MORPHOLOGICAL RESPONSE OF *Amaranthus hybridus* UNDER DROUGHT STRESS AMERIATED WITH PLANT GROWTH REGULATORS

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OCTOBER, 2025

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A PROJECT SUBMITTED TO THE DEPARTMENT OF SCIENCE LABORATORY TECHNOLOGY, UNIVERSITY OF BENIN, BENIN CITY, IN PARTIAL FUFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF BACHELOR OF SCIENCE DEGREE (B.Sc Hons) IN SCIENCE LABORATORY TECHNOLOGY (BIOTECHNOLOGY TECHNIQUES)

OCTOBER, 2025

CERTIFICATION

This is to certify that the research work titled MORPHOLOGICAL RESPONSE OF *Amaranthus hybridus* UNDER DROUGHT STRESS AMERIATED WITH PLANT GROWTH REGULATORS was carried out by **Blessing Osariemen UGBO** (Miss) with Matriculation number **LSC2007357** in the Department of Science Laboratory Technology (Biotechnology Techniques), Faculty of Life Sciences, University of Benin, Edo State, Nigeria.

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DEDICATION

This project is dedicated to my Alpha and Omega, my provider, my protector, God Almighty for giving me strength and protecting me throughout the process of this project work.

ACKNOWLEDGEMENTS

My profound gratitude goes to God Almighty for giving me the strength to finish this project work and his divine empowerment to complete my first degree. Special thanks to my mom, Mrs Evelyn Ugbo, for all her support and the love during my course of study, I love and honour you. My sincere gratitude goes to my project supervisior, Mrs B.O. Ogbeide for her guidance throughout the process of this project work.

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ABSTRACT

Amaranthus hybridus , a significant crop in arid and semi-arid regions, faces substantial challenges due to drought stress. This study investigates the morphological responses of A. *hybridus* to water-limited conditions. The research involved controlled experiments where plants were subjected to different treatments (40mg/l, 200mg/l, 360mg/l, 17.61mg/l, 88.06mg/l, 176.12mg/l) and growth regulators (SA and AA). Morphological traits such as plant height, stem girth, root length, and weight of seed, number of leaves were meticulously measured and analyzed. The results revealed significant increase in plant height, number of leaves and weight of seed under drought stress, indicating a strategy to minimize water loss. In contrast, root development showed an increase, suggesting an adaptive response to enhance water uptake from deeper soil layers. These morphological adaptations demonstrate the plant's resilience and ability to survive in drought environments. The findings underscore the importance of understanding these responses for improving drought tolerance in A. *hybridus* , thereby enhancing its productivity in water-scarce regions."

CHAPTER ONE

INTRODUCTION

1.1 Background of the study

Hidden hunger, a subtle form of undernutrition, weakens the immune system, leaving children and the elderly more vulnerable to various diseases (Jimoh et al., 2018). In Sub-Saharan Africa, deficiencies in essential nutrients manifest through conditions such as underweight, overweight, wasting, stunting, and other metabolic dysfunctions (Webb et al., 2018). To address these nutritional challenges, both developing and developed nations have implemented numerous interventions, including continuous food supplementation programs for low-income communities, improved maternal and child healthcare access, and agricultural and social incentives aimed at increasing food production (Webb et al., 2018). Despite these comprehensive strategies, the number of households struggling with poor dietary intake continues to grow.

Ironically, many of the populations most affected by nutritional deficiencies live in areas rich in a wide variety of indigenous vegetables (Mavengahama et al., 2013). Wild vegetables, which serve as significant sources of essential nutrients, hold great potential for combating global nutrient deficiencies (Flyman et al., 2006). These plants contain both macro- and micronutrients vital for cellular functions and tissue repair. Leafy vegetables, in particular, help counter oxidative stress by neutralizing free radicals and related biomolecules (Sakar et al., 2018). However, limited utilization of these vegetables has contributed to widespread metabolic imbalances, including malnutrition, stunting, and undernutrition (Webb et al., 2018).

Among these nutrient-rich vegetables, *Amaranthus* species stand out as some of the oldest cultivated plants. Globally, amaranths have been used as grain crops, leafy vegetables, ornamental plants, dyes, and even weeds, thriving across tropical, subtropical, and temperate zones (Jimoh et al., 2018). The

Amaranthus genus comprises about 74 annual species, displaying significant morphological diversity, with both monoecious and dioecious reproductive systems (Waselkov et al., 2018). These plants are promising sources of plant-based proteins, high-quality nutrients, unsaturated fatty acids, and other essential organic minerals found in their leaves, seeds, and roots (Jimoh et al., 2020).

Amaranthus species are well adapted to harsh environmental conditions due to their C_4 photosynthetic pathway, which enhances efficient food production even under stress (Rastogi et al., 2013). They possess physiological and genetic traits that promote resilience against pathogens, easy cultivation, and significant phenotypic plasticity (Jimoh et al., 2020). Numerous studies have highlighted the diverse bioactive compounds present in *Amaranthus* species, including phenolic compounds, lectins, anthocyanins, flavonoids, and antioxidant nutrients. These compounds act as free radical scavengers, protecting biological systems from oxidative damage (Young et al., 2001).

1.2 Justification of the Study

Amaranthus hybridus is distinguished by its outstanding nutritional composition. The plant contains high-quality protein that provides all essential amino acids, particularly lysine, which is typically deficient in staple grains such as wheat and maize (Jimoh *et a*l., 2018) This makes A. *hybridus* a complete and valuable protein source essential for human nutrition. In addition, it is rich in vital vitamins—including vitamin A and several B-complex vitamins—and minerals such as iron, calcium, and magnesium, which are indispensable for numerous physiological processes (Jimoh *et al.*, 2019). Owing to this nutrient density, A. *hybridus* represents an important dietary resource, particularly in areas where food diversity is limited and malnutrition remains prevalent. In the context of global climate change, resilient crops capable of withstanding environmental stress are increasingly important. A. *hybridus* demonstrates exceptional tolerance to drought and can thrive with minimal water input (Oni *et al*., 2011).

It also adapts well to various soil types, including marginal and saline soils, providing a major advantage in regions affected by soil degradation and erratic rainfall. Understanding the mechanisms underlying this resilience offers valuable insights for future crop-improvement programs aimed at developing varieties with enhanced stress tolerance and productivity (Eleazu *et al*., 2013).

Furthermore, A. *hybridus* is an agriculturally versatile crop. It can be cultivated for multiple purposes—its leaves as a leafy vegetable, its seeds as a grain source, and its biomass as animal forage. Its relatively short growth cycle enables several harvests within one growing season, increasing yield potential and food availability. This adaptability makes the crop suitable for both smallholder farms and commercial agricultural systems. The plant's low input requirements and environmental tolerance also make it a promising candidate for sustainable agriculture in drought-prone regions (Adeyemi *et al*., 2024).

Economically, cultivating A. *hybridus* can generate substantial income for farmers. Growing consumer awareness of its nutritional and health benefits has increased market demand both locally and internationally. The crop can be processed into diverse food products—such as flour, cereals, and snack items—creating opportunities for value-added production and agro-enterprise development. Research into optimal cultivation methods, post-harvest processing, and market strategies can help maximize profitability and enhance livelihoods for farming communities (Akindahusi *et al*., 2005).

From a conservation perspective, A. *hybridus* belongs to the Amaranthaceae family, which encompasses a broad range of cultivated and wild species. Studying the genetic diversity within A. *hybridus* and its wild relatives is crucial for safeguarding valuable traits for future breeding and ensuring the conservation of this important plant group. While A. *hybridus* has received growing scientific attention, knowledge gaps still exist, particularly concerning yield improvement, nutritional enhancement, and resistance to pests and diseases. Additional investigations are needed to identify the best growth conditions, effective processing methods, and sustainable market frameworks for this crop.By addressing these research gaps, scholars can contribute to the long-term productivity and utilization of *Amaranthus hybridus* . Such studies not only promote sustainable agriculture but also improve food security, nutritional health, and economic resilience among farmers—especially those affected by drought and changing climate conditions (Murray *et al*., 2013).

