EFFECTS OF METHYLJASMONATE AND SALICYCLIC ACID ON SELECTED BIOCHEMICAL PARAMETERS IN PATHOGEN- INFESTED OIL PALM SEEDLINGS

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BENIN CITY

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A PROJECT REPORT SUBMITTED TO THE DEPARTMENT OF BIOCHEMISTRY, FACULTY OF LIFE SCIENCES, UNIVERSITY OF BENIN, BENIN CITY IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF A BACHELOR OF SCIENCE (B.Sc Hons) DEGREE IN BIOCHEMISTRY

APRIL, 2024.

CERTIFICATION

This is to certify that this project work was thoroughly researched into by **EKEBAFE OMOTINE EXCEL** matriculation number **LSC1906484** as part of the requirements for the award of the Bachelor of Science Degree Award (B.SC) in Biochemistry.

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DEDICATION

I dedicate this to God and also to Dr. (PST) and Dr. (Mrs.) Ekebafe, who was always there for me, even on the tough days. their steadfast commitment, love, and dedication will not be forgotten.

ABSTRACT

Fungal infections are major hazards to crop health and yield in agricultural settings. This study investigates into how well salicylic acid and methyljasmonate work to lessen the negative effects of fungal pathogen exposure on oil palm seedlings.The effects of these phytohormones on the production of important plant biomolecules and antioxidants, