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# Study on Physiological Response of Drought Tolerant Groundnut Genotypes Associated with Different Stress Levels

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## ARTICLE INFO

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## ABSTRACT

Identification of drought tolerant genotypes with superior drought tolerant physiological traits is essential for the success of drought tolerance breeding program. This study was conducted to observe physiological response of drought tolerant groundnut genotypes associated with different stress levels. During 2021-2022 post-monsoon season, eight groundnut genotypes were evaluated under non-stress (field capacity), moderate stress (50 % available water) and severe stress (25 % available water) conditions. The experiment was undertaken using split plot design. Increasing drought stress levels decreased relative water content and increased canopy temperature and proline content. Total chlorophyll content increased under moderate stress condition and decreased under severe stress condition in some genotypes. The genotype ICGV-07235 and the drought tolerant check variety, Sinpadetha-12 showed consistent RWC values under different stress levels. The genotypes ICGV-07235, ICGV-07406 and Sinpadetha-12 possessed minimum canopy temperature values among the tested genotypes. Total chlorophyll content of the genotypes ICGV-07286 and ICGV-07235 were higher than that of the other tested genotypes. The genotypes ICGV-07390, YZG-07084 and ICGV-07286 had the highest proline content under stress conditions. Based on the results, the genotypes YZG-07084, ICGV-07286, ICGV-07235, ICGV-07390 and ICGV-07406 possessed desired physiological traits and these genotypes could be effectively utilized for developing drought tolerant groundnut genotypes.

## 1. Introduction

Groundnut (*Arachis hypogaea* L.) is a self-pollinated annual legume crop, mainly grown for its high-quality edible oil and food use in the tropical and warm temperate regions of the world. Groundnut is one of the major oilseed crops of Myanmar. Now a days, it is not only used as a source of edible oil but it is also consumed directly and forms an important base of many food products. Edible oil plays an important role in Myanmar as its traditional uses. In order to reduce the imported palm oil because of insufficient amount of edible oil production for local consumption, efforts are being implemented to increase yield and production of oilseed crops (MOALI, 2016). Among the oilseed crops, groundnut is the highest oil yielding crop per unit area and considered as a highly potential crop to fulfill the requirement of self-sufficiency in the country.

In Myanmar, groundnut is generally grown in monsoon season (June-October) and post-monsoon season (November-March). Monsoon season groundnut is mostly grown in Central Dry Zone areas which accounts for more than 70% of production. As a post-monsoon season crop, it is mainly grown in the Ayeyarwady delta, on plains and river banks relying solely on residual moisture. Yield of groundnut during monsoon and post-monsoon in Myanmar is 1.01ton $ha^{-1}$ and 1.48-ton $ha^{-1}$, respectively (MOALI, 2020). The lower yield in groundnut (monsoon) is mainly due to the effect of drought as the major abiotic stress during the growing season.

Drought is one of the most important factors restricting agricultural production, which seriously affects crop yield (Muhammad *et al.*, 2021). Drought induces morphological and physiological alterations in plants, including growth reduction, changes in water relation, decline of stem elongation, stomatal movement, and ion imbalance leading to yield losses and limiting crop production (Hasanuzzaman *et al.*, 2018). The susceptibility of plants to drought stress varies in dependence of stress degree, different accompanying stress factors, plant species, and their developmental stages (Demirevska *et al.*, 2009).

Drought stress disturbs turgor pressure and affects cell enlargement due to loss of cell turgidity resulting in poor plant growth (Mondal *et al.*, 2012). Drought stress negatively impacts the inherent traits such as leaf water potential ($\psi$), RWC (relative water content) and OP (osmotic potential) (Shanker *et al.*, 2014). Water stress adversely affected the chlorophyll pigments (Marcinska *et al.*, 2013). Canopy temperature is an integrative trait that reflects the plant water status or the resultant equilibrium between the root water uptake and shoot transpiration (Berger *et al.*, 2010). Under the high solar radiation and drought conditions, stomatal conductance decreases, soil moisture deficit reduces normal transpiration rate, which in turn increases canopy temperature (Rebetzke *et al.*, 2013). Thus, canopy temperature can be used to study drought tolerance in plants.

Plants have developed adaptive cellular responses to cope with the adverse effects of drought stress. The drought tolerance capacity varies among the different plant species, and most crop plants are either sensitive or moderately tolerant to drought stress (Todaka *et al.*, 2015). Osmotic adjustment is one of the biochemical strategies that protects the cellular membranes, plant proteins and other important cellular structures, and maintains water uptake under drought stress (Zhang *et al.*, 2018). Proline, an amino acid, plays an important role in plants. It protects the plants from various stresses and also helps plants to recover from stress more rapidly. Proline accumulation is a common physiological response in many plants in response to drought stress (Hayat *et al.*, 2012). Thus, in germplasm screening studies for drought tolerance, the accumulation of proline content is used as an important selection criterion (Kumar *et al.*, 2011).

Identifying groundnut varieties that are tolerant to drought stress will be of immense importance to improve crop yield. Breeding for drought tolerant genotypes is an attainable strategy to increase and sustain yield levels under challenging environments. Identification of drought tolerant genotypes with superior drought tolerant traits is essential for the success of drought tolerance breeding program. Information on physiological traits under drought stress might reveal the underlying mechanism from which improved strategies could be developed to enhance the effectiveness and progress in drought tolerance breeding. Therefore, this study was conducted to observe physiological response of drought tolerant groundnut genotypes associated with different stress levels.

