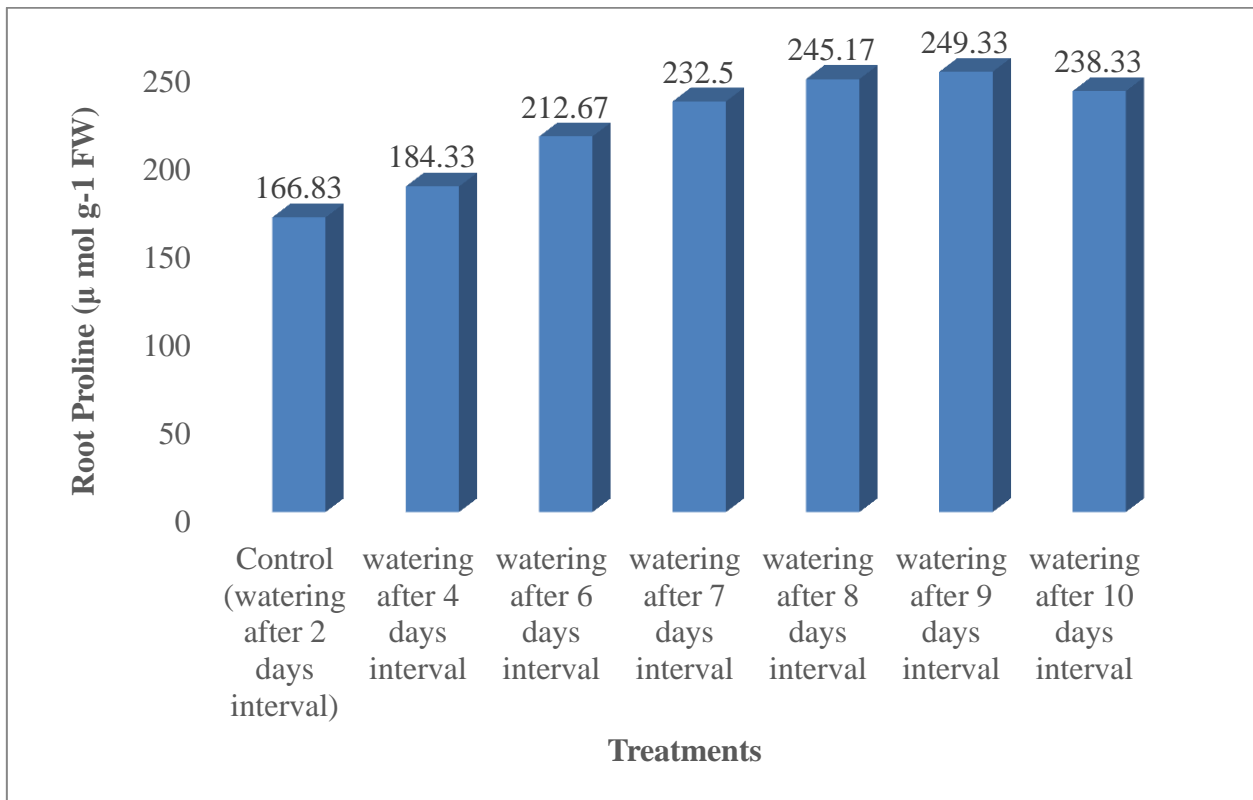


Fig. 15: Effect of different water regimes treatments on root proline (μ mol g⁻¹ FW) of seedlings of guava.

3.3. Shoot Length and root length (cm)

Shoot Length and root length are an imperative vegetative growth factor in plants. Larger shoot length results into additional space for the plant and have extra leaves for photosynthesis. Data concerning shoot length and root length are shown in Figure 3 and Figure 4. Shoot and root lengths were increased along with irrigation water applications. Plant watered after 2 days interval produced maximum shoot length (35.717cm) and root length (15.050 cm) exhibited, while minimum shoot length (18.917 cm) and root length (6.267 cm) was recorded in plant water after 10 days interval. A progressive significant reduction in shoot length root length was observed by increasing water stress by declining total available water of the soil. The present results were supported by the findings obtained by Khodarahmpour, (2011) who discovered that shoot length and root length drastically affected by irrigation regimes [18]. According to Hale and Orcutt (1987), the effect of stress may be recognized to turgor pressure loss which influences the rate of cell extension and cell size. As a consequence, water deficit reduced growth rate and stem elongation [19].

