



Comparative Evaluation of Relative Growth Rate and Net Assimilation Rate in Soybean (*Glycine max* L.) Varieties under Drought and Waterlogging Stress

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ABSTRACT

Water is crucial for plant physiological functions including germination, photosynthesis, nutrient uptake, and biomass accumulation. Under changing climate conditions, water stress - either drought or waterlogging - significantly impairs crop performance. This study investigated the effects of water stress on the growth parameters of soybean at the nursery unit of the Department of Botany, University of Ibadan, Oyo State, Nigeria. Using a completely randomized design (CRD), three soybean varieties (TGX 1987-10F, TGX 1835-10E, and TGX 1987-62F) were subjected to seven water regimes (waterlogged, watered once daily, watered twice a week, watered once a week, watered once in two weeks, watered once in three weeks, and watered once in four weeks) simulating drought and waterlogging conditions. Statistical analysis revealed that high moisture stress significantly suppressed biomass accumulation ($p \leq 0.05$), with maximum reductions of 76.91% and 76.89% in shoot and root fresh weights, and 83.83% and 61.46% in their respective dry weights. Interestingly, the waterlogging treatment promoted the highest root fresh weight across all studied varieties, despite simultaneously inducing substantial losses in other parameters; specifically, shoot fresh weight, root fresh weight, and root dry weight declined by 35.0%, 36.01%, and 46.33%, respectively, under anaerobic conditions. Furthermore, physiological efficiency was markedly compromised by hydrological extremes. Severe water deficit led to a drastic decline of 82.66% in the Relative Growth Rate (RGR) and 88.97% in the Net Assimilation Rate (NAR), whereas waterlogging resulted in comparatively moderate reductions of 17.06% and 49.27% for the same indices. Optimal growth was observed under daily and twice-weekly watering regimes. Drought stress had a more pronounced impact on soybean growth than waterlogging. The three soybean varieties exhibited similar responses to water stress, indicating minimal varietal influence. These findings highlight the critical role of water availability in optimizing soybean production under stress-prone environments.

Keywords: Soybean, Water stress, Relative growth rate (RGR), Net assimilation Rate (NAR), Climate stress adaptation

INTRODUCTION

Water plays a fundamental role in the survival of all living organisms. It participates directly in all cellular activities and molecular processes. Water is the fundamental driver of crop productivity, serving as a critical trigger

for seed germination, a primary substrate for photosynthetic carbon fixation, and the source of cellular turgor pressure essential for maintaining plant structural integrity and expansion (Kakati *et al.*, 2022). Its

availability, excess and scarcity are a strong factor that determines rates of growth in crop (Biswas *et al.*, 2025; Lawal *et al.*, 2024). The role of water in plant performance has garnered intensified research interest in recent years, primarily driven by the increasing frequency of extreme weather events and the urgent global demand for climate-resilient agriculture (Franco-Navarro *et al.*, 2025). This is critical given the escalating frequency and intensity of extreme water stress events ranging from flash flooding to prolonged drought, driven by accelerated global climate shifts. Recent assessments indicate that human-induced climate change has intensified the global water cycle, leading to more frequent and severe droughts and heavy precipitation events (Arias *et al.*, 2021).

Soybean (*Glycine max* L. Merrill) is globally recognized as a 'wonder crop' or 'miracle legume' because it serves as the cheapest, richest, and most accessible source of high-quality plant protein and oils (Ramlal *et al.*, 2023). Soybean serves as a highly versatile global commodity, providing essential nutrition for human consumption, serving as the primary protein source for livestock, and acting as a critical raw material for industrial biofuels and pharmaceutical applications (Delele, 2021). Global demand for soybean remains on a significant upward trajectory, driven by its versatile applications in the human food industry, its role as a primary protein source for livestock feed, and its expanding use in industrial biofuels and bio-based products (Ramlal *et al.*, 2023). This demand translates to good returns that give farmers better income. It facilitates fixing soil nitrogen through nitrogen fixation, thereby enhancing soil fertility and this is an added advantage in African agricultural systems where soil degradation is common (Hamza *et al.*, 2024). Prolonged dry spell and low rainfall are prevalent in areas of soybean production. This results in low yield, reduced pod production, reduces photosynthetic rate and biomass subsequently reducing seed weight by 40% (Du *et al.*, 2020). Flooding also reduces soybean by up to 27% (Zhou *et al.*, 2021). Soybean can be subjected to excess

water due to excess rainfall. Waterlogging stress induces severe physiological disruptions in soybean, primarily by reducing chlorophyll content and photosynthetic rates, inhibiting symbiotic nitrogen fixation in root nodules, and impairing nutrient uptake, which collectively result in stunted growth and significant yield losses (Sathi *et al.*, 2022).

