



Effect of Intermittent Water Supply on Yield and Seed Quality of Peanut (*Arachis hypogaea* L.) under Drought Stress

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ABSTRACT

Peanut (*Arachis hypogaea* L.) is a globally cultivated legume crop, valued for its nutritional properties and wide range of applications. However, its production is highly susceptible to water scarcity and the detrimental effects of drought stress. This study aimed to evaluate the impact of intermittent water supply on peanuts under drought conditions (60% field capacity). The findings demonstrated that intermittent water supply treatments positively affected peanut growth and yield compared to continuous drought stress. The treatment applied from day 80 until harvest significantly improved pod number per plant, seed number per pod, and seed weight, approaching levels observed in the control group. Additionally, the oil content of seeds from the intermittent water supply treatment increased by approximately 20% compared to the control. The intermittent water supply (60-80% field capacity treatment, applied from day 80 until harvest) also enhanced chlorophyll *a* content and respiration rate, bringing them to levels comparable to the control. Furthermore, the intermittent water supply treatment from day 80 until harvest notably increased starch content, particularly soluble sugars, proteins, and proline, which were similar to those observed in the control group. These results underscore the potential of intermittent water supply as an effective irrigation strategy for sustainable peanut production under drought conditions.

Introduction

Peanut (*Arachis hypogaea* L.) is a legume crop of significant economic importance, cultivated globally for its nutritional value and diverse applications (Boukid, 2022). However, peanut production is highly susceptible to water scarcity and drought stress, primarily due to its high water demand during critical growth stages (Palmero et al., 2022). Drought stress has profound negative

effects on peanut growth and development, resulting in notable reductions in the number of pods, seeds, and seed weight, with respective decreases of 16.60 to 2.40, 31.10 to 5.10, and from 0.62 to 0.51 g. Moreover, the photosynthetic rate of peanut plants under drought stress is significantly reduced by approximately one-third compared to control conditions (Tran et al.,

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In response, researchers have increasingly explored alternative irrigation methods, such as intermittent water supply (IWS), to enhance water use efficiency and sustain acceptable yields under water-limited conditions (Sarisen et al., 2022). IWS involves providing water to crops at pulsed or intermittent intervals, rather than through continuous irrigation (Fan et al., 2014). This approach aims to conserve water by delivering it during key growth stages or critical periods when crops are most vulnerable to water stress. The intermittent nature of water application allows plants to adapt and optimize water usage, potentially improving water use efficiency and enhancing drought tolerance (Bodner et al., 2015).

Several studies have demonstrated the success of IWS in various crop species. For instance, research on soybeans subjected to intermittent water supply (every three days, with each watering session lasting two hours) reported improved water use efficiency and maintained acceptable yields during the dry season, exceeding 1.25 tons per hectare (Haryati et al., 2021). Similarly, intermittent irrigation has been shown to enhance water use efficiency and sustain productivity in mungbean during arid periods (Ukwu et al., 2024). In a study of ten mungbean accessions, no significant differences were observed in growth and yield traits between plants under control conditions and those subjected to a 3-day intermittent irrigation regime, further illustrating the potential of IWS as an effective strategy in diverse crop systems.

Despite these promising findings, the effects of intermittent water supply on peanut growth and its capacity to mitigate drought stress remain relatively underexplored. Therefore, this study aims to assess the impact of IWS on peanut crops under drought stress. By examining the physiological, biochemical, and morphological responses of peanut plants subjected to different IWS treatments, this research seeks to provide valuable insights into the effectiveness and limitations of IWS in peanut cultivation. The findings will contribute to a broader understanding of IWS as a sustainable irrigation strategy for peanut production in water-limited environments.

Materials and Methods

Plant materials

The plant material used in this study was the peanut variety VD01-2, obtained from the Research Institute of Oil and Oil Plant in Ho Chi Minh City, Vietnam. VD01-2 is a locally adapted

cultivar known for its favorable growth characteristics and high yield potential.

Effect of intermittent water supply on peanut growth under drought stress

Seeds were saturated with water and planted in plastic pots (22 x 16 x 17 cm) filled with experimental soil weighing 4.5 kg. The physical and chemical characteristics of the experimental soil consisted of 63.4% sand, 28.5% silt, and 8.1% clay. The soil had an organic matter content of 24.91 g kg⁻¹, a total nitrogen content of 0.165%, an available phosphorus content of 0.062%, and an available potassium content of 0.93%. The concentrations of zinc, boron, copper, and molybdenum in the soil were measured as 733 mg kg⁻¹, 98 mg kg⁻¹, 26 mg kg⁻¹, and 0.9 mg kg⁻¹, respectively. The pH value of the soil was approximately 5.07, and its cation exchange capacity was 3.4 meq 100 g⁻¹. The pots were placed in the experimental orchard under a measured light intensity of 60,000 ± 500 lux at 12:00 h. The day/night temperatures were maintained at 35/25 ± 2 °C, and the day/night air humidity levels were kept at 60/65 ± 5%.

The experiment on intermittent water supply included the following five treatment conditions:

(1) Control group - T1: This group was continuously watered to maintain the soil at 75% field capacity (FC).

(2) Drought stress group - T2: This group experienced continuous watering to maintain the soil at 60% FC during the entire duration of the experiment (Tran et al., 2022).

(3) Stress 60-80 group - T3: This group received 60% FC from d 60 to 80 after sowing, during the period of fruit enlargement. For the remaining stages, the soil was maintained at 75% FC.

(4) Stress 60-harvest group - T5: This group received 60% FC from d 60 until harvest, covering both the period of fruit enlargement and lipid accumulation. For the remaining stages, the soil was maintained at 75% FC.

(5) Stress 80-harvest group - T4: This group received 60% FC from d 80 until harvest, during the period of lipid accumulation. For the remaining stages, the soil was maintained at 75% FC.

Field capacity was maintained using drip irrigation and a moisture sensor system (SSR1025, Handsontec, Singapore). When the moisture level dropped below the set value, the automatic pumping system supplied water to reach the desired value (Tran et al., 2022). Water use efficiency (WUE) was calculated using the formula $WUE (kg m^{-3}) = Y/ET$, where Y represents peanut yield (g plant⁻¹), and ET

represents water consumption ($L\ plant^{-1}$) (Zhang et al., 2021). The development of peanuts, their productivity, and water consumption were closely monitored throughout the experiment.

Determination of chlorophyll and carotenoid

The chlorophyll and carotenoid content of the leaves at the 90-d time point was assessed following a method described by Lichtenthaler (1987). Leaf samples were ground using 95% ethanol, and the resulting extract was centrifuged. Absorbance measurements were taken at wavelengths of 664, 648, and 470 nm using a spectrophotometer. The pigment content was calculated using the following formulae:

$$\begin{aligned} \text{Chlorophyll } a &= 13.36 \times OD_{664} \\ &\quad - 5.19 \times OD_{648} \\ \text{Chlorophyll } b &= 27.43 \times OD_{648} \\ &\quad - 8.12 \times OD_{664} \\ \text{Carotenoid} &= \frac{(1000 \times OD_{470} - 2.13 \times Chl\ a - 97.64 \times Chl\ b)}{209} \end{aligned}$$

Determination of respiration and photosynthesis rate

The respiration and photosynthesis rates of the leaves at the 90-d time point were assessed using a CO_2 sensor (EA80, Extech, USA). The measurement of respiration intensity was conducted under dark conditions, whereas the measurement of photosynthetic intensity was performed under a light intensity of 10,000 lux (Tran, 2022).

Determination of soluble carbohydrate, starch, soluble protein, lipid, and proline

Samples were extracted with ethanol to obtain a supernatant, which was subsequently mixed with phenol and sulfuric acid. The resulting mixture was compared to a sucrose standard curve at 540 nm to determine soluble carbohydrate content (Coombs et al., 1985). The residue was then hydrolyzed with HCl and $HClO_4$, yielding a supernatant that was mixed with DNS reagent, heated, and compared to a glucose standard curve at 490 nm to determine starch content (Miller et al., 1972).

For protein quantification, samples were ground in a buffer containing Tris (0.1 M) and EDTA (1 mM, pH 7.8). After centrifugation, the supernatant was used for protein analysis by mixing it with Bradford reagent and measuring absorbance at 595 nm (Bradford, 1976).

The oil content of the seeds was extracted using a modified method based on Shiva et al. (2018). Ground seeds were mixed with 2-propanol

containing 0.01% butylated hydroxytoluene. After heating and cooling, a mixture of chloroform, methanol, and water was added. The samples were incubated, shaken, and centrifuged, and the lipid content was determined by weighing the residue after solvent evaporation.

Proline content was quantified spectrophotometrically following extraction with 95% ethanol. The samples were centrifuged at 5000 g for 10 minutes to obtain the supernatant. One milliliter of this supernatant was mixed with 2 mL of acid ninhydrin in a test tube and heated at 100°C for 60 minutes. The absorbance of the resulting solution was measured at 520 nm, with proline content determined using L-proline as a standard (Paquin and Lechasseur, 1979).

Statistical analysis

The experimental procedures were conducted in three independent repetitions, with each repetition consisting of 30 pots, and one plant was grown in each pot. A randomized block design was used for the experiment, and a valid analysis of variance (ANOVA) was performed on the collected data. Mean separation was conducted using Duncan's Multiple Range Test, with a significance level of 5%. The results were presented as mean values accompanied by their corresponding standard deviations.

Results

Effect of intermittent water supply on growth and seed quality of peanuts under drought stress

At harvest, the data reveal the impact of intermittent water supply on peanut growth and yield in comparison to drought stress (Fig. 1). The control treatment exhibited the highest values for pod number, pod weight, seed number, and seed weight. In contrast, drought stress led to significant reductions in all yield parameters, with pod number decreasing from 13.80 to 2.80 pods per plant, seed number dropping from 26.80 to 5.60 seeds per pod, pod weight decreasing from 2.30 to 2.12 g, and seed weight declining from 0.63 to 0.49 g.

Among the intermittent water supply treatments, the 80-harvest treatment demonstrated the most significant improvements in pod number, seed number, pod weight, seed weight, and seed yield compared to the continuous drought stress treatment, with increases of 336%, 335%, 7.1%, 26.5%, and 2173%, respectively. Additionally, the 80-harvest treatment produced pod and seed weight values that were closer to those of the control treatment (Table 1).



Fig. 1. Effect of intermittent water supply on peanut yield under drought stress at harvest time. Scale bar = 5 cm.

Table 1. Effect of intermittent water supply on yield and water use efficiency under drought stress.

Treatment	T1	T2	T3	T4	T5
Pod number plant ⁻¹	13.80 ± 0.84 ^a	2.80 ± 0.45 ^d	9.40 ± 1.34 ^c	5.00 ± 0.71 ^d	12.20 ± 0.84 ^b
Pod weight (g)	2.30 ± 0.05 ^a	2.12 ± 0.02 ^c	2.19 ± 0.01 ^b	2.14 ± 0.01 ^c	2.27 ± 0.04 ^a
Seed number pod ⁻¹	26.80 ± 1.10 ^a	5.60 ± 0.89 ^d	18.80 ± 2.68 ^c	10.00 ± 1.41 ^d	24.40 ± 1.67 ^b
Seed weight (g)	0.63 ± 0.01 ^a	0.49 ± 0.05 ^c	0.61 ± 0.01 ^a	0.55 ± 0.01 ^b	0.62 ± 0.01 ^a
Seed yield (g plant ⁻¹),	232.98 ± 13.72 ^a	7.68 ± 0.20 ^d	157.79 ± 5.67 ^b	27.53 ± 0.53 ^c	174.56 ± 7.65 ^b
ET (L plant ⁻¹)	117.67 ± 5.73 ^a	93.65 ± 2.07 ^d	102.34 ± 4.12 ^b	99.67 ± 1.63 ^c	103.72 ± 3.51 ^b
WUE (kg m ⁻³)	1.98 ± 0.06 ^a	0.08 ± 0.01 ^d	1.06 ± 0.03 ^b	0.27 ± 0.02 ^c	1.77 ± 0.04 ^a