## 2. Materials and Methods

### 2.1 Experimental site

This experiment was conducted at Water Utilization Research Section, Department of Agricultural Research (DAR), Myanmar during November to February, 2021-2022.

### 2.2 Experimental materials

Seven drought tolerant groundnut genotypes and drought tolerant check variety, Sinpadetha-12 that was released from DAR, were utilized. Seven drought tolerant genotypes were selected based on yield response to drought stress and studying the root and shoot characteristics by imposing Polyethylene Glycol 6000 solution.

### 2.3 Experimental design and layout

This study was conducted under field capacity as non-stress conditions (FC), moderate stress conditions (50 % available water) and severe stress conditions (25 % available water) in split plot design with three replications. Different stress levels were assigned in the main plot and groundnut genotypes were assigned in sub plot. The concrete tanks were used as the experimental plots. The size of each concrete tank was 1.8 m long, 0.9 m wide and 0.2 m deep containing 330 kg of soil. In each concrete tank, there were 6 rows of groundnut plants with 9 plants per row with a spacing of 30 cm between rows and 10 cm between plants. The total plant population was 54 plants per plot. Two seeds per hill of each genotype were sown. The plants were thinned to individual plant per hill at 7 days after sowing (DAS). Recommended cultural practices were followed throughout the growing season. Prior to sowing, soil moisture of all plots was placed at field capacity to achieve uniform germination. Soil moisture was maintained at FC until harvest under non-stress conditions. Water was withheld to allow soil moisture to decrease to meet predetermined levels of 50 % available water and 25 % available water at 40 DAS up to 80 DAS under moderate stress conditions and severe stress conditions, respectively. Irrigation was applied regularly to control soil moisture contents at predetermined levels.

### 2.4 Data collection

The following physiological parameters were recorded at 45 days after sowing (DAS) (reproductive stage R2, beginning peg), 60 DAS (reproductive stage R4, full pod), and 75 DAS (reproductive stage R6, full seed).

#### *2.4.1 Relative water content (RWC)*

Relative water content was estimated following the procedure of Barrs & Weatherly (1968). RWC was recorded between 10 to 12 am from leaflets of the third fully expanded leaf from the top of the main stem. Leaf discs of the third fully expanded leaf from the top were collected and fresh weight was recorded using electronic balance. These leaf discs were floated in distilled water for four hours in petridish. Then the discs were removed and blotted gently and weighed to record turgid weight. After that, the leaf discs were oven dried at 80 ˚C for 48 hours and dry weight was recorded. The RWC was calculated by using the following formula and expressed in percentage.

$$\text{Relative Water Content} = \frac{\text{Fresh weight (g)-Dry weight (g)}}{\text{Turgid weight (g) - Dry weight (g)}} \times 100$$

(Gonzalez *et al.*, 2001)

#### *2.4.2 Canopy temperature (˚C)*

Canopy temperature was measured from top five leaves at 11.00 am to 12.00 noon using Infrared Thermometer.

#### *2.4.3 Estimation of total chlorophyll content*

Total chlorophyll content was determined by following dimethyl sulfoxide (DMSO) method of Hiscox & Israeltam (1979). Third fully expanded leaf from the top was brought in polyethylene bags kept in an ice box from the field and was cut into small pieces; known weight of leaves containing 7.0 ml of DMSO. The test tube incubated at 65 ˚C for 30 minutes, leaf residue was removed by decanting the solution and final volume was made to 10 ml with DMSO. The absorbance of the extract was measured at 663, 645 and 470 nm in a UV-VIS spectrophotometer (Elico, SL-159) and a blank was run using DMSO. Total chlorophyll content was calculated by using the following formula and expressed in mg per g fresh weight.

$$\text{Total Chlorophyll content (mg g}^{-1}) = \frac{20.2\,(A645) + 8.02\,(A663) \times V}{1000 \times W}$$

Where,
A663 = Absorbance of the extract at 663 nm
A645 = Absorbance of the extract at 645 nm
W = Fresh weight of the sample (g)
A = Path length of cuvette (cm)
V = Final volume of the chlorophyll extract (ml)

#### *2.4.4 The amino acid proline content*

The amino acid proline content was determined with a ninhydrin-based method using cuvette spectrophotometer as described by Bates *et al.* (1973). A 2% homogenate of the fresh leaf was prepared with 10 ml of 3% sulfosalicylic acid and centrifuged at 3000 rpm for 15 min. Two milliliters of supernatant were taken and 2 ml of glacial acetic acid and acid ninhydrin reagent were added. The reaction mixture was boiled in water bath for 60 min and then cooled on ice for 5 min. Then, 4 ml of toluene was added and incubated at room temperature for 30 min. Tubes were then shaken for 15 sec and allowed to stand for 30 min for phase separation. The upper phase was separated and absorbance was measured using spectrophotometer at 520 nm and the concentration of free proline was calculated using proline standard.

### 2.5 Statistical analysis

Analysis of variance (ANOVA) was performed using Statistix (Version-8) software and mean comparison was done using Least Significant Difference (LSD) test at 5% level.

## 3. Results and Discussion

### 3.1 Relative Water content (RWC)

The values of RWC as affected by different stress levels were significantly different at 45 and 60 DAS and not significantly different at 75 DAS (Table 1). The highest RWC value was found at 45 DAS and then, it steadily declined at 60 and 75 DAS. In the initial stages of leaf development, the relative water content of leaves was higher, and it declined when the leaf matures. At 45 DAS, there was no significant difference in RWC value between non-stress and moderate stress conditions. The value of RWC under severe stress conditions was significantly lower than those under non-stress and moderate stress conditions. At 60 DAS, RWC was decreased markedly in response to declining soil water availability and the lowest value was observed under severe stress conditions indicating that RWC decreased with increased soil moisture deficit. Sepehri & Golparvar (2011) found that relative water content (RWC) contains amount of available water in leaf, increasing stress causes to decreasing it. Relative water contents of groundnut genotypes experienced to non-stress and stress treatments were not statistically different at 75 DAS, indicating that the tested groundnut genotypes did not respond much to the soil moisture conditions to change RWC of the plant as it matures.