1.3 Statement of the Problem

Amaranthus hybridus is a vital crop across many tropical and subtropical regions; however, its productivity is severely constrained by water scarcity. Drought stress adversely affects the plant's growth and yield by reducing biomass accumulation, altering its morphology, and impairing physiological processes. Despite the crop's growing agricultural relevance, scientific understanding of its specific morphological and physiological adjustments to drought stress remains limited.

The key challenge is to determine how A. *hybridus* adapts morphologically to water-limited environments and how these adaptations influence its overall performance and yield potential. Critical research questions include:

What morphological modifications—such as changes in leaf size, root architecture, and stem elongation—occur in A. *hybridus* under varying levels and durations of drought stress?

How do these morphological traits relate to physiological parameters such as water-use efficiency and photosynthetic rate? Are there observable variations in drought tolerance among different genotypes or cultivars, and which morphological features contribute to this variation? (Amusat et al., 2011).Addressing these questions will enhance understanding of the mechanisms that enable A.

hybridus to survive and perform under drought conditions, providing a foundation for improving crop management and resilience.

1.3.1 Natural Growth Regulators

Natural growth regulators are organic compounds produced by plants in response to environmental stimuli, particularly stress conditions. They help regulate growth, development, and adaptive responses. The primary natural regulators include:

1.Auxins – responsible for cell elongation, apical dominance, and root initiation.

2.Gibberellins – promote stem elongation, seed germination, and flowering.

3.Abscisic acid – functions as a stress hormone that induces stomatal closure and mediates drought responses.

1.3.2 Synthetic (Manufactured) Growth Regulators

Synthetic growth regulators are chemically formulated substances that imitate the functions of natural plant hormones. They are widely used to enhance crop performance, growth, and stress resistance. Examples include:

1 .Ethylene

2. Kinetin

3. Salicylic acid

4. 6-Benzyladenine (BA)

5 .2,4-Dichlorophenoxyacetic acid (2,4-D)

This study specifically focuses on the effects of salicylic acid and ascorbic acid as key regulators that influence A. *hybridus* responses to drought stress.

1.3.2.1 Salicylic Acid

Salicylic acid (SA) is a naturally occurring plant signaling compound with the molecular structure HOC_6H_4COOH. It plays a pivotal role in numerous physiological and biochemical processes, including growth regulation, nutrient absorption, biotic interactions, and defense against environmental stressors. SA enhances a plant's tolerance to drought by regulating stomatal behavior, activating antioxidant enzymes, and promoting systemic acquired resistance mechanisms that mitigate the adverse effects of stress (Amoanimaa-Dede et al., 2022).

1.3.2.2 Ascorbic Acid

Ascorbic acid (vitamin C) is a water-soluble antioxidant essential for various biological functions in plants and other organisms. With the chemical formula $C_6H_8O_6$, it acts as a potent electron donor capable of neutralizing free radicals and preventing oxidative damage In plants, ascorbic acid supports photosynthesis, growth, and cellular protection during drought conditions by maintaining redox balance and stabilizing physiological processes (Carr et al., 2017).

1.4 Aim of the Study

The overarching aim of this research is to evaluate the morphological responses of *Amaranthus hybridus* under drought stress ameriated with growth regulators and to determine how plant growth regulators influence the crop's capacity to tolerate and survive under water-limited conditions.

1.5 Objectives of the Study

1. To assess the role of plant growth regulators in improving the yield and productivity of A. *hybridus* under drought conditions, thereby supporting farmers' economic resilience.

2. To evaluate the effectiveness of selected plant regulators in mitigating drought stress in A. *hybridus* .

3 .To investigate the morphological adaptations of A. *hybridus* in response to drought stress.

4. To identify optimal plant growth regulator treatments for enhancing drought tolerance and to recommend practical solutions for improving A. *hybridus* cultivation in drought-affected regions.

1.6 Limitations of the Study

This study will employ controlled experimental conditions to simulate drought stress, incorporating detailed morphological observations, physiological measurements, and data analysis to quantify adaptive responses. While these experiments will provide valuable insights into drought-tolerance mechanisms, results may vary under field conditions due to environmental variability. Nevertheless, the findings are expected to contribute to the development of strategies that enhance the drought resilience and productivity of *Amaranthus hybridus* in water-deficient environments.

CHAPTER TWO

LITERATURE REVIEW

2.1 Literature Review

The genus *Amaranthus* belongs to the order Caryophyllales, within the family Amaranthaceae, subfamily Amaranthoideae (Montoya et al., 2015). Plants in this genus are herbaceous or shrubby and may be either annual or perennial. Their floral structures differ among species, with flowers containing either three or five sepals and stamens, while the pollen grains consistently exhibit a seven-pore (7-porate) structure throughout the family.

Species of *Amaranthus* possess concentric rings of vascular bundles and utilize the C_4 photosynthetic pathway, which enhances their carbon fixation efficiency, especially under high temperature and light intensity. The leaves, typically 6.5–15 cm long, are oval or elliptical in shape and arranged alternately or oppositely, depending on the species. Most leaves are simple, entire, and have smooth margins. The root system is characterized by a main taproot that extends deep into the soil, supported by a network of fibrous secondary roots that improve stability and nutrient absorption.

The inflorescence of *Amaranthus* takes the form of a large terminal or axillary panicle, which varies in size, color, and orientation among species. Flowers are radially symmetrical and may be bisexual or unisexual, with small, bristly perianths and pointed bracts. The genus contains both monoecious species (such as A. *hybridus*) and dioecious species (such as A. palmeri). The fruit is a capsule, known as a unilocular pixidio, which opens upon maturity to release its seeds (Keding et al., 2007).

Plate 2.1: *Amaranthus hybridus*

Source: Elias *et a*l., 2009

The capsule's operculum opens to release the seed, which is typically round, 1–1.5 mm in diameter, and varies in color, usually possessing a smooth and glossy surface. The crop matures approximately 200 days after planting, yielding between 1,000 and 3,000 seeds per gram (Keding et al., 2007).

Historically, *Amaranthus* originated in the Americas and is considered one of the world's oldest food crops, with archaeological evidence of its cultivation dating back to around 6700 BC. The genus comprises nearly 60 species, several of which are cultivated as leafy vegetables, grain crops, or ornamentals, while others exist as weeds.

According to the United Nations Development Programme (UNDP), Sub-Saharan Africa continues to face widespread food insecurity and inadequate health facilities, particularly in rural areas. One of the major challenges to food security in this region is the lack of recognition and utilization of indigenous plant species with high nutritional potential. In Nigeria and other tropical African nations where starchy staples dominate the diet, vegetables remain the most affordable and accessible sources of essential vitamins, minerals, and amino acids.

Despite their availability, the nutritional composition of commonly consumed leafy vegetables, including A. *hybridus* , remains under-researched. The scarcity of comprehensive nutritional data limits their integration into formal nutrition and agricultural policies. Detailed knowledge of their nutrient content would benefit food processors, pharmaceutical developers, and policy makers, and could support government and non-governmental initiatives aimed at combating malnutrition. These gaps have provided the foundation for this study—to evaluate the nutritional properties and potential of *Amaranthus hybridus* as a sustainable dietary resource.

2.3 Medicinal Benefits of *Amaranthus hybridus*

Amaranthus hybridus , commonly referred to as green amaranth or slender amaranth, is widely recognized for its nutritional and therapeutic properties. The plant is valued not only as a leafy vegetable but also for its potential in traditional and modern medicine due to its bioactive compounds (O'Brien, 2012).

Nutritional Value: The leaves and seeds of A. *hybridus* are highly nutritious, containing substantial amounts of vitamins such as A, C, K, and B-complex.

Minerals such as calcium, iron, magnesium, and potassium are also abundant, contributing to bone health, oxygen transport, enzymatic functions, and electrolyte balance. Additionally, the plant is a source of antioxidants that counteract oxidative stress (O'Brien, 2012).

1. Antioxidant Properties: The presence of flavonoids, polyphenols, and other antioxidants in A. *hybridus* enables it to scavenge free radicals, protecting cells from oxidative damage. This action can help mitigate the risk of chronic diseases such as cardiovascular disorders, cancer, and neurodegenerative conditions (O'Brien, 2012).
2. Anti-inflammatory Effects: Certain phytochemicals in A. *hybridus* have anti-inflammatory properties. These compounds may inhibit inflammatory pathways, reducing chronic inflammation linked to diseases like arthritis, metabolic syndrome, and gastrointestinal disorders (Oni et al., 2011).
3. Digestive Health: The plant is a rich source of dietary fiber, which enhances digestive health by promoting peristalsis, improving gut microbiota, and preventing constipation. Fiber also contributes to satiety and can aid in weight management (Oni et al., 2011).
4. Cholesterol Reduction: Studies have shown that consuming amaranth may help reduce low-density lipoprotein (LDL) cholesterol, thereby supporting cardiovascular health (O'Brien, 2012).
5. Blood Sugar Regulation: A. *hybridus* may have hypoglycemic effects due to bioactive compounds that enhance glucose uptake and improve insulin sensitivity, making it beneficial for diabetic individuals (Oni et al., 2011).
6. Bone Health: The high calcium and magnesium content supports bone density and reduces the risk of osteoporosis (O'Brien, 2012).

7. Wound Healing: Traditionally, A. *hybridus* has been applied in herbal remedies for wound care, leveraging its antibacterial and anti-inflammatory activities to accelerate tissue repair (Oni et al., 2011).
8. Gluten-Free: The plant is naturally gluten-free, making it suitable for individuals with celiac disease or gluten sensitivity (O'Brien, 2012).

2.4 Characteristics of *Amaranthus hybridus*

A. *hybridus* is an annual herbaceous plant that can grow several feet in height. Its leaves are typically diamond-shaped or ovate, while the plant produces dense clusters of small flowers. Known for prolific seed production, A. *hybridus* is highly adaptable, thriving in a range of soil types including disturbed soils. Both the leaves and seeds are edible, making it a valuable food and medicinal resource (Oni et al., 2011).

2.5 Taxonomic Hierarchy

The taxonomic classification of *Amaranthus hybridus* is as follows (Murray et al., 2001):

Kingdom: Plantae

Unranked: Angiosperms, Eudicots

Order: Caryophyllales

Family: Amaranthaceae

Genus: *Amaranthus*

Species: A. *hybridus*

2.6 Physical Characteristics

Seeds: The seeds are small (less than 1 mm), lens-shaped, and typically dark brown or black. They have a smooth, shiny surface and are produced in large quantities within inflorescences. Seeds serve as both the reproductive unit and a nutrient source (Agbo et al., 2013).

Leaves: Leaves are ovate to lanceolate with entire margins and are alternately arranged along the stem. Their color varies from green to reddish-purple depending on environmental conditions. Leaves are the primary photosynthetic organs, producing energy for plant growth (Agbo et al., 2012).

Flowers: Flowers are small, green or reddish, and lack petals. They are monoecious and wind-pollinated, arranged in dense terminal or axillary clusters (Amusat et al., 2018).

Roots: The taproot system anchors the plant and absorbs water and nutrients. Lateral roots enhance nutrient acquisition and contribute to overall plant stability (Kobore et al., 2018).

Stem: The stem is generally erect, sometimes slightly sprawling, and ranges in color from green to reddish. It supports leaves and flowers while transporting water and nutrients (Osaman et al., 2004).

2.7 Effects of Drought Stress on A. *hybridus*

Drought stress profoundly affects A. *hybridus* physiology and productivity.

Reduced soil moisture triggers stomatal closure, decreasing CO_2 uptake and photosynthetic efficiency. This limits biomass accumulation and leaf yield due to lowered chlorophyll content and reduced activity of enzymes such as PEP carboxylase (Adeyemi et al., 2024).

The plant attempts osmotic adjustment by accumulating solutes like proline, sugars, and ions to retain water, but prolonged drought overwhelms these mechanisms. Root growth is restricted, reducing the plant's capacity to access deeper water reserves (Kumar et al., 2025).

Oxidative stress is a significant consequence of drought, with reactive oxygen species (ROS) damaging cell membranes, proteins, and DNA. Antioxidant enzymes such as superoxide dismutase (SOD) and catalase (CAT) are activated, but under severe drought, these defenses are insufficient, leading to reduced photosynthesis, leaf area, and seed quality (Adeyemi et al., 2024).

Drought also affects nutritional composition. Leaf protein content decreases, carbohydrates may accumulate as an energy reserve, and heat-sensitive vitamins decline, reducing the plant's value as a nutrient-dense crop (Akin-Idowu et al., 2020).

Plant growth regulators (PGRs) such as salicylic acid (SA) and jasmonic acid can mitigate these effects by enhancing antioxidant activity, nutrient uptake, and photosynthetic efficiency, improving both yield and nutritional quality under water-limited conditions (Adeyemi et al., 2024; Kumar et al., 2025).

2.8 Soil Requirements

A. *hybridus* grows best in well-drained loamy or sandy-loam soils with a pH of 5.5–7.5. Organic matter (2–3%) enhances nutrient uptake and biomass production. Proper tillage (15–20 cm) improves aeration and supports taproot growth. Adding compost or manure (5–10 t/ha) improves fertility, particularly under drought, while PGRs like SA and ascorbic acid enhance nutrient assimilation and leaf quality (Adedibu et al., 2024).

2.9 Moisture and Temperature Requirements

Moisture: Optimal soil moisture is 60–70% of field capacity (20–30 mm per week during vegetative growth). The plant tolerates moderate drought due to its C4 metabolism and deep taproot but experiences 20–30% yield reduction under water stress (Adedibu et al., 2024). PGRs improve water use efficiency by 10–15%.

Temperature: Seeds germinate at 20–25°C, while vegetative growth is optimal at 25–35°C. A. *hybridus* is heat and drought tolerant, suitable for tropical and semi-arid regions below 800 meters altitude (Adeyemi et al., 2024; Kumar et al., 2025).

2.10 Plant Growth Regulators

PGRs are organic compounds that regulate plant growth and responses to environmental stress. Exogenous application can enhance plant growth, stress tolerance, and yield (Verma et al., 2016). Principal PGRs include:

1. Auxins: Promote cell elongation, root initiation, and apical dominance
2. Gibberellins: Stimulate stem elongation, flowering, and seed germination
3. Cytokinins: Enhance cell division, shoot proliferation, and delay senescence
4. Abscisic Acid: Mediates stress responses and stomatal closure
5. Ethylene: Regulates fruit ripening, senescence, and abscission (Osman et al., 2016)

2.11 Salicylic Acid as a Growth Regulator

SA is a naturally occurring plant hormone that regulates defense, growth, and stress tolerance. It enhances photosynthesis by increasing leaf expansion, chlorophyll content, and stomatal efficiency. SA also improves nutrient uptake, antioxidant activity (SOD, CAT, POD), and secondary metabolite production, which are important for medicinal and nutritional quality (Arun et al., 2024; Li, 2022).

Application:

Foliar spray: Optimal at 10 µM (~1.38 mg/L), improving growth, biomass, antioxidant enzyme activity, chlorophyll content, and nutrient assimilation (Arun et al., 2023).

Other methods, such as seed priming, soil drench, and root dipping, are well-studied in related *Amaranthus* species but limited in A. *hybridus* .

Functions of Salicylic Acid

1. **Systemic Acquired Resistance (SAR):**

   Salicylic acid (SA) serves as a crucial signaling molecule in SAR, a widespread defense mechanism that provides long-lasting protection against a variety of pathogens.

2. **Local Defense:**

   SA accumulates at infection sites, triggering localized defense mechanisms such as the hypersensitive response (HR). This leads to programmed cell death in infected cells, limiting the spread of pathogens.

3. **Stomatal Regulation:**

   SA can induce stomatal closure, which reduces water loss and prevents pathogen entry through the leaves.

4. **Thermotolerance:**

   SA contributes to heat stress adaptation by enhancing thermogenic responses, helping plants cope with high temperatures.