Growth data reflect biomass accumulation driven by photosynthesis. It indicates the rate at which plants convert light energy to chemical energy. Growth rates assist in farm resource management, yield optimization by making farmers informed about irrigation, fertilization and pest management decisions (Raza *et al.*, 2021) and assessment of the genetic potential of plants with a good growth rate is used in breeding. Quantitative growth parameters, such as Relative Growth Rate (RGR) and Net Assimilation Rate (NAR), are key functional traits that determine the biomass allocation strategy and overall ecological performance of crops under environmental constraints (Amjid and Üstün, 2025). Analyzing fresh weight and dry weight is crucial for evaluating water content, biomass accumulation, and overall plant productivity, all of which are vital for enhancing soybean yield. Valuable growth analyses include relative growth rate (RGR) and net assimilation rate (NAR). RGR indicates how efficiently a plant converts raw resources (such as light and nutrients) into biomass. A higher RGR indicates that a plant is more effective at utilizing available resources, which is vital for assessing productivity in agricultural systems and natural ecosystems (Lamont *et al.*, 2023). Net Assimilation Rate (NAR) serves as a critical physiological index of photosynthetic efficiency, integrating changes in biomass with leaf area expansion to accurately track crop growth trajectories (Amjid and Üstün, 2025). By quantifying the net gain in dry matter per unit of leaf surface area, NAR provides a robust measure of a plant's metabolic health and its capacity to convert solar energy into yield under variable environments (Wang *et al.*, 2022). Collectively, these factors help identify high-

performing genotypes, enhance stress tolerance, and inform better crop management practices to maximize soybean productivity. Plant biomass represents the total mass of living plant material. Plant biomass, in most cases, forms the foundation and basis of the food chain, and the herbivores feed on it. They also improve soil fertility and structure, thereby improving water retention and nutrient content, which improves the overall health of the ecosystem (Johnson and Sankararajan, 2025). Soybean biomass demand, especially in the form of soybean meal and silage, is important in the livestock industry. This demand is driven by the need for high-quality protein sources for animal feed, which supports livestock health and productivity (Parrini *et al.*, 2023). Soybeans can be significantly affected by varying water stress. The objective of the study is to evaluate the relationship between the growth

analyses of a soybean variety in response to varying water stress. We aimed to ascertain the variation in fresh and dry, relative growth rate and net assimilation rate under varying water stress.

MATERIALS AND METHODS

The Study Area

The study was carried out at the Screen House at Nursery Unit of the Department of Botany, University of Ibadan, Ibadan, Oyo State. It is located at Latitude 7.444° N and longitude: 3.895° E. It is in the forest zone of Nigeria with two distinct season which are the wet season from March to October and dry season from November to February influenced in late December to early January by the dusty Harmattan winds. The mean daily temperature is 26.5°C, which can rise up to 34°C in March (Sule *et al.*, 2023).

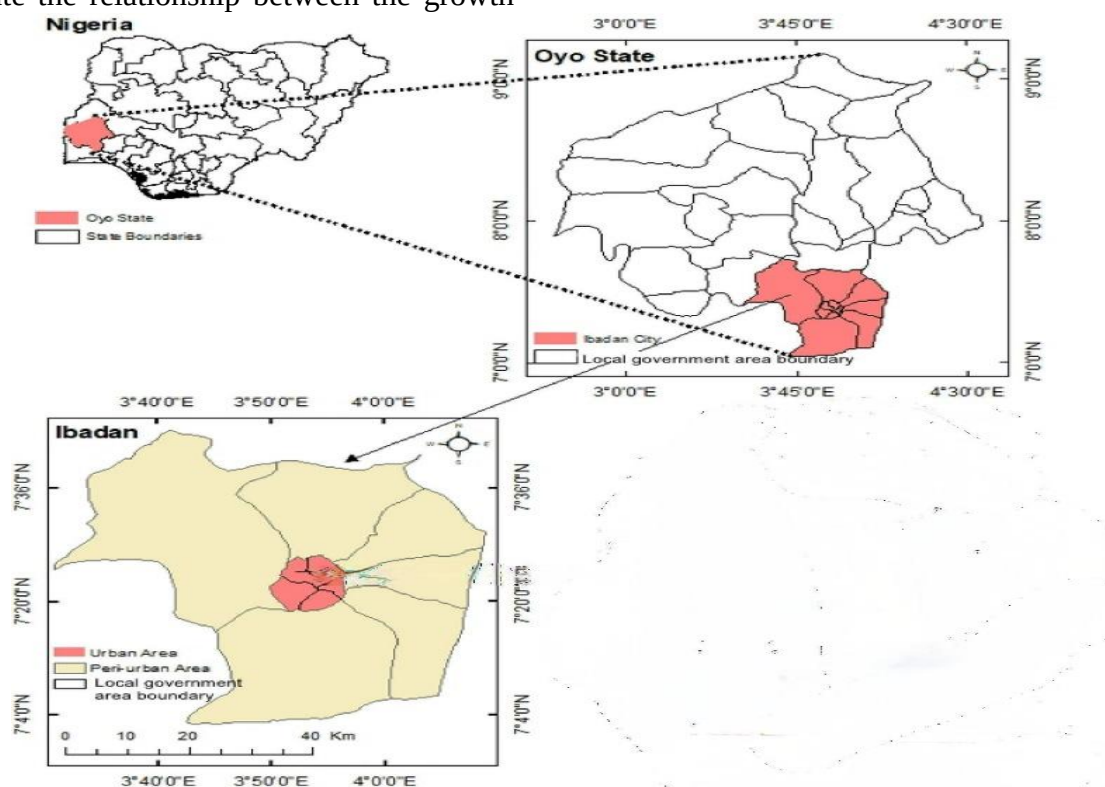


Figure 1: A Map of Nigeria and Oyo State showing Ibadan Metropolis
Source: Olokeogun and Kumar (2022)

Experimental Design Layout and Treatment Allocation

Polythene pots were filled with loamy soil obtained from the top soil in the nursery. The experiment was a 3 x 7 factorial design laid in a completely randomized design (CRD). Three varieties were obtained from International Institute for Tropical Agriculture (IITA), Ibadan and are as follows: Variety A - TGX 1987-10F (broad-spectrum rhizobia compatibility), Variety B - TGX 1835-10E (disease tolerant), Variety C - TGX 1987-62F (high yielding), to validate our evaluation whether effects water stress is consistent across the species or specific to a single genotype. The soil was watered till field capacity, and soybean seed was allowed to germinate for two weeks before imposition of water stress. The germinated seedlings were then subjected to seven water stress treatments as follows: Wat - Waterlogged treatment; 1dy - Once daily; 3dy - Twice a week; 1wk - Once a week; 2wk - Once in two weeks; 3wk - Once in three weeks; and 4wk - Once in four weeks (Kekere *et al.*, 2024). The soybean seedlings were watered based on the watering regimes.

Data Collection

At the time of soybean harvest, selected plants were randomly and carefully uprooted for each treatment and variety, labelled in polythene bags to reduce transpiration and respiration to a minimum and carried to the laboratory. They were then washed with running tap water and blotted with paper to remove adhering dirt and water. The plants were then separated into shoot and root and weighed. The components were oven dried at 78°C for 2 days until a constant dry weight was observed and reweighed. Shoot Fresh Weight (SFW), Shoot Dry Weight (SDW), Root Fresh Weight (RFW) and Root Dry Weight (RDW) were determined by accumulating the fresh and dry weight of each portion of the plant. The relative growth rate (RGR); the mass increase per

unit time and mass of plant, and net assimilation rate (NAR); produced dry matter per unit leaf area per unit time, were calculated using the formula (Hunt *et al.*, 2002).

$$RGR = \frac{\ln W_2 - \ln W_1}{t_2 - t_1} \quad 1$$

Where: $\ln$ is the natural log, W_1 and W_2 refer to the dry weight of the plant at sampling times t_1 and t_2 and is expressed as g/g/day (Hunt *et al.*, 2002).

$$NAR = \frac{(W_2 - W_1)(\log A_2 - \log A_1)}{(t_2 - t_1)(A_2 - A_1)} \quad 2$$

Where: W_1 , A_1 and W_2 , A_2 refer to dry weight and leaf area of plant at sampling time t_1 and t_2 , respectively and is expressed as g/cm²/day (Hunt *et al.*, 2002).

Data Analysis

The collected growth data under different water stress treatments were analyzed using IBM SPSS Statistics 2021. Means of all were calculated for each water treatment and variety. Analysis of variance (ANOVA) was conducted to determine the effect of water stress on shoot and root biomass at a significance level of $p < 0.05$.

RESULTS AND DISCUSSION

Effects of Water Stress on Shoot and Root Fresh and Dry Weights:

Data presented in Table 1 indicate that biomass accumulation was significantly influenced by watering frequency. The highest shoot fresh weight (SFW) was achieved by Variety A (TGX 1987-10F) under the twice-weekly watering regime (3dy), yielding 7.62g, whereas the lowest record for the same variety occurred under the three-week interval (3wk), dropping to 1.58g. Peak values for shoot dry weight (SDW), as well as root fresh and dry weights (RFW and RDW), were consistently observed under daily (1dy) or twice-weekly (3dy) regimes (Table 1). These optimal results were followed in descending order by the waterlogged (Wat) and once-per-week (1wk) treatments. A comparative analysis across

all varieties revealed a severe growth inhibition under moisture deficit; specifically, the transition from twice-weekly watering to a three-week deficit resulted in mean reductions of 76.91% in SFW, 76.89% in RFW, 83.83% in SDW, and 61.46% in RDW. In contrast, the waterlogging treatment induced a different physiological response, characterized by average reductions of 35.0% in SFW, 36.01% in SDW, and 46.33% in RDW. Notably, despite these declines in dry matter, the waterlogging treatment facilitated the highest root fresh weights across all genotypes studied. Analysis of variance (ANOVA) confirmed that the watering regime exerted a statistically significant effect on all evaluated biomass parameters ($p \leq 0.05$), as detailed in Table 2. Variety differences do not have a significant effect on the shoot and root fresh weight of soybean. Crop plants are frequently being subjected to either drought or waterlogging or both stresses during their growing period in regions (Bashir et al., 2021; Topali et al., 2024). Fresh and dry weights are major parameters reflecting the growth and development of a plant under varying environmental conditions. Waterlogging on roots slows down root elongation, root

hair formation and causes rotting and drying of roots and eventually death of the plant (Fujita et al., 2021). Exposure to water stress, particularly drought, resulted in a decrease in fresh and dry weight of the shoot and root of the crop plant. Soybean varieties' biomass decreases with increasing water stress. Drought conditions have been shown to reduce shoot biomass by approximately 33.53% and seed weight by 41.65% during the reproductive stages of growth. This decline is attributed to decreased leaf photosynthetic rates and impaired sucrose metabolism, which are crucial for energy supply during seed development (Du et al., 2020). Exposure to drought causes drastic changes in biomass production. Plants undergo cell modification under stress, which significantly affects the total biomass of the plant (Farooq et al., 2009). Plant weight reduction under varying water stress has been reported in barley (Rollins et al., 2013), apple (Bolat et al., 2014), sugar beet (Mohammadian et al., 2005), beans (Islam et al., 2021), rice (Saha et al., 2019) and soybean (Wang et al., 2022). Fresh and dry weights are major parameters reflecting the growth and development of a plant under varying environmental conditions.

Table 1: Effect of Water Stress on Shoot and Root Fresh and Dry Weights of Soybean

Trt	Shoot Fresh Weight (g)			Shoot Dry Weight (g)			Root Fresh Weight (g)			Root Dry Weight (g)		
	Var. A	Var. B	Var. C	Var. A	Var. B	Var. C	Var. A	Var. B	Var. C	Var. A	Var. B	Var. C
Wat	6.44	5.08	3.96	1.87	1.67	1.19	6.89	6.26	6.54	0.86	1.16	1.28
1dy	7.23	6.90	7.48	1.70	1.96	1.76	4.98	5.65	6.22	0.70	1.21	1.58
3dy	7.62	9.34	6.81	2.18	3.17	1.69	4.42	4.43	4.91	1.21	1.45	1.29
1wk	6.05	6.37	5.84	1.76	1.85	1.91	2.65	3.21	3.13	0.72	1.35	1.10
2wk	3.29	2.44	3.51	1.04	1.06	1.28	1.75	1.31	1.70	0.59	0.56	0.65
3wk	1.58	1.89	1.83	0.67	0.74	0.65	1.09	1.23	1.26	0.51	0.50	0.52
4wk	2.63	1.66	2.00	0.68	0.43	0.53	1.62	0.84	1.38	0.58	0.34	0.37

Trt = Treatment, Wat - Waterlogging, 1dy - Daily watering, 3dy - Twice a week, 1wk - Once a week, 2wk - Once in two weeks, 3wk - Once in three weeks, 4wk - once in 4 weeks

Table 2: Analyses of Variance for Soybean Shoot and Root Fresh and Dry Weights

Effect	Df	Shoot Fresh Weight (g)	Shoot Dry Weight (g)	Root Fresh Weight (g)	Root Dry Weight (g)
W. Stress	6	8.54*	4.56*	8.47*	4.94*
Variety	2	0.37	0.43	0.11	0.67
W. stress x Variety	12	0.38	0.37	0.07	0.37

Key: * = $p \leq 0.05$

Effects of Water Stress on the Relative Growth Rate (RGR) and the Net Assimilation Rate (NAR) of Soybean

The effect of water stress on RGR as observed in Table 3, the highest values of RGR was recorded when planting soybean in daily watering treatment for varieties A (TGX 1987-10F) and C (TGX 1987-62F) with 0.0339g/g/day and 0.0427g/g/day, respectively, while 0.0395g/g/day stands for the highest RGR in variety B (TGX 1835-10E) under twice a week water application. The lowest ones: 0.0027g/g/day, 0.0100g/g/day and 0.080g/g/day were registered by planting soybean once in four weeks or once in three weeks among the varieties as shown in Table 3. An average percentage reduction of 82.66% was recorded across

all varieties in severe water deficit as against 17.06% reduction in waterlogging treatment.

Water stress on soybean showed a favourable effect on NAR in twice-a-week water application and daily water treatment, with net assimilation as high as 5.88g/cm²/day, 11.63g/cm²/day and 7.53g/cm²/day in the varieties, respectively, as shown in Table 3. In reverse, the lowest values of NAR of 1.05g/cm²/day, 0.66g/cm²/day and 0.72g/cm²/day were all achieved by planting soybean at watering under once in four weeks. This showed a 88.97% reduction in the net assimilation rate of severe moisture deficit while waterlogging reduction effect stands at 49.27%.

Table 3: Effects of Water Stress on Relative Growth Rate (RGR) and Net Assimilation Rate (NAR) of Soybean Varieties

Water Stress	Relative Growth Rate (g/g/day)			Net Assimilation Rate (g/cm ² /day)		
	Var. A	Var. B	Var. C	Var. A	Var. B	Var. C
Wat	0.0259	0.0311	0.0400	3.49	3.97	4.42
1dy	0.0339	0.0302	0.0427	3.55	4.66	7.53
3dy	0.0266	0.0395	0.0348	5.88	11.63	5.58
1wk	0.0293	0.0305	0.0394	3.95	5.81	4.16
2wk	0.0047	0.0246	0.0223	2.55	2.67	1.86
3wk	0.0088	0.0100	0.0080	1.45	1.34	1.26
4wk	0.0027	0.0104	0.0080	1.05	0.66	0.72

Wat - Waterlogging, 1dy - Daily watering, 3dy - Twice a week, 1wk - Once a week, 2wk - Once in two weeks, 3wk - Once in three weeks, 4wk - once in 4 weeks

Production of dry mass is directly related to the amount of water transpired, as there is a reduction in growth of plants by affecting either the efficiency with which photosynthates aid to new growth or the rate at which they are used in maintaining the existing dry matter (Koehler *et al.*, 2023). Drought stress alters dry matter partitioning patterns, favoring greater allocation to roots over shoots, while

promoting temporal and spatial adjustments in root distribution, quantity, and functional root length to enhance water foraging (Cai *et al.*, 2023) where higher root growth under water deficit conditions can increase drought tolerance in plants (Seleiman *et al.*, 2021).

Waterlogging causes CO₂ reduction uptake, which inhibits photosynthesis as the carbon assimilation is affected, thereby

negatively affecting the NAR (Sathi *et al.*, 2022). Soybean plants are sensitive to waterlogging at the vegetative and reproductive stages, which reduces their internal CO₂ concentration and stomatal conductance and reduces photosynthesis. Reduced photosynthesis leads to a reduction in NAR (Pan *et al.*, 2021). Under drought stress, NAR generally decreases due to impaired photosynthesis and stomatal limitations (Elferjani and Soolanayakanahally, 2018; Lupitu *et al.*, 2023), as observed in this study and crops like Cauliflower and cabbage (Lupitu *et al.*, 2023) and Corianda (Jamali, 2013). This is attributed to impaired carbon assimilation, thereby limiting photosynthesis and stomatal conductance, leading to reduced photosynthetic activity (Elferjani and Soolanayakanahally, 2018). NAR is a great determinant of RGR in crop plants, as NAR reduction under water-deficient conditions is a common response of crop plants.

The general trend of the soybean species' growth rates was to decline with time, though there was an increase in the relative growth rate with the inception of flowering and fruiting, indicating that there may be a demand for water with the inception of this developmental stage. This reduction in growth rate with plant age has been well documented and explained by ontogenetic changes, higher allocation to low-efficiency tissues, and self-shading (Shipley, 2006). However, experiments show RGR increasing with time or exhibiting complex patterns during specific periods (Rees *et al.*, 2010; Paine *et al.*, 2012; Pepe *et al.*, 2022), often tied to NAR shifts. In other experiments, the temporal changes in RGR have also been associated with variation in NAR (Li *et al.*, 2016).

CONCLUSION

Plant growth and productivity depend heavily on water availability, particularly in soybeans, a crop of immense nutritional,

economic, and ecological importance. The study examined the effects of water stress (drought and waterlogging) on soybean, with particular focus on their effects on growth parameters. The findings confirm that while all varieties studied, TGX 1987-10F, TGX 1835-10E, and TGX 1987-62F, possess inherent genetic advantages such as disease tolerance and high yield potential, their physiological performance remains highly sensitive to hydrological extremes. Statistical evidence shows that severe moisture deficit is more detrimental to biomass accumulation than waterlogging. Specifically, the drastic reductions in Relative Growth Rate (RGR) and Net Assimilation Rate (NAR) under drought (up to 88.97%) indicate a near-complete shutdown of photosynthetic activity, whereas the waterlogging treatment showed a more moderate physiological impairment. A unique anatomical response was observed under anaerobic conditions, where waterlogging stimulated the highest root fresh weight across all varieties, likely as a compensatory mechanism for impaired root respiration. From a management perspective, the results suggest that twice-weekly watering is the optimal regime for maximizing biomass accumulation in soybean. Given the high sensitivity of these varieties to prolonged moisture stress, it is recommended that irrigation strategies in drought-prone regions prioritize maintaining water availability during the critical flowering and fruiting stages to prevent the irreversible decline in growth indices observed in this study. Understanding how drought and waterlogging affect key physiological processes like RGR and NAR is vital for developing effective strategies to improve crop resilience against environmental challenges posed by climate change. Data highlight the need for developing resilient soybean varieties capable of maintaining optimal growth under adverse water stress

and possibly other environmental stress. Further study is necessary to identify specific mechanism of underlying stress response which will enable stable yields across varying climatic stress condition.

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