**Table 1.** Effect of different stress levels and genotypes on RWC of groundnut plants

| Treatment | Relative water content (RWC) | | |
|---|---|---|---|
| | **45 DAS** | **60 DAS** | **75 DAS** |
| **Stress levels** | | | |
| Non-stress | 95.66 a | 93.76 a | 90.82 a |
| Moderate stress | 87.43 a | 82.11 b | 80.74 ab |
| Severe stress | 74.82 b | 71.41 c | 70.16 b |
| **LSD $_{0.05}$** | 8.75 | 3.24 | 19.17 |
| **Genotypes** | | | |
| YZG-07084 | 82.19 b | 80.95 a | 78.37 ab |
| YZG-04060 | 87.18 ab | 82.67 a | 79.21 ab |
| YZG-08010 | 80.98 b | 82.05 a | 78.21 ab |
| ICGV-07286 | 87.46 ab | 83.74 a | 78.31 ab |
| ICGV-07235 | 89.79 a | 83.40 a | 80.06 b |
| ICGV-07390 | 83.76 ab | 82.56 a | 77.74 b |
| ICGV-07406 | 86.45 ab | 84.10 a | 83.88 a |
| Sinpadetha-12 | 89.96 a | 82.86 a | 81.31 ab |
| **LSD $_{0.05}$** | 6.84 | 5.32 | 9.19 |
| **Pr>F** | | | |
| Stress levels (S) | 0.0068 | 0.0001 | 0.0952 |
| Genotypes (G) | 0.0852 | 0.8626 | 0.2548 |
| S × G | 0.0144 | 0.9972 | 0.4601 |
| **CV $_a$ (%)** | 14.99 | 14.97 | 16.75 |
| **CV $_b$ (%)** | 9.6 | 8.97 | 12.72 |

*DAS = Days after sowing*
*Means followed by the same letter in each column are not significantly different at 5% level.*

There were no significant differences in RWC among groundnut genotypes at all recorded dates. No significant interaction between different stress levels and genotypes was observed in RWC at 60 and 75 DAS. This pointed out that groundnut genotypes did not respond to different stress levels in RWC of the plant at 60 and 75 DAS. Painawadee *et al.* (2009) indicated that RWC was sensitive in identifying drought stress even in the same groundnut genotypes with different stress levels in case of appropriate stress level. Significant interaction between different stress levels and groundnut genotypes was found in RWC values at 45 DAS. This explained that the effect of different stress levels varied with groundnut genotypes at 45 DAS. Under non-stress conditions, groundnut genotypes were not significantly different in RWC values (Figure 1). In case of moderate stress, there was no significant difference in RWC value among groundnut genotypes. This is possibly due to slow response of groundnut genotypes in RWC to declining stress levels and response of groundnut genotypes did not vary with different stress levels under moderate stress condition. Under severe stress conditions, groundnut genotypes were significantly different in RWC values. This is possibly due to variation of groundnut genotypes for drought tolerance under severe stress condition. The genotype ICGV-07235 had maximum RWC as similar as drought tolerant check variety, Sinpadetha-12, and can be a promising genotype with the high RWC at 45 DAS. Generally, although groundnut genotypes were significantly different in RWC values among different stress levels, RWC values of the drought tolerant check, Sinpadetha-12 and ICGV-07235 were consistent under different stress levels. This may be due to the fact that these genotypes could maintain higher leaf turgor levels during periods of drought stress and they are desirable genotypes by RWC.

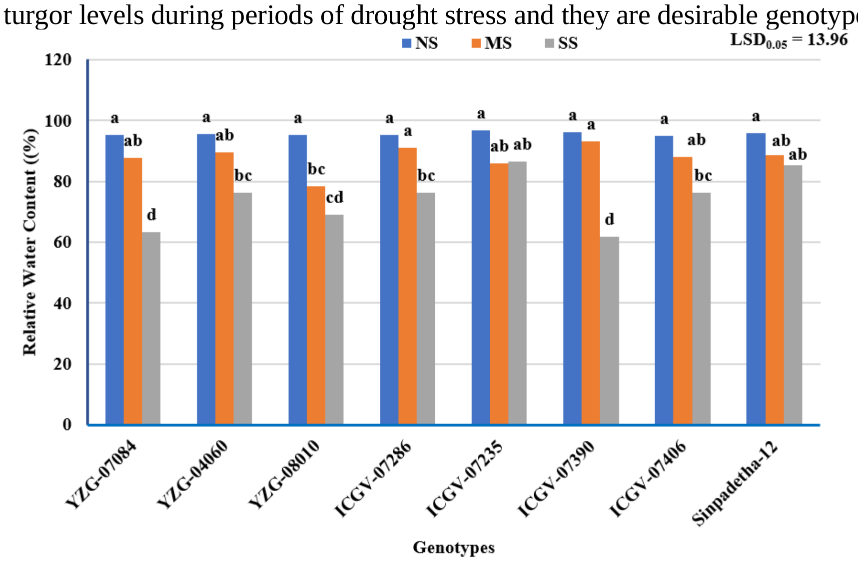



**Figure 1.** Mean value of relative water content (RWC) as affected by combination of different stress levels (NS = Non-stress; MS = Moderate stress; SS = Severe stress) and groundnut genotypes at 45 DAS