Values with different letters in a row indicate significant differences, $P = 0.05$ (Duncan's test).

Analysis of water use efficiency indicated that the 80-harvest treatment reduced water consumption (from 117.67 to 103.72 L per plant) while maintaining a comparable water use efficiency to the control treatment, ranging from approximately 1.77 to 1.98 kg m³.

In terms of seed quality, the drought stress group exhibited a higher lipid content (16.75%) but a lower starch content compared to the control group (33.41%). All intermittent water supply treatments displayed similar lipid contents, with

the 60-harvest stress treatment showing approximately a 20% increase in lipid content relative to the control. Notably, the 80-harvest stress treatment had the highest starch and soluble sugar content among the intermittent water supply treatments.

Protein content across all treatments was comparable, with no significant differences observed between the intermittent water supply treatments and the continuous drought stress treatment (Table 2).

Table 2. Effect of intermittent water supply on seed quality under drought stress at harvest time.

Treatment	Content in seed (mg g ⁻¹ DW)			
	Lipid	Starch	Soluble sugar	Protein
T1	459.72 ± 6.67 ^b	64.05 ± 4.48 ^a	43.69 ± 1.28 ^c	268.68 ± 19.53 ^b
T2	552.19 ± 38.06 ^a	42.65 ± 2.91 ^d	52.62 ± 1.18 ^b	309.24 ± 6.39 ^a
T3	552.59 ± 25.40 ^a	54.63 ± 1.48 ^b	56.03 ± 1.34 ^a	312.39 ± 17.95 ^a
T4	568.86 ± 32.87 ^a	48.76 ± 4.33 ^c	51.84 ± 1.49 ^b	309.33 ± 7.70 ^a
T5	535.06 ± 23.40 ^a	60.37 ± 1.37 ^a	55.37 ± 1.41 ^a	314.98 ± 13.21 ^a

Values with different letters in a column indicate significant differences, $P = 0.05$ (Duncan's test).

Changes in chlorophyll and carotenoid

Under drought stress conditions, chlorophyll a and chlorophyll b levels were reduced compared to the control group, while carotenoid content increased. Among the intermittent water supply treatments, the 60-80 stress treatment notably

improved chlorophyll a content, bringing it to levels comparable to the control. Additionally, all intermittent water supply treatments increased chlorophyll b content relative to continuous drought stress. However, these treatments resulted in a decrease in carotenoid content compared to drought stress conditions (Fig. 2).