3.4. Shoot and shoot Fresh Weight

Maximum shoot fresh weight (7.30 g) and root fresh weight (2.32 g) was observed in control (T_0) that was statistically dissimilar from other treatments. On the other hand minimum shoot and root dry weights were 1.87 g and 0.72 g respectively in plants irrigated at ten days of interval. Water stress reduced the phenotypic expression of the seedling trait like fresh shoot and root weights as clear from Figure 5 and Figure 6. The findings are in union of Ali *et al.* (2011) [20]. Bibi *et al.* 2010 noticed that most of the physiological as well as morphological characteristics at seedling stage are influenced by moisture stress in sorghum [21]. Moisture stress concealed shoot enlargement more than root development moreover in certain cases root development improved.

3.5. Shoot Dry Weight (g)

Dry shoot and root weights were differed significantly among different watering regimes. Maximum dry shoot (3.50g) and root (1.32g) weights were recorded under control that was statistically dissimilar from other treatments. But lowest fresh shoot (0.78g) and root (0.42g) weights were seen for treatment T_6 (watering after 10 days interval) according to the Figure 7 and Figure 8. These results showed that shoot and root dry masses were influenced by water scarcity. Root and shoot fresh as well as dry weights were reduced under moisture stress in sorghum. Dry and fresh weight of roots and shoots were reduced throughout the drought phase because their leaf size remained undersized to decrease transpiration. Corresponding results were found by Shiralipour and West (1984) [22]. Plants first encounter drought strain in roots means that roots can intellect as well as react to the stress stipulation [18].

3.6. Photosynthesis rate ($\mu \text{ mol Co}_2 \text{ dm}^{-2} \text{ S}^{-1}$)

According to the results presented in Figure 9 maximum photosynthesis activity ($6.63 \mu \text{ mol Co}_2 \text{ dm}^{-2} \text{ S}^{-1}$)

was observed in control T_0 (watering after 2 days interval) for all treatments; maximum photosynthesis was. As water stress is limiting factor so increased irrigation application interval reduced rate of photosynthesis for transplants. Seedlings showed slightly better results at T_1 (watering after 2 days interval) $5.5133 \mu \text{ mol Co}_2 \text{ dm}^{-2} \text{ S}^{-1}$. Decreased rate of photosynthesis was observed for T_5 ($1.9783 \mu \text{ mol Co}_2 \text{ dm}^{-2} \text{ S}^{-1}$) and T_6 ($1.4100 \mu \text{ mol Co}_2 \text{ dm}^{-2} \text{ S}^{-1}$). Results represent a significant difference between the treatments. Photosynthesis decreases under water scarcity due to metabolic damage and closing of stomata. Stomata in the leaves opened more slowly in daylight and close up more rapidly in the dark under water stress [23]. Major effect of drought strain is the decline in carbon fixation coupled with closing of stomata and the subsequent raise in resistance to CO_2 flow in the leaves. This effect consequences in a decrease in the rate of leaf photosynthesis and photochemical Chl *a* fluorescence parameters [24]. Furthermore, the decrease in carbohydrates synthesis reduces plant growth and, therefore, it has a great effect on crop yield [25].

3.7. Transpiration rate ($\text{mmol H}_2\text{O m}^{-2} \text{ S}^{-1}$)

The rate of transpiration was higher in the well-watered plants (irrigated after 2 days interval) compared to the extremely stressed plants (irrigated after 8, 9 and 10 days interval) (Figure 10). The lowest rate of transpiration ($1.11 \text{ mmol H}_2\text{O m}^{-2} \text{ S}^{-1}$) was observed in plants irrigated after 10 days. On 6 and 7 days watering interval there was a non-significant difference among the treatments. However at T_6 (10 days interval) a decline in the rate of transpiration was observed under extreme water stress as compared to control where an increase in transpiration $3.61 \text{ mmol H}_2\text{O m}^{-2} \text{ S}^{-1}$ was observed. Transpiration decreased drastically in the plants under severe stress as compared to the control. Similar findings have been reported in soya beans [26], in tomato [27] and in wheat [28]. Decrease in rate of transpiration within plants under moisture stress may also be recognized to morphological changes such as increased cell wall lignification and cell wall thickness. Decreased transpiration is an essential physiological effect of stress. Nuruddin *et al.* (2003) stated that transpiration and photosynthesis are repressed instantaneously after receiving the water stress [23].

3.8. Stomatal conductance

The tendency in stomatal conductance is almost similar to that of transpiration. During this study, significant results were found concerning stomatal conductance. The stomatal conductance was highest (79.61 mmS^{-1}) in the well-watered plants (control) and lowest (13.77 mmS^{-1}) in the extremely water stressed plants (Figure 11). Decline in leaf water potential may have led to the development of a water deficit in the leaves results into loose of turgor pressure in guard cells and consequently stomatal pores to condense. Furthermore, the augmented stomatal conflict possibly will lead to condensed water transportation in the foliage promote reduction in the stomatal conductance. Stomatal conductance of plants decreased by water stress as roots of plant are incapable for absorption of water from soil under stress. Plants adopted this process under water deficit. In this stipulation a difference between loss of water by transpiration and absorption of water via roots occurred, as a result stomatal conductance reduces ultimately wilting of

plant occurred. Plants closed their stomata under moisture deficit to protect from dehydration. On the other hand, closing of stomata also stop the exchange of carbon dioxide and oxygen between its internal tissue and outside atmospheric air. In this situation uptake of nutrients by plant decreased in addition to slows down various metabolic activities in plant and probability of plant survival decreased [29].

3.9. Water use efficiency

Maximum value of water use efficiency was in control (6.8767) followed by other treatments. It is also vital to observe that control treatment represented highest water use efficiency as well as lowest water use efficiency at T₆ (1.0483) according to the Figure 12. The same results referred that in receipt of regular watering had maximum water utilization than plants in receipt of less watering under related weather conditions [30]. Crop yield reduced under drought stress despite of the growth stage at which it occurred. The drought stress influence a number of molecular and biochemical processes, which results in diminish rate of transpiration, photosynthesis, stomatal closure, pigment content in that way partial or full restriction in growth and development [31]; diminution in water-use-efficiency and leaf size, restriction of enzymatic activities .

3.10. Chlorophyll Content

Data concerning chlorophyll content is presented in Figure 13 and represented that highest chlorophyll content (37.167mg g⁻¹) was observed for the control treatment which was statistically significant from all other treatments. Lowest chlorophyll content (9.053 mg g⁻¹) was recorded for treatment T₆ (10 days interval) which was also statistically significant from all other treatments. Our results are in harmony with Dias and Bruggemann (2010) who originate that chlorophyll was decreased with increasing drought stress [32]. Kirnaket *et al.* (2001) reported that main reduction in chlorophyll content, electrolyteleakage, leaf relative water content and vegetative growth in drought stress [33]. Moreover plants developed under high moisture stress have poor fruit quality with less yield.

3.11. Leaf and root proline content

Maximum mean value of proline in leaf and root contents were 171.83 μ mol g⁻¹ FW and 238.33 μ mol g⁻¹ FW respectively were observed in stressed plant (irrigated after 10 days), while Lowest mean value of proline contents in leaf (132.33 μ mol g⁻¹ FW) and roots (166.83 μ mol g⁻¹ FW) were observed in control (Figure 14 and Figure 15).

Regarding proline content, it was remarkable that water stress increased proline accretion in the leaves. Karimi *et al.* (2012) reported a patent increase in proline content in a water stress tolerant fig cultivar under water stress [34]. Water shortage induces proline accumulation in many plant species by inactivation of its degradation or increasing its biosynthesis. Proline as an osmo-protector or as an osmoregulator, may help plant tolerate moisture scarcity. Verslues *et al.* (2006) found that proline acts as a cell membrane stabilizer and may protect cells against oxidative stress during lack of moisture [35]. Ghaderi and Siosemardeh (2011) revealed that when transplants olive

cultivars exposed to different water stress go ahead to the accumulation of proline content [36].

4. Conclusion

It is concluded that seedlings are very susceptible to water immediately after transplanting. Seedlings rising under deficit water levels were coupled with poor growth. As seedlings were under moisture stress, the water pressure within the leaves reduces furthermore seedlings show wilting. The major effect of water deficit was reduced development and growth caused by reduced photosynthesis. The water scarcity at seedling stage of growth is more constraint as compare to the later stages.

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