**3.2 Canopy temperature**

Canopy temperatures were highly and significantly different between different stress levels throughout 45 to 75 DAS (Table 2). Canopy temperature was increased with increasing water deficit levels in all observations. This may be due to dense canopies under non-stress condition and sparse canopies under severe stress condition. Canopy temperature values were significantly different among groundnut genotypes at 45 and 60 DAS and no significant difference was observed among groundnut genotypes at 75 DAS. This may be due to difference in canopy performance. The drought tolerant check, Sinpadetha-12 showed low values of canopy temperature at 45 and 60 DAS. Canopy may be cooler because of their ability to transfer relatively more heat back to the atmosphere by reflection.

The highly significant interaction between different stress levels and groundnut genotypes was observed in canopy temperature at 45 DAS. This indicated that the effect of different stress levels on canopy temperature varied with groundnut genotypes at 45 DAS. No significant difference in canopy temperature values was observed among groundnut genotypes under non-stress and moderate stress conditions but canopy temperature values were significantly different among groundnut genotypes under severe stress conditions at 45 DAS (Figure 2). All the tested groundnut genotypes showed increasing trend of canopy temperature due to drought stress that might have occurred due to increased respiration and decreased transpiration resulting from stomatal closure. Under moderate stress conditions, the genotypes YZG-07084, ICGV-07235 and ICGV-07390 showed the lower canopy temperature value as similar as drought tolerant check variety, Sinpadetha-12 and these genotypes also possessed the lowest and statistically similar canopy temperature value to drought tolerant check under severe stress conditions. Therefore, these genotypes can be promising genotypes with low canopy temperature value at 45 DAS. Genotypes with lower canopy temperatures may possess enhanced capacity to take up soil moisture or to maintain a better plant water status.

No significant interaction between different stress levels and groundnut genotypes was observed in canopy temperature value at 60 and 75 DAS, indicating that the effect of different stress levels on canopy temperature did not vary with groundnut genotypes at 60 and 75 ADS.

**Table 2.** Effect of different stress levels and genotypes on canopy temperature of groundnut plants

| Treatment | Canopy temperature (˚C) 45 DAS | 60 DAS | 75 DAS |
|---|---|---|---|
| **Stress levels** | | | |
| Non-stress | 30.33 c | 29.16 b | 27.36 c |
| Moderate stress | 35.02 b | 34.45 a | 33.92 b |
| Severe stress | 39.25 a | 35.09 a | 38.45 a |
| **LSD $_{0.05}$** | 0.69 | 1.37 | 2.30 |
| **Genotypes** | | | |
| YZG-07084 | 34.63 bc | 32.71 b | 33.33 a |
| YZG-04060 | 34.46 b | 33.46 a | 32.20 a |
| YZG-08010 | 36.52 a | 31.83 b | 33.70 a |
| ICGV-07286 | 35.46 b | 32.29 b | 32.72 a |
| ICGV-07235 | 33.48 d | 32.17 b | 32.72 a |
| ICGV-07390 | 34.30 cd | 35.46 a | 34.04 a |
| ICGV-07406 | 35.06 bc | 31.36 b | 32.56 a |
| Sinpadetha-12 | 34.17 cd | 31.62 b | 32.63 a |
| **LSD $_{0.05}$** | 0.99 | 2.74 | 1.75 |
| **Pr>F** | | | |
| Stress levels (S) | <0.0001 | 0.0005 | 0.0005 |
| Genotypes (G) | <0.0001 | 0.0035 | 0.3080 |
| S × G | <0.0001 | 0.1735 | 0.1359 |
| **CV $_a$ (%)** | 3.35 | 8.39 | 8.43 |
| **CV $_b$ (%)** | 3.19 | 4.61 | 5.26 |

*DAS= Days after sowing*
*Means followed by the same letter in each column are not significantly different at 5% level.*

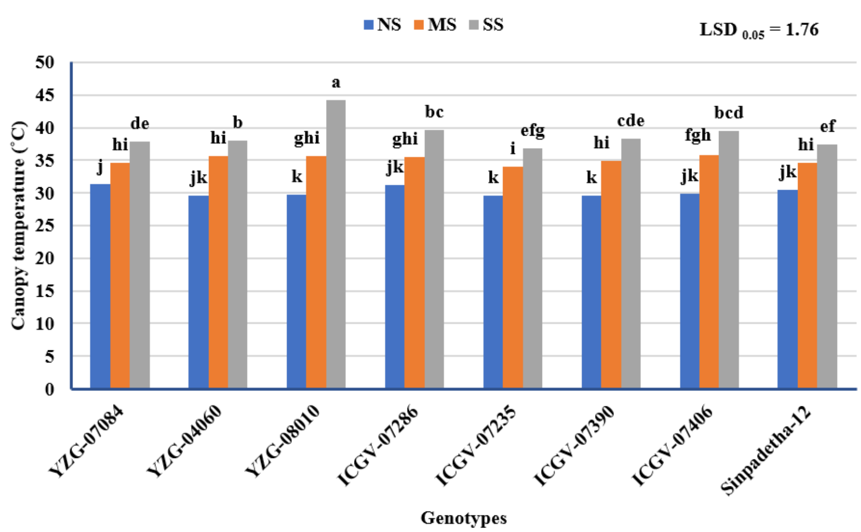



**Figure 2.** Mean value of canopy temperature as affected by combination of different stress levels (NS = Non-stress; MS = Moderate stress; SS = Severe stress) and groundnut genotypes at 45 DAS.

### 3.3 Total chlorophyll content

The values of total chlorophyll content were significantly different among different stress levels at 45 and 60 DAS but not significantly different at 75 DAS (Table 3). The maximum total chlorophyll content was achieved in moderate stress conditions although no significant difference with non-stress condition, and minimum total chlorophyll content was found in severe stress conditions in all observations. Moderate drought stress increased total chlorophyll content and the levels of total chlorophyll content was decreased under severe stress condition. Moderate drought stress may increase the concentration of chlorophyll per unit area because water losses cause to increasing of contraction of cells resulting to increased cell concentration whereas severe stress may stop making chlorophyll. Sepehri & Golparvar (2011) indicated that chlorophyllase and peroxidase enzymes increased, at result, chlorophyll content decreased under severe drought stress conditions. The decrease in chlorophyll was attributed to the inhibition of chlorophyll synthesis as well as to the accelerated turnover of the chlorophyll already present.

From 45 to 75 DAS, the total chlorophyll content was highly and significantly different among the groundnut genotypes. The genotype ICGV-07286 showed higher total chlorophyll content than the other genotypes in all observations and ICGV-07235 also possessed higher total chlorophyll content than other genotypes at 45 and 75 DAS. This indicated that ICGV-07286 and ICGV-07235 had high chlorophyll in leaves and increased photosynthetic capacity. Chlorophyll is an important photosynthetic pigment to the plant, largely determining photosynthesis, the most important source of energy for plant growth. Sepehri & Golparvar (2011) reported that effect of drought stress on chlorophyll depends on plant genotypes and environmental conditions; in some varieties, drought stress reduces and, in some varieties, increases chlorophyll content.

The significant interactions between different stress levels and genotypes were observed in total chlorophyll content in all observations (Table 3), indicated that the effect of different stress levels on total chlorophyll content varied with genotypes. In some genotypes (e.g., Sinpadetha-12), total chlorophyll content tended to decrease with increasing stress levels. In some genotypes (e.g., YZG-08010, ICGV-07286, ICGV-07235 and ICGV-07406 at 45 DAS; YZG-07084 and YZG-04060 at 60 DAS; YZG-04060, ICGV-07286 and ICGV-07390 at 75 DAS), maximum total chlorophyll content was observed under moderate stress condition (Figure 3). Drought stress decreased total chlorophyll content although some genotypes increased total chlorophyll content under moderate stress conditions. At 45 DAS, the genotypes ICGV-07286 and ICGV-07235 under moderate stress conditions possessed the higher total chlorophyll content which was not statistically different under non-stress conditions but significantly different under severe stress conditions. Severe stress decreased total chlorophyll content of these genotypes at 45 DAS. The genotype ICGV-07390 and YZG-07084 resulted in no significant changes in total chlorophyll content under different stress levels at 45 DAS.

**Table 3.** Effect of different stress levels and genotypes on total chlorophyll content of groundnut plants

| Treatment | Total chlorophyll content (mg $g^{-1}$) | | |
|---|---|---|---|
| | **45 DAS** | **60 DAS** | **75 DAS** |
| **Stress levels** | | | |
| Non-stress | 2.3387 a | 2.4209 a | 2.3533 ab |
| Moderate stress | 2.3902 a | 2.5604 a | 2.4429 a |
| Severe stress | 2.1571 b | 2.2653 b | 2.0174 b |
| **LSD $_{0.05}$** | 0.1339 | 0.1554 | 0.3405 |
| **Genotypes** | | | |
| YZG-07084 | 2.2495 b | 2.3559 cd | 2.0411 c |
| YZG-04060 | 2.3365 b | 2.3071 d | 2.1264 bc |
| YZG-08010 | 2.0228 c | 2.3538 cd | 2.3049 ab |
| ICGV-07286 | 2.6028 a | 2.6598 a | 2.4923 a |
| ICGV-07235 | 2.5442 a | 2.4825 bc | 2.4840 a |
| ICGV-07390 | 2.2970 b | 2.5627 ab | 2.1854 bc |
| ICGV-07406 | 2.2480 b | 2.3063 d | 2.4234 a |
| Sinpadetha-12 | 2.0619 c | 2.2962 d | 2.1118 bc |
| **LSD $_{0.05}$** | 0.1044 | 0.1327 | 0.2049 |
| **Pr>F** | | | |
| Stress levels (S) | 0.0180 | 0.0158 | 0.0529 |
| Genotypes (G) | < 0.0001 | < 0.0001 | < 0.0001 |
| S × G | < 0.0001 | < 0.0001 | 0.0013 |
| **CV $_a$ (%)** | 14.58 | 15.97 | 13.19 |
| **CV $_b$ (%)** | 13.38 | 12.48 | 12.34 |

*DAS= Days after sowing*

*Means followed by the same letter in each column are not significantly different at 5% level.*

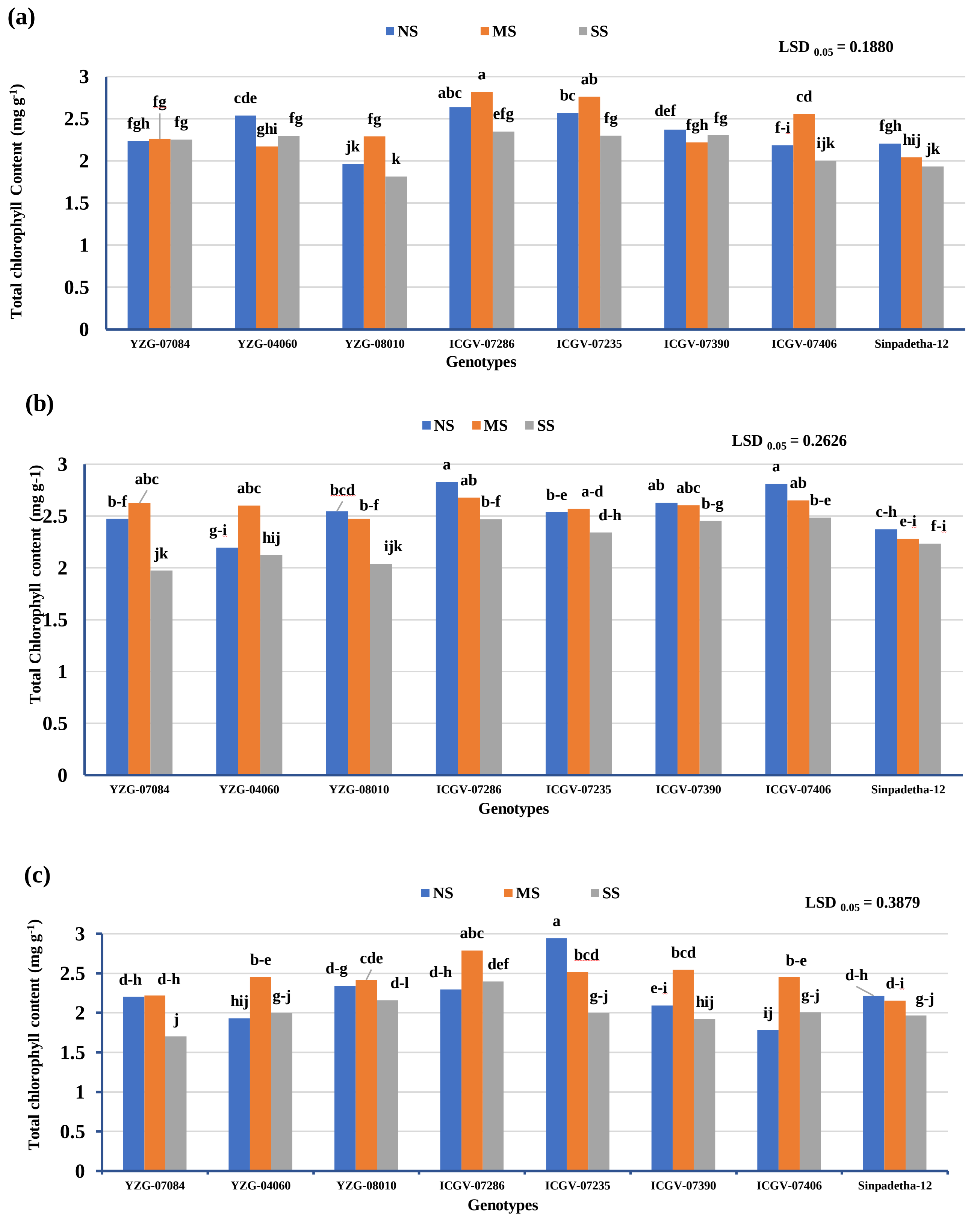



**Figure 3.** Mean value of total chlorophyll content as affected by combination of different stress levels (NS= Non-stress, MS = Moderate stress, SS = Severe stress) and groundnut genotypes at (a) 45 DAS, (b) 60 DAS and (c) 75 DAS

### 3.4 Proline content

The highly significant difference in proline content was observed among different stress levels from 45 to 75 DAS (Table 5). Proline content continuously increased from 45 to 75 DAS under moderate and severe stress conditions. Proline content increased with increasing stress levels. Pireivatloum *et al.* (2010) reported that proline content increased sharply by about four-to-sevenfold under water stressed conditions.

Groundnut genotypes were significantly different in proline content in all observations. Proline content of all tested groundnut genotypes continuously increased from 45 to 75 DAS. Among the tested genotypes, significantly highest proline content was observed in the genotype ICGV-07390 and it was followed by YZG-07084 at all observation dates.

Highly significant interaction between different stress levels and groundnut genotypes was observed in proline content in all observations. Among the genotypes, the highest proline content was observed in ICGV-07390 under severe stress conditions followed by YZG-07084 at 45 and 60 DAS (Figure 4 a and b, respectively). At 75 DAS, the genotype ICGV-07286 under severe stress conditions produced the significantly highest proline content followed by YZG-07084 and it was significantly higher than the drought tolerant check, Sinpadedtha-12. Although proline content increased as soil moisture deficit increased, effect of different stress levels on proline content varied with genotypes, for example, ICGV-07235 at 45 and 60 DAS and YZG-08010 at 75 DAS. In these genotypes, proline content did not increase under severe stress condition. This may be due to the fact that prolonged severe drought stress caused serious metabolic damages and decreased proline accumulation in some genotypes. Liu *et al.* (2011) indicated that prolonged drought treatments significantly increased proline content in six woody plant species under moderate stress, increased proline content in one species under severe stress, but decreased proline content in three species under severe stress.