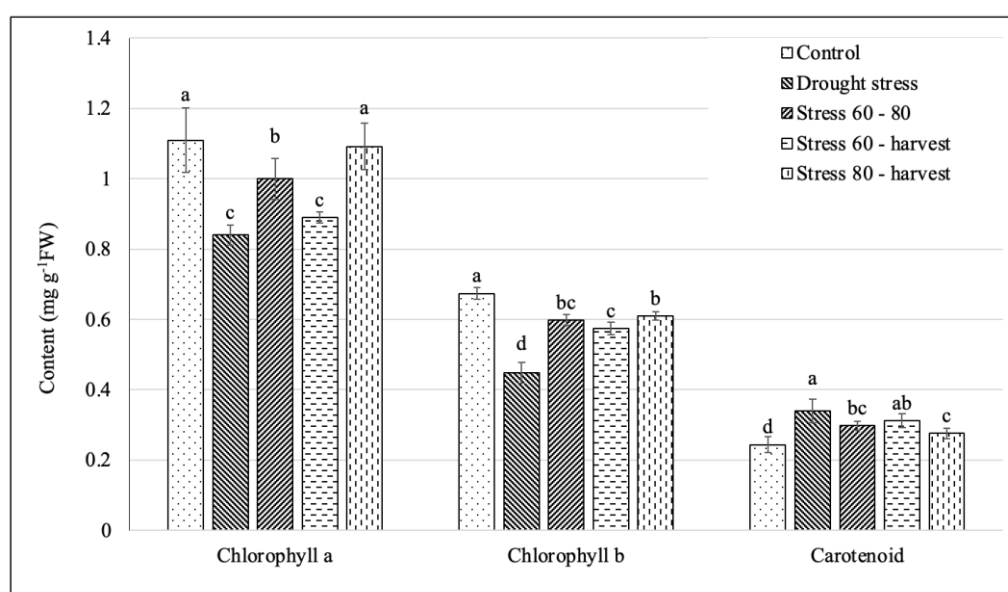


Fig. 2. Effect of intermittent water supply on chlorophyll and carotenoid content of leaves after 90 d under drought stress. According to the Duncan's test, there were differences in values for the letters in the columns ($P = 0.05$).

Changes in respiration and photosynthesis rate

Figure 3 illustrates the impact of intermittent water supply on leaf respiration and photosynthesis rates after 90 days. The continuous drought stress treatment resulted in a one-third reduction in respiration rate and a one-fourth reduction in photosynthesis rate compared

to the control. Among the intermittent water supply treatments, the 60-80 stress and 80-harvest stress treatments showed an increase in both respiration and photosynthesis rates relative to continuous drought stress. Notably, applying intermittent water supply from day 80 until harvest significantly improved respiration rates, reaching levels comparable to the control treatment.