5. **Hormonal Interactions**:

   SA interacts with other plant hormones, including jasmonic acid and ethylene, to fine-tune defense responses.

Overall, SA is a vital component of plant immunity, enabling effective recognition and response to pathogen attacks (Eleksu et al., 2019).

2.12 Ascorbic Acid as a Growth Regulator

Ascorbic acid (vitamin C) is an essential compound in plants, playing multiple roles in growth, metabolism, and stress responses. Although studies specifically on *Amaranthus hybridus* are limited, research on related amaranth species shows its effectiveness.

For example, Sarker and Oba (2018) observed that drought stress increased the accumulation of vitamin C, phenolic compounds, and overall antioxidant capacity in amaranth leaves, suggesting that ascorbic acid is central to the plant's drought response. Exogenous application of ascorbic acid can complement this natural defense system. It has been shown to enhance the ascorbate–glutathione (AsA–GSH) cycle, strengthen antioxidant defenses, reduce oxidative damage, and improve growth under water-deficit conditions.

Practically, foliar application is the most effective delivery method. Recommended concentrations range from 100–200 mg·

Functions of Ascorbic Acid (Vitamin C) as a Growth Regulator

Ascorbic acid (AsA) is an essential compound in plants, playing multiple roles in growth, development, and stress adaptation. Although research on *Amaranthus hybridus* is limited, studies on related amaranth species indicate its significant role in improving plant resilience.

Stress Response Enhancement:

Drought stress often increases vitamin C accumulation in amaranth leaves, along with phenolic compounds and antioxidant activity. Exogenous application can complement these natural defenses, reducing oxidative damage caused by stress (Sarker & Oba, 2018).

Antioxidant Activity:

Ascorbic acid scavenges reactive oxygen species (ROS) such as hydrogen peroxide and superoxide, protecting cells from oxidative injury under environmental stresses like drought, heat, and UV radiation (Farooq et al., 2020).

Photosynthesis Support:

Application of AsA helps maintain chlorophyll content, improves photosynthetic efficiency, and supports leaf expansion, which collectively enhances growth even under water-limited conditions.

Osmotic and Metabolic Regulation:

AsA contributes to cellular osmotic adjustment and stabilizes metabolic processes, helping the plant maintain redox balance and nutrient uptake during drought stress. This supports overall growth and yield stability in A. *hybridus* (Younis et al., 2024).

Hormone and Developmental Roles:

Ascorbic acid influences plant growth by participating in hormone signaling pathways, regulating cell division, and supporting developmental transitions, including reproductive stages and senescence (Khan et al., 2011; Smirnoff, 2018).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Materials

Amaranthus hybridus seeds, top soil (0-10cm), nursery bags, and distilled water, hand trowel, shovel, sieve cloth, spray bottles, watering cans, foil paper, 5 litres, 6 kegs, bowl, salicylic acid solution, absorbic acid solution, masking tape, chloroform, acetic acid, mayer's reagent, olive oil, sulphuric acid, stirring bar, hot plate, acetic acid, weighing balance, hand gloves, detergent, beaker, micropipette, measuring cylinder, measuring tape, test tubes with a test tube rack.

3.2 Description of study area

This study was conducted at the Botanical Garden of Department of Plant Biology and Biotechnology, Faculty of Life Sciences, University of Benin, Benin City, Nigeria.

3.3 Source of planted seeds

The A. *hybridus seeds* were sourced from IAR&T (Institute of Agricultural Research and Technology) Ibadan, Nigeria).

3.3.1 Soils:

Top soil (0-10cm) were collected from the Department of Soil Science, Faculty of Agriculture, University of Benin, Benin City, Nigeria, in the morning (7:00am) and placed in a polythene sheets that were spread on an open surface and left in the sun until evening (5:00pm) for drying.

3.3.2 Source of plant growth regulator

Plant growth regulator were obtained from Pyrex Medical Laboratory opposite UBTH (University of Benin Teaching Hospital), Ugbowo, Benin City, Edo state, Nigeria.

3.3.3 Determination of water holding capacity of soil used

Soil was gotten from Department of soil science, Faculty of Agriculture, University of Benin. The soil`s water holding capacity was assessed in a laboratory setting using precise measuring scale, the weight of an empty 1000ml beaker was recorded as 555g. 1kg of soil was added to the beaker resulting in a total weight of 1555g. The weight of beaker was subtracted confirming the 1kg of the weight of the soil. Water was added to the soil until it was fully saturated and the mixture was stirred to ensure moisture distribution. The top of the beaker was covered with sieve cloth with multiple rubber bands to create a semipermeable seal. The beaker was converted and place in the sink to drain, allowing it to rest for 24hours. After this period, the weight of the remaining content was measured. The weight difference between the drained soil and the dry soil was recorded as 1211.9grams. The water holding capacity of the soil was calculated using this formula:

(VWC %) = Vw/Vt x 100

Vw = Mw

And

My = Mt - Ms

Where;

Mw = mass of water in grams

Mt = total mass of the container and wet soil in grams

Ms = total mass of the container and dry soil in grams

Vw = volume of water

Vt = total volume of saturated soil recorded to be 1211.9g

(VWC %) = water holding capacity

Mathematical calculations

Mw = Mt - Ms

Mw = 4112.1g - 1555g = 2557.1g

Where;

Vw = Mw

Vw = Mw = 2557.1g

(VWC%) = (Vw/Vt) x 100

= (2557.1g/1211.9g) x 100

= 210999 x 100

= 211mg/kg.

3.4 Preparation of soil bags for planting

A total of sixty-six (66) planting bags was prepared for the *Amaranthus hybridus* seeds using 7.5kg of soil from the sun dried soil collected from soil science Department, Faculty of Agriculture and were left for 2weeks to attenuate. Each bag was labelled according to the treatment it would receive.

The bag was labelled according to the specific treatments to which the *Amaranthus* seeds will be subjected.

It was twenty-two (22) nursery bags with three (3) replicates each including control which sum up to sixty-six (66) bags and they were watered regularly without the use of plant growth regulator to enable the plant grow to a mature stage where plant growth regulator can then be applied.

Twenty- seven (27) nursery bags was assigned to seeds treated with salicylic acid (SA), another twenty-seven (27) nursery bags was assigned to seeds treated with absorbic acid (AA). Nine (9) nursery bags was subjected to controlled water (drought stress) and Three (3) nursery bags served as control group.

3.5 Planting of *Amaranthus hybridus* seeds

To break seed dormancy, the seeds was presoaked in water for 2hours. Then the viable ones were planted on each bags by broadcasting method, ensuring they were evenly distributed across the different bags. This uniform distribution helped minimize overcrowding and reduced competition among seedlings for essential resources such as light, nutrients, and water, thereby promoting healthy and uniform growth.

3.6 Preparation of growth regulator solutions (Salicylic acid and Absorbic acid)

The value of treatment were gotten using this formula below

C1V1 = C2V2

Where; C1 = Initial concentration

C2 = Final concentration

V1 = Initial volume

V2 = Final volume

Stock solution of Salicylic acid available was 500ppm

Stock solution of Absorbic acid available was 1000ppm

Calculation for Salicylic Acid

For 5ppm treatment; C1V1 = C2V2

C1 = 500ppm

V1 = ?

C2 = 5ppm

V2 = 500ml

V1 = C2V2/C1

V1 = (5 x 500)/500

V1 = 5ml

For 15ppm treatment; C1V1 = C2V2

C1 = 500ppm

V1 = ?

C2 = 15ppm

V2 = 500ml

V1 = C2V2/C1

V1 = (15 x 500)/500

V1 = 15ml

For 45ppm treatment; C1V1 = C2V2

C1 = 500ppm

V1 = ?

C2 = 45ppm

V2 = 500ml

V1 = C2V2/C1

V1 = (45 x 500)/500

V1 = 45ml

Calculation for Absorbic Acid

For 5ppm treatment; C1V1 = C2V2

C1 = 1000ppm

V1 = ?

C2 = 5ppm

V2 = 1000ml

V1 = C2V2/C1

V1 = (5 x 1000)/1000

V1 = 5ml

For 15ppm treatment; C1V1 = C2V2

C1 = 1000ppm

V1 = ?

C2 = 15ppm

V2 = 1000ml

V1 = C2V2/C1

V1 = (15 x 1000)/1000

V1 = 15ml

For 45ppm treatment ; C1V1 = C2V2

C1 = 1000ppm

V1 = ?

C2 = 45ppm

V2 = 1000ml

V1 = C2V2/C1

V1 = (45 x 1000)/1000

V1 = 45ml

3.7 Germination and care

The seed was observed to germinate approximately 72hours after planting.

Each nursery bag was properly labeled according to its designated treatment group. During the first four weeks, all seedlings received equal and adequate watering to promote uniform early growth and establishment.

After this initial phase, drought stress treatments were imposed by adjusting irrigation based on specific percentages of the soil's total water-holding capacity—namely 5%, 15%, and 45%—depending on the treatment group. The control group, however, was provided with sufficient water to sustain normal growth.

Throughout the experimental period, seedlings were carefully monitored, and standard cultural practices were maintained to minimize external environmental influences. This approach ensured that any variations observed in plant growth and development could be accurately attributed to the effects of drought stress and the applied growth regulators.

3.7.1 Experimental Design

After the soil has been gathered in a plastic bags, small holes was made in the bags to enable aeration and the soil was slightly watered. To carry out experimentation on drought stress of *Amaranthus hybridus* plant, drought stress was instigate by regimented watering, then the *Amaranthus hybridus* was subjected different percentages of drought represented in 5%, 15%, 45%. To also test the effectiveness of growth regulators in different treatments was grouped into;

1. 40mg/l
2. 200mg/l
3. 360mg/l
4. 17.61mg/l

5. 88.06mg/l
6. 176.12mg/l

Table 3.1: Drought stress with treatment plot

Treatment with Salicylic acid

40mg/l	200mg/l	360mg/l
DSA_1, DSA_1, DSA_1	DSA_1, DSA_1, DSA_1	DSA_1, DSA_1, DSA_1
DSA_2, DSA_2, DSA_2	DSA_2, DSA_2, DSA_2	DSA_2, DSA_2, DSA_2
DSA_3, DSA_3, DSA_3	DSA_3, DSA_3, DSA_3	DSA_3, DSA_3, DSA_3

Treatment with Absorbic acid

17.61mg/l	88.06mg/l	176.12mg/l
DAA1, DAA1,DAA1	DAA1, DAA1,DAA1	DAA1, DAA1,DAA1
DAA2,DAA2, DAA2	DAA2,DAA2, DAA2	DAA2,DAA2, DAA2
DAA3, DAA3, DAA3	DAA3, DAA3, DAA3	DAA3, DAA3, DAA3

Table 3.2: Drought stress without treatment plot

5%	15%	45%
DS1, DS1, DS1	DS2, DS2,DS	DS3, DS3, DS3

Table 3.3: Control plot

Control plot
Control 1
Control 2
Control 3

Regimented water was calculated using the formula below:

% x water holding capacity (211mg/kg) + number of days

Where;

% = 5%, 15%, 45%

Water holding capacity = 211mg/kg

Number of days = watering was done 4 days in a week

The water was calculated for 1week before exposing it to plant growth regulator. Application of growth regulator through the use of spray bottles started after 21days and was applied 3 times a week at the early hours in the morning for 4weeks.

3.7.2. Measurement of plant height

A measuring tape was used to measure the height of the plants with different treatments.

3.7.3. Determination of leaf number per plant

This was done to each plant to different treatments by counting each leaves in the plants and recording it appropriately. This was done weekly.

3.7.4. Determination of leaf death

The leaves of the plant which has fallen in the bags was picked as leaf death and this was done 3 times a week.

3.7.5. Determination of chlorosis

A leaf colour chart ranging from dark green to light yellow was used to compare *Amaranthus hybridus* leaves to check for chlorosis.

3.7.6. Determination of weight of seed per plant

The seeds were weighed and recorded appropriately

3.7. 7 Determination of length of root per plant

The length of the roots were measured accurately using a ruler.

3.7.8. Determination of weight of root hairs per plants

The weight of root hairs was weighed with a weighing balance and recorded accurately.

3.7.9. Determination of stem girth per plant

The vernier caliper was used to measure the stem of each of the plant from top to middle to bottom.

CHAPTER FOUR

RESULTS

4.1 Results

The result obtained from the study are shown in table 4.1, 4.2, 4.3, 4.4, 4.5, 4.6 and alongside with diagrams which are plates 4.1, 4.2, 4.3, 4.4, 4.5. and a graphical representation of the impact the growth regulators (salicylic acid and absorbic acid) on *Amaranthus hybridus* plant shown in figure1, figure 2, figure3, figure 4, figure 5, and figure 6.

Table 4.1 shows the record of the height of *Amaranthus hybridus* plant between 4 to 6 weeks of being exposed to growth regulator (salicylic acid and absorbic acid) and regimented water.

Table 4.2 shows the record of the number of leaves per plants between 4 to 6 weeks of being exposed to growth regulators (salicylic acid and absorbic acid) and regimented water. Table 4.3 shows the record of stem girth per plant of *Amaranthus hybridus* for 6weeks of being exposed to growth regulators (salicylic acid and absorbic acid) and regimented water.

Table 4.4 shows the record of length of root of *Amaranthus hybridus* plant for 6weeks after being exposed to growth regulators (salicylic acid and absorbic acid) and regimented water.

Table 4.5 shows the record of weight of *Amaranthus hybridus* seeds for 6weeks after being exposed to growth regulators (salicylic acid and absorbic acid) and regimented water. Table 4.6 shows the record of colour chart showing the colours of *Amaranthus hybridus* of different treatments.

Table 4.1: Height of *Amaranthus hybridus* plants between 4 to 6 weeks of being exposed to growth regulators (SA and AA) and regimented water

Plant treatment conditions	Week 4	Week 5	Week 6
CONTROL	22	32	49
DS1 5%	20	32	70
DS2 15%	35	40	50
DS3 45%	19	23	59
DSA1 40mg/l	25	30	60
DSA1 200mg/l	37	41	70
DSA1 360mg/l	30	38	69
DAA1 17.61mg/l	36	40	65
DAA1 88.06gm/l	25	30	85
DAA1176.12mg/l	30	38	90
DSA2 40mg/l	18	25	68
DSA2 200mg/l	19	35	92
DSA2 360mg/l	20	30	50
DAA2 17.61mg/l	22	29	81
DAA2 88.06mg/l	32	40	45
DAA2 176.12mg/l	30	39	56
DSA3 40mg/l	17	26	49
DSA3 200mg/l	20	29	82
DSA3 360mg/l	22	38	100
DAA3 17.61mg/l	29.6	40.1	133
DAA3 88.06mg/l	25	37	110
DAA3 176.12mg/l	39	41	89

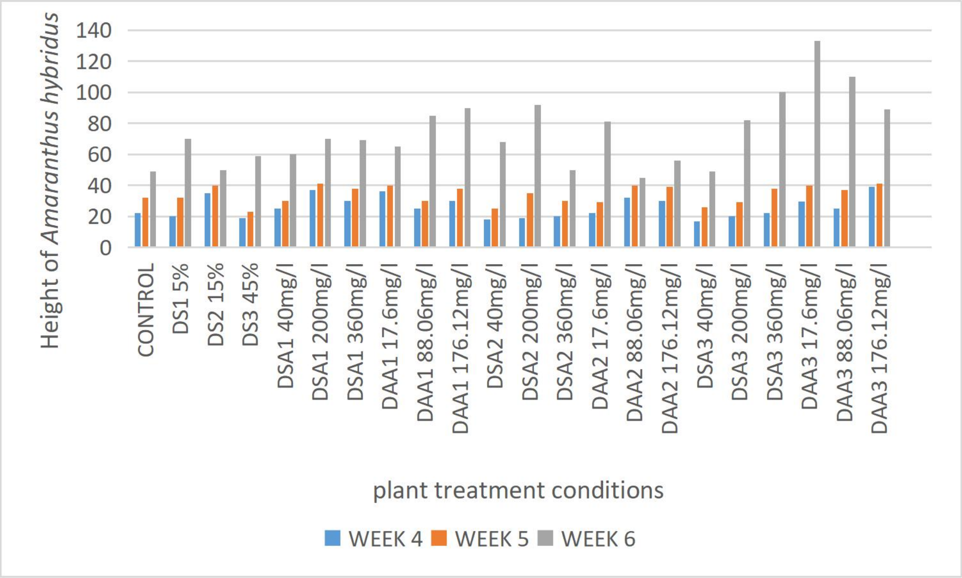


Figure 4.1: Graphical representation of height of *Amaranthus hybridus* plant between 4 to 6 weeks of being exposed to growth regulators (SA and AA) and regimented water

Note: The graph shows how the height of A. *hybridus* progressively increased from week 4 to week 6 for different plant treatments. DAA3 17.61mg/l had the highest height in week 6 followed by DAA3 88.06mg/l and DSA3 360mg/l . Then DAA2 88.06mg/l had the lowest height in week 6 followed by DSA3 40gm/l and Control.

Table 4.2: Number of leaves per plants of *Amaranthus hybridus* between week 4 to week 6 of being exposed to growth regulators (SA and AA) and regimented water

Plant treatment conditions	Week 4	Week 5	Week 6
Control	25	32	43
DS1 5%	19	77	95
DS2 15%	29	62	100
DS3 45%	19	23	58
DSA1 40mg/l	16	30	62
DSA1 200mg/l	13	41	89
DSA1 360mg/l	20	52	60
DAA1 17.61mg/l	33	31	61
DAA1 88.06gm/l	25	36	82
DAA1176.12mg/l	30	25	90
DSA2 40mg/l	18	30	100
DSA2 200mg/l	19	18	96
DSA2 360mg/l	22	19	59
DAA2 17.61mg/l	22	20	87
DAA2 88.06mg/l	15	36	40
DAA2 176.12mg/l	22	31	59
DSA3 40mg/l	17	62	215
DSA3 200mg/l	20	40	89
DSA3 360mg/l	13	69	100
DAA3 17.61mg/l	32	50	119
DAA3 88.06mg/l	25	80	110
DAA3 176.12mg/l	21	69	200

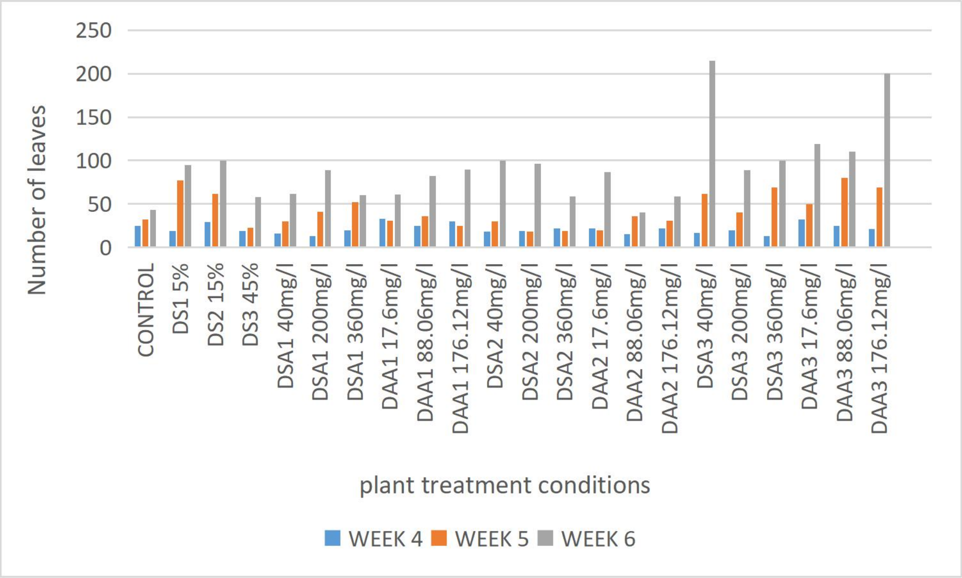


Figure 4.2: Graphical representation of number of leaves of A. *hybridus* plant between 4 to 6 weeks of being exposed to growth regulators (SA and AA) and regimented water

Note: week 4: blue, week 5: brown, week 6: ash

The graph shows how the number of leaves of A. *hybridus* progressively increased from week 4 to week 6 for different plant treatments. DSA3 40mg//l had the highest number of leaves in week 6 followed by DAA3 176.12mg/l and DAA3 17.61mg/l. Then DAA2 88.06mg/l had the lowest number of leaves in week 6 followed by DS3 45% and Control.

Table 4.3: Stem girth per plant of *Amaranthus hybridus* for 6weeks of being exposed to growth regulators (SA and AA) and regimented water

Plant treatment conditions	Top	Middle	Bottom
Control	0.2	0.3	0.3
DS1 5%	0.1	0.2	0.2
DS2 15%	0.1	0.2	0.3
DS3 45%	0.1	0.3	0.3
DSA1 40mg/l	0.3	0.2	0.3
DSA1 200mg/l	0.1	0.2	0.3
DSA1 360mg/l	0.1	0.1	0.3
DAA1 17.61mg/l	0.2	0.2	0.3
DAA1 88.06gm/l	0.1	0.3	0.3
DAA1 176.12mg/l	0.2	0.2	0.2
DSA 2 40mg/l	0.3	0.1	0.3
DSA2 200mg/l	0.2	0.2	0.3
DSA2 360mg/l	0.3	0.1	0.3
DAA2 17.61mg/l	0.2	0.2	0.3
DAA2 88.06mg/l	0.2	0.3	0.3
DAA2 176.12mg/l	0.1	0.1	0.3
DSA3 40mg/l	0.2	0.2	0.4
DSA3 200mg/l	0.3	0.2	0.3
DSA3 360mg/l	0.1	0.1	0.1
DAA3 17.61mg/l	0.1	0.2	0.2
DAA3 88.06mg/l	0.3	0.2	0.3
DAA3 176.12mg/l	0.2	0.1	0.1

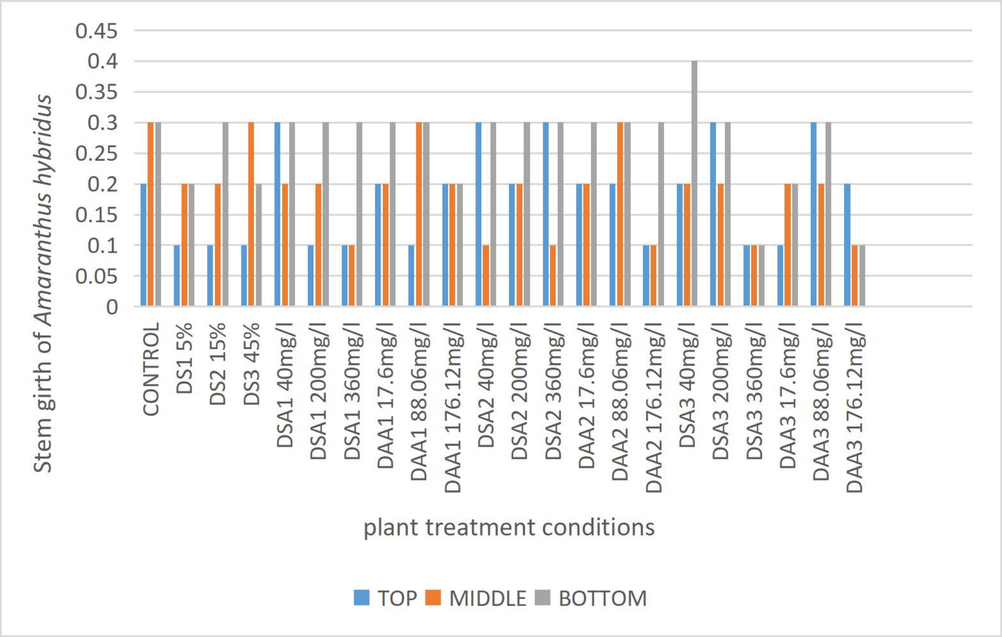


Figure 4.3: Graphical representation of stem girth per plant of A. *hybridus* for 6 weeks of being exposed to growth regulators (SA and AA) and regimented water

Note: The graph shows the stem girth per plant of A. *hybridus* for week 6 for different plant treatments. DSA3 40mg//l had the highest stem girth per plant in week 6. Then DAA3 176.12mg/l had the lowest stem girth per plant in week 6.

Table 4.4: The length of root of *Amaranthus hybridus* plant for 6weeks after being exposed to growth regulators(SA and AA)and regimented water

Plant treatment conditions	Length of root(6weeks)
Control	**20.1**
DS1 5%	**20**
DS2 15%	**31**
DS3 45%	**22**
DSA1 40mg/l	**23.4**
DSA1 200mg/l	**21.6**
DSA1 360mg/l	**30**
DAA1 17.61mg/l	**25.1**
DAA1 88.06gm/l	**85**
DAA1176.12mg/l	**20.3**
DSA2 40mg/l	**31**
DSA2 200mg/l	**40**
DSA2 360mg/l	**28**
DAA2 17.61mg/l	**22.4**
DAA2 88.06mg/l	**32**
DAA2 176.12mg/l	**17.5**
DSA3 40mg/l	**20**
DSA3 200mg/l	**19.2**
DSA3 360mg/l	**27**
DAA3 17.61mg/l	**18.2**
DAA3 88.06mg/l	**31**
DAA3 176.12mg/l	**24**

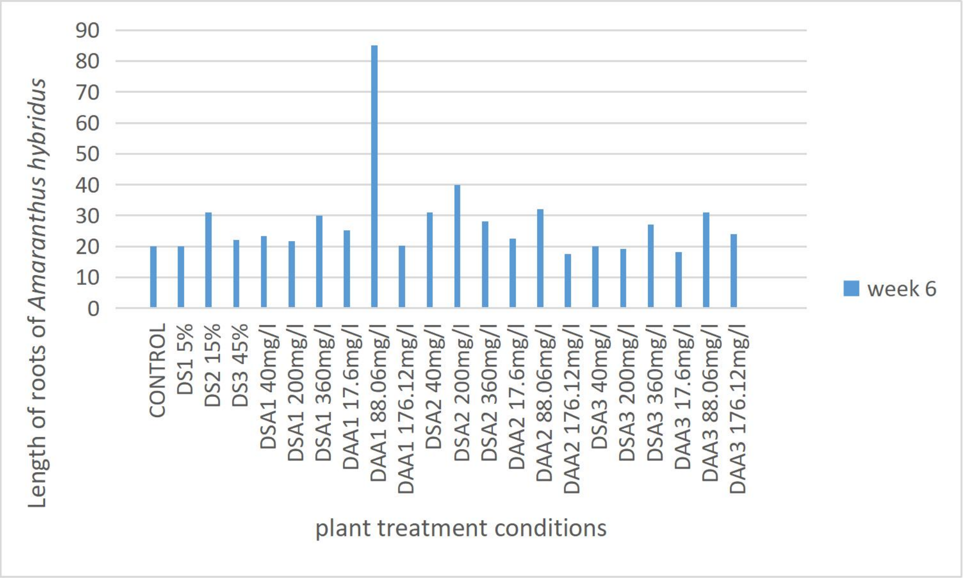


Figure 4.4: Graphical representation of length of roots of A. *hybridus* plant for 6 weeks of being exposed to growth regulators (SA and AA) and regimented water

Note: The graph shows the length of A. *hybridus* plant for week 6 for different plant treatments. DAA1 88.06gm/l had the highest length of root in week 6. Then DAA2 176.12mg/l had the lowest length of root in week 6.

Table 4.5: Weight of seed per plant of *Amaranthus hybridus* for 6weeks after being exposed to plant growth regulators (SA and AA) and regimented water

Plant treatment conditions	Weight of seed per plant (Week 6)
Control	3.261
DS1 5%	2.431
DS2 15%	4.065
DS3 45%	4.119
DSA1 40mg/l	2.731
DSA1 200mg/l	2.409
DSA1 360mg/l	1.387
DAA1 17.61mg/l	1.363
DAA1 88.06gm/l	2.096
DAA1176.12mg/l	2.509
DSA2 40mg/l	3.067
DSA2 200mg/l	2.601
DSA2 360mg/l	3.222
DAA2 17.61mg/l	2.904
DAA2 88.06mg/l	2.945
DAA2 176.12mg/l	2.450
DSA3 40mg/l	0.123
DSA3 200mg/l	0.234
DSA3 360mg/l	1.342
DAA3 17.61mg/l	0.239
DAA3 88.06mg/l	0.435
DAA3 176.12mg/l	0.051

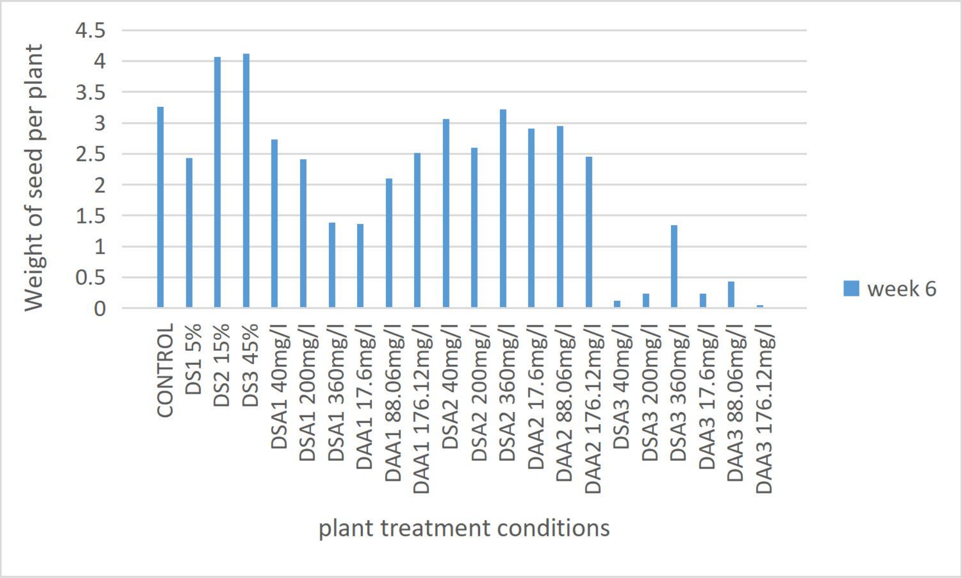


Figure 4.5: Graphical representation of weight of seed per plants of A. *hybridus for* 6 weeks for different treatments (SA and AA) and regimented water

Note: The graph shows the weight of seed per plant of of A. *hybridus* for week 6 for different plant treatments. DSA2 360mg/I had the highest weight of seed in week 6 and DAA3 176.12mg/l had the lowest weight of seed in week 6

Table 4.6: Colour chart showing the colours of *Amaranthus hybridus* for different treatments after being exposed to growth regulators (SA and AA) and regimented water

Plant treatment conditions	Week 6
CONTROL	Colour 2
DS1 5%	Colour 2
DS2 15%	Colour 3
DS3 45%	Colour 3
DSA1 40mg/l	Colour 2
DSA1 200mg/l	Colour 3
DSA1 360mg/l	Colour 2
DAA1 17.61mg/l	Colour 3
DAA1 88.06gm/l	Colour 2
DAA1176.12mg/l	Colour 2
DSA2 40mg/l	Colour 2
DSA2 200mg/l	Colour 1
DSA2 360mg/l	Colour 2
DAA2 17.61mg/l	Colour 2
DAA2 88.06mg/l	Colour 1
DAA2 176.12mg/l	Colour 3
DSA3 40mg/l	Colour 3
DSA3 200mg/l	Colour 2
DSA3 360mg/l	Colour 2
DAA3 17.61mg/l	Colour 3
DAA3 88.06mg/l	Colour 1
DAA3 176.12mg/l	Colour 1

KEYS:

DSA- drought stress plant exposed to salicylic acid only

DAA-drought stress plant exposed to absorbic acid only

DS- drought stress plant exposed to water deficit only

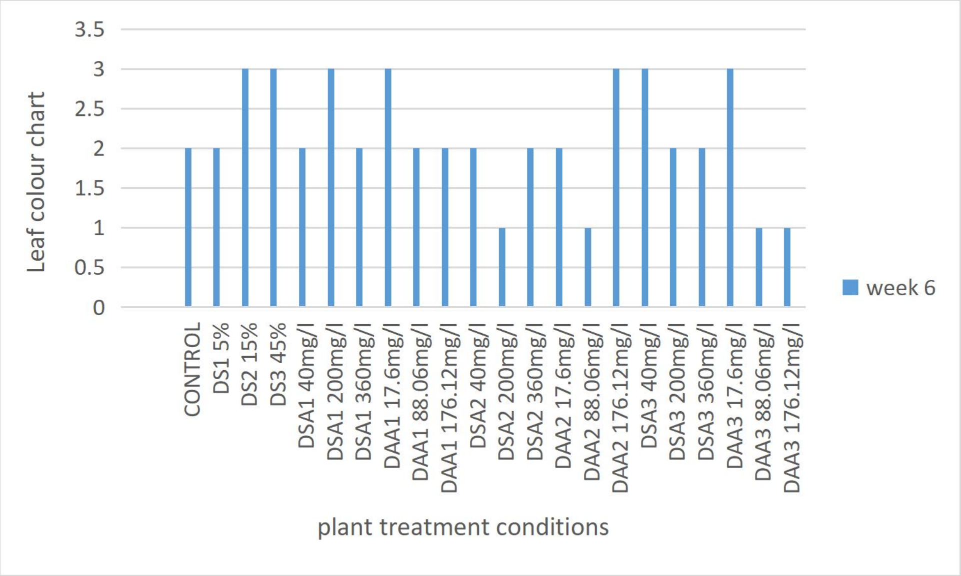


Figure 4.6: Graphical representation of colour chart showing the colour of A. *hybridus* for different treatments for 6 weeks of being exposed to growth regulators (SA and AA) and regimented water

Note: The graph shows the colour chart of A. *hybridus* in week 6 of different plant treatments. The highest colour chart were gotten from different treatments in week 6. The lowest leaf colour chart were gotten from DSA2 200mg/l, DAA2 176.12mg/l, DAA3 88.06mg/l, and DAA3 176.12mg/l in week 6.

Plate 4.1 Height of A. *hybridus* plant in 4 weeks after being exposed to sufficient watering for different treatments (DSA3 40mg/l, DSA3 200mg/l, DSA360mg/l, DAA3 17.6mg/I, DAA3 88.06mg/l, DAA3 176.12mg/I)

Plate 4.2: Height of *Amaranthus hybridus* plant in 6weeks after being exposed to treatment (DAA3 88.06mg/I)

Plate 4.3: colour chart showing the colour of *Amaranthus hybridus* leaf in 6weeks after being exposed to treatment (DSA3 40mg/I)

Plate 4.4: Detaching of flourescence for drying of treatment (DSA3 40mg/l) in 6weeks after being exposed to salicylic acid and regimented water

CONTROL

DS1 5%

DS2 15%

DS3 45%

DSA1 40mg/l

DSA1 200mg/l

DSA1 360mg/l

DSA2 40mg/l

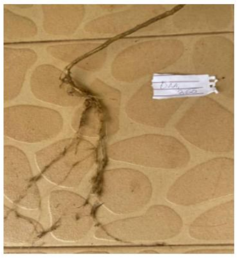
DSA2 200mg/l

DSA2 360mg/l

DSA3 40mg/l.

DSA3 200mg/l.

DSA3 360mg/l



DAA1 17.61mg/l.

DAA1 88.06mg/l

DAA1 176.12mg/l

DAA2 17.61mg/l. DAA2 88.06mg/l DAA2 176.12mg/l

DAA3 17.6mg/l DAA3 88.06mg/l DAA3 176.12mg/l

Plate 4.5: Some root hairs of *Amaranthus hybridus* in 6weeks after being exposed to SA and AA.

CHAPTER FIVE

DISCUSSION

This study examined the effects of salicylic acid (SA) and ascorbic acid (AA) on the growth of *A. hybridus* under drought stress and well-watered conditions.

Over the past two decades, growing evidence has demonstrated that SA plays a vital role in regulating plant growth and development (Vanacker et al., 2001; Scott et al., 2004; Miura et al., 2010; Fujikura et al., 2020).

The height of the plants varied across different treatments with DAA3 (17.6 mg/L) showing the greatest height (133 cm) in the sixth week following exposure to the growth regulator (ascorbic acid). Zelalem (2018) similarly reported that higher concentrations of ascorbic acid enhance plant height, particularly in well-watered plants. In addition, DSA3 (40 mg/L) exhibited the highest number of leaves (215 leaves) after six weeks of exposure to salicylic acid under controlled water conditions. Previous studies have also confirmed that increased concentrations of salicylic acid can enhance leaf production in plants (Khan et al., 2016; Jabbarzadeh et al., 2009).

For root development, DAA1 (88.06 mg/L) recorded the longest root length (85 cm) after six weeks of treatment with salicylic and ascorbic acids under regulated water conditions. This finding aligns with (Worku M, 2020), who observed that higher concentrations of ascorbic acid promoted root elongation in *Amaranthus hybridus* . Overall, the results suggest that both ascorbic and salicylic acids contribute to stem girth development in A. *hybridus* (Tesfaye, 2019). The treatment DSA3 (40 mg/l) showed the largest stem girth at the base (0.4 cm) after exposure to the growth regulators and controlled watering.

Previous research has documented various morphological adaptations of A. *hybridus* to drought stress (Sarker et al., 2019). Such adaptations often include alterations in root architecture and stem thickness, which help improve water absorption from deeper soil layers (Mepha et al., 2017). Other studies have also reported that drought stress tends to reduce plant height, stem girth, and overall biomass accumulation (Sarker et al., 2018).

Regarding seed development, DSA2 360mg/l had the highest seed weight per plant (3.222) after six weeks of exposure to AA, and regulated water conditions, while DAA3 176.12 mg/l recorded the lowest seed weight (0.051 g). According to Akombi et al. (2018), the application of ascorbic and salicylic acids in A. *hybridus* can enhance seed establishment and potentially increase seed yield. Salicylic acid has been shown to influence seed germination and seedling growth, which may lead to higher seed weights.

Furthermore, the observed effects of SA and AA on the leaf color of A. *hybridus* *i*ndicate their influence on chlorophyll synthesis (Mavenghama et al., 2011). SA helps plants maintain higher chlorophyll levels under stress, while AA functions as an antioxidant that protects chlorophyll from degradation, resulting in deeper green foliage (Jimoh et al., 2012).

These morphological and physiological adaptations are essential for A. *hybridus* survival and resilience in water-limited environments (Amusat et al., 2018).

CONCLUSION

The morphological responses of *Amaranthus hybridus* to drought stress involve a combination of adaptations that aim to conserve water and enhance survival. They also play roles in increased number of leaves , root development, stem girth and plant height, and changes in leaf colour and weight of seeds of *Amaranthus hybridus* . Absorbic acid was observed to result in significant increase in height of plants and length of root, While salicylic acid had increase in the number of leaves, These responses enable the plant to cope with water scarcity and maintain its physiological functions under drought stress conditions.

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RECOMMENDATION

To mitigate the effects of drought stress on *Amaranthus hybridus*, several recommendations can be made based on morphological responses. Implementing irrigation strategies to maintain soil moisture levels is crucial. Selecting drought-tolerant varieties of *Amaranthus hybridus* can also be beneficial. Additionally, practices like mulching can help conserve soil moisture and reduce water evaporation, while adjusting planting density can optimize resource utilization. These measures collectively aim to enhance the plant's ability to cope with water scarcity and maintain growth.

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