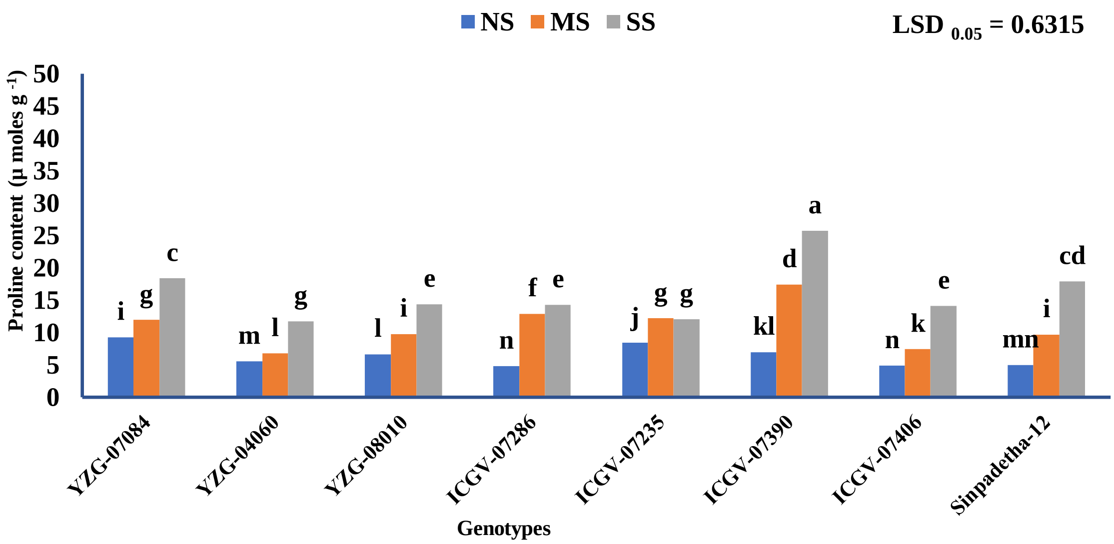
**Table 4.** Effect of different stress levels and genotypes on proline content of groundnut plants

| **Treatment** | **Proline content (μ moles $g^{-1}$)** | | |
|---|---|---|---|
| | **45 DAS** | **60 DAS** | **75 DAS** |
| **Stress levels** | | | |
| Non-stress | 5.959 c | 7.032 c | 6.398 c |
| Moderate stress | 12.283 b | 13.356 b | 24.623 b |
| Severe stress | 15.533 a | 16.606 a | 36.001 a |
| **$LSD_{0.05}$** | 0.3680 | 0.3742 | 0.0188 |
| **Genotypes** | | | |
| YZG-07084 | 13.194 b | 14.267 b | 26.203 ab |
| YZG-04060 | 8.024 g | 9.097 g | 18.322 e |
| YZG-08010 | 10.269 e | 11.342 e | 23.474 c |
| ICGV-07286 | 10.678 d | 11.751 d | 25.424 b |
| ICGV-07235 | 10.894 c | 11.967 c | 26.405 ab |
| ICGV-07390 | 16.703 a | 17.776 a | 26.763 a |
| ICGV-07406 | 8.808 f | 9.881 f | 23.277 c |
| Sinpadetha-12 | 10.862 d | 11.935 d | 19.556 d |
| **$LSD_{0.05}$** | 0.322 | 0.183 | 1.086 |
| **Pr>F** | | | |
| Stress levels (S) | <0.0001 | <0.0001 | <0.0001 |
| Genotypes (G) | <0.0001 | <0.0001 | <0.0001 |
| S × G | <0.0001 | <0.0001 | <0.0001 |
| **$CV_a$ (%)** | 4.08 | 3.85 | 4.73 |
| **$CV_b$ (%)** | 3.23 | 3.3 | 4.69 |

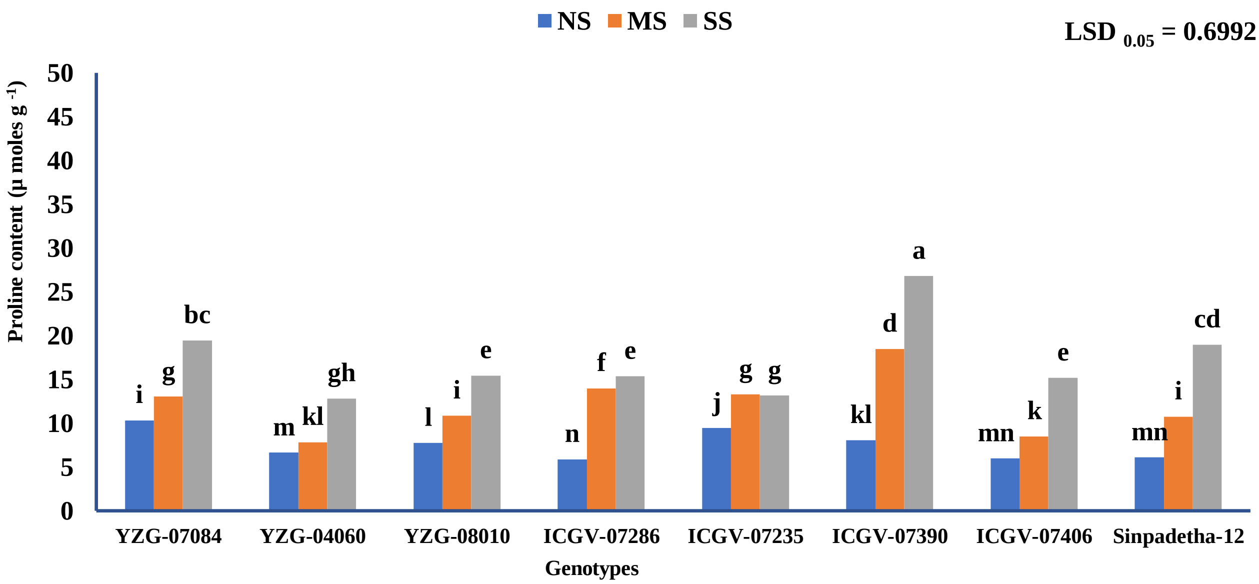
*DAS= Days after sowing*

*Means followed by the same letter in each column are not significantly different at 5% level.*

(a)


NS MS SS
LSD 0.05 = 0.6315
Proline content (μ moles g -1)
50
45
40
35
30
25
20
15
10
5
0
i g c
m l g
l i e
n f e
j g g
kl d a
n k e
mn i cd
YZG-07084
YZG-04060
YZG-08010
ICGV-07286
ICGV-07235
ICGV-07390
ICGV-07406
Sinpadetha-12
Genotypes

(b)


NS MS SS
LSD 0.05 = 0.6992
Proline content (μ moles g -1)
50
45
40
35
30
25
20
15
10
5
0
i g bc
m kl gh
l i e
n f e
j g g
kl d a
mn k e
mn i cd
YZG-07084
YZG-04060
YZG-08010
ICGV-07286
ICGV-07235
ICGV-07390
ICGV-07406
Sinpadetha-12
Genotypes

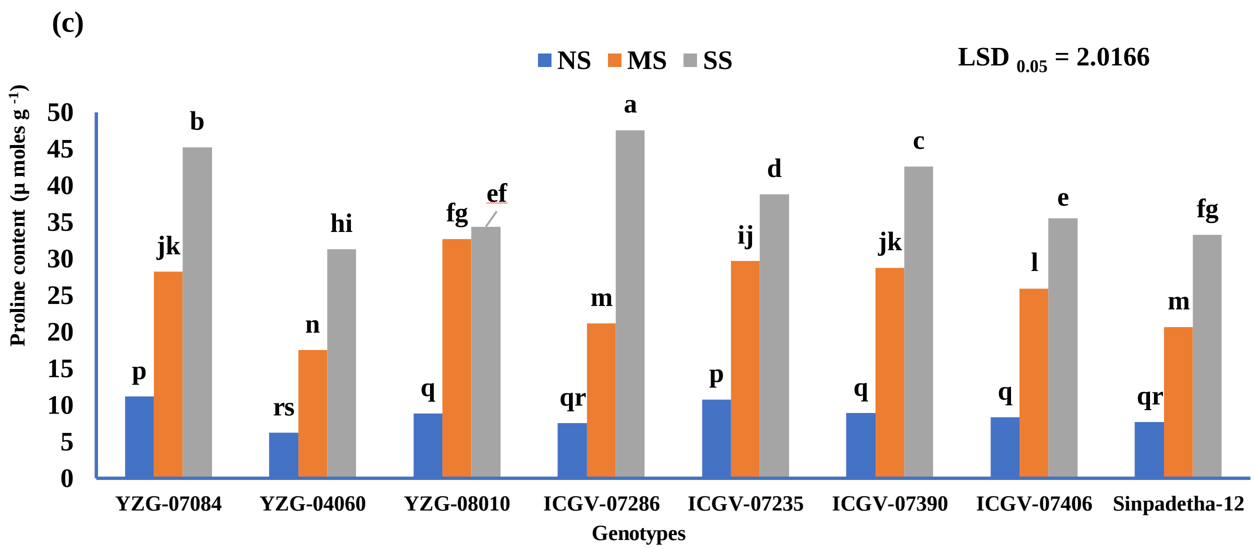



**Figure 4.** Mean value of proline content as affected by combination of different stress levels (NS = Non-stress, MS = Moderate stress, SS = Severe stress) and groundnut genotypes at (a) 45 DAS, (b) 60 DAS and (c) 75 DAS

## 4. Conclusion

Drought stress affected relative water content, canopy temperature, total chlorophyll content and proline content. Increasing drought stress levels decreased relative water content, increased canopy temperature and proline content. Total chlorophyll content increased under moderate stress conditions and decreased under severe stress condition in some genotypes. Different physiological responses were observed among the tested groundnut genotypes. In the present study, decreased RWC values under soil moisture deficit conditions showed a higher degree of plant stress due to drought. The genotypes ICGV-07235 and the drought tolerant check, Sinpadetha-12 showed consistent RWC values under different stress levels implying that these two genotypes could maintain higher leaf turgor levels during periods of drought stress. The genotypes ICGV-07235, ICGV-07406 and Sinpadetha-12 possessed minimum canopy temperature values among the tested genotypes and thus these genotypes seemed to have a better capacity for maintaining a better plant water status. Total chlorophyll content of the genotypes ICGV-07286 and ICGV-07235 were higher than that of the other tested genotypes and, thus, these genotypes may have enhanced photosynthetic capacity. Among the tested groundnut genotypes, ICGV-07390, YZG-07084 and ICGV-07286 showed the highest proline content under stress conditions. The increased proline accumulation plays adaptive role to impart tolerance in plants and thus the genotypes with the highest proline content can be considered as the most drought tolerant genotypes. Based on the results, the genotypes; YZG-07084, ICGV-07286, ICGV-07235, ICGV-07390 and ICGV-07406 possessed desired physiological traits. The finding of this study suggested that selecting a drought tolerant groundnut genotype with this desired combination of physiological characters will be a challenging task in future breeding programs for increasing groundnut production.

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## 6. Authors Note

The authors declare that there is no conflict of interest regarding the publication of this article. Authors confirmed that the paper was free of plagiarism.

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