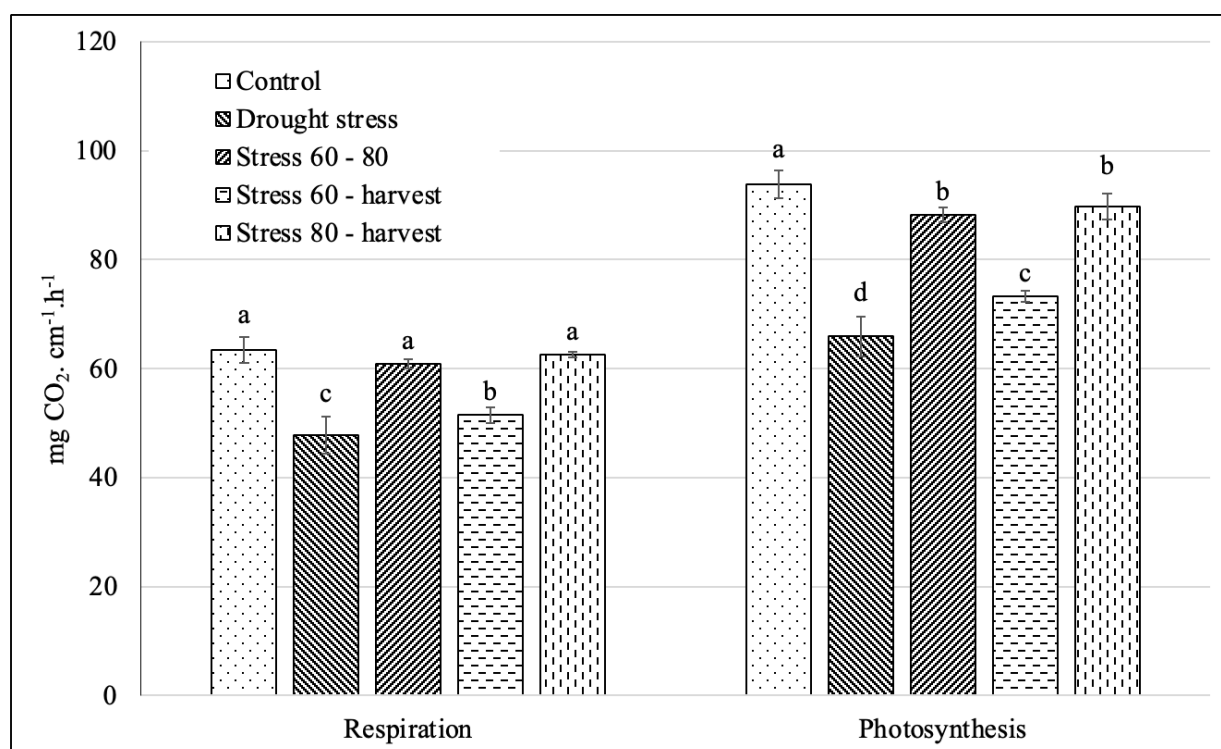


Fig. 3. Effect of intermittent water supply on respiration and photosynthesis rate of leaves after 90 d under drought stress. According to Duncan's test, there were differences in values for the letters in the columns ($P = 0.05$).

Changes in soluble sugar, starch, protein, and proline content

The impact of intermittent water supply on the starch, soluble sugar, protein, and proline content of leaves after 90 d is depicted in Figure 4. Continuous drought stress led to a reduction in starch content by approximately 40%. However, it resulted in a significant increase in the levels of total soluble sugars, proteins, and proline. Notably, the proline levels showed an eight-fold increase compared to the control group. Conversely, applying intermittent water supply treatments from d 80 until harvest helped elevate the starch content to approximately 45 mg g⁻¹. Furthermore, these treatments also contributed to comparable levels of soluble sugars, proteins, and proline to those observed in the control group, with levels of approximately 27, 15, and 0.3

mg g⁻¹, respectively.

Discussion

The role of intermittent water supply (IWS) in agriculture is increasingly important, especially in regions prone to water scarcity or drought. These strategies enable farmers to manage water resources more efficiently, ensuring crops receive adequate moisture for growth and development while simultaneously reducing water consumption (Bodner et al., 2015). A study on soybeans found that IWS significantly increased seed yield by 20% compared to the control group (Haryati et al., 2021). Moreover, intermittent water supply, applied as 2-hour watering sessions every three days, reduced the time to harvest by 10 days compared to watering every two days.

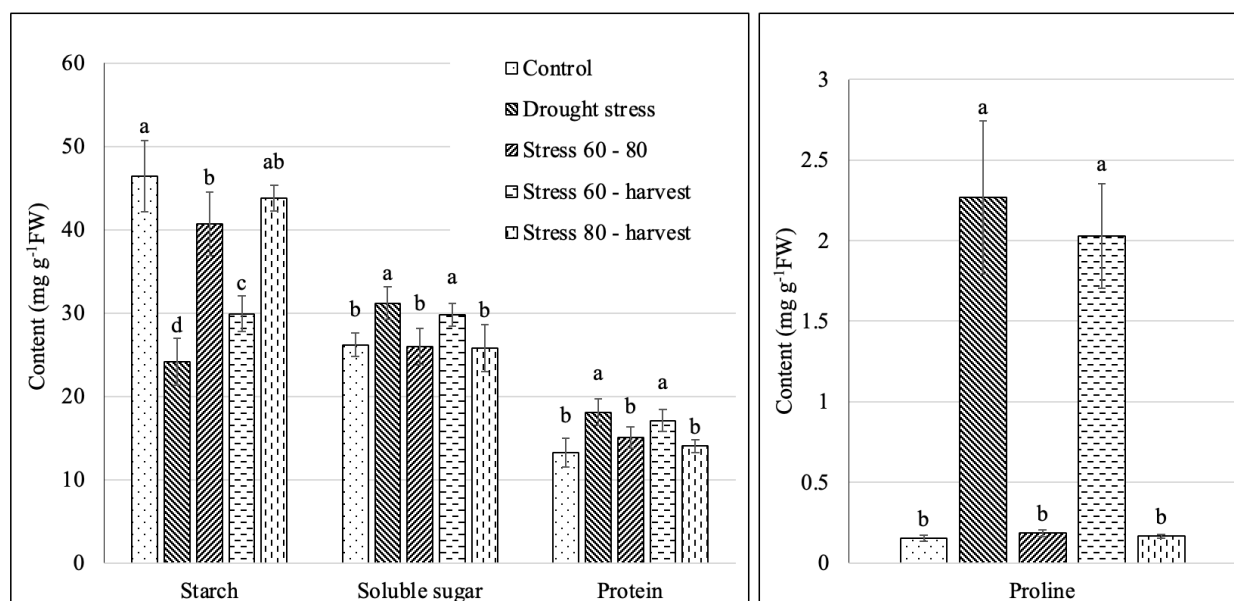


Fig. 4. Effect of intermittent water supply on starch, soluble sugar, protein, and proline content of leaves after 90 d under drought stress. According to Duncan's test, there were differences in values for the letters in the columns ($P = 0.05$).

In peanuts, the impact of IWS on growth parameters was evident (Fig. 1). Among the treatments, the stress 80-harvest regime showed the most promising results, improving pod number, seed number, and seed weight compared to continuous drought stress. This treatment also reduced water consumption (from 117.67 to 103.72 L per plant) while maintaining comparable water use efficiency to the control (Table 1). These findings suggest that IWS, particularly during specific growth stages, can lower irrigation demands and mitigate the adverse effects of drought stress on peanut yield. Furthermore, when water stress was applied from day 80 until harvest, at a time when plants were fully developed, growth remained largely unaffected, and there was a noticeable increase in seed lipid accumulation. This increase may be attributed to restricted irrigation during the period of lipid accumulation, which enhances the production of abscisic acid (ABA). Indeed, all water stress treatments resulted in higher lipid content in seeds (Table 2). According to Shi et al. (2021), ABA influences oleate-to-linoleate conversion in the endoplasmic reticulum membrane of oil palm by inducing higher expression levels of the FAD gene. Similar results have been observed in soybeans and Arabidopsis, where seed lipid content increases under ABA's influence during water stress, driven by the expression of AIL6 (INTEGUMENTA-like 6), a gene regulating FUSCA 3, which controls oil droplet accumulation (Dastmalchi, 2021; Liu et al., 2023).

In addition to increasing lipid content, the stress 80-harvest treatment also exhibited the highest starch and soluble sugar content among the IWS treatments (Table 2). This suggests that IWS can positively affect key seed quality attributes, such as lipid and starch content, which are crucial for both nutritional value and marketability.

IWS has also been shown to help maintain stable water levels within plants, preserving essential physiological and biochemical processes (Makonya et al., 2020). Studies on photosynthetic pigments, respiration, and photosynthesis have consistently demonstrated that IWS improves these parameters compared to continuous water stress (Figs. 2 and 3).

Additionally, analysis of soluble sugar, starch, protein, and proline content revealed that applying IWS from day 80 until harvest increased starch content while maintaining soluble sugars, proteins, and proline at levels similar to the control (Fig. 4). These findings indicate that IWS can regulate the accumulation of key biochemical compounds, enhancing plant stress tolerance and overall growth. Increased assimilation and metabolic capacity in peanuts during the vegetative growth phase under IWS has been documented (Collino et al., 2001).

In conclusion, intermittent water supply not only reduces water usage but also enhances crop yield, seed quality, and stress tolerance in peanuts, making it a promising irrigation strategy for water-limited conditions.

Conclusion

The findings suggested that intermittent water supply can mitigate the negative impacts of water deficit on peanut growth. The observed changes in yield parameters, seed quality, water-use-efficiency, and bio-physiological parameters showed that the intermittent treatment applied from d 80 until the harvest time improved the growth of peanuts effectively.

Conflict of Interest

The authors indicate no conflict of interest in this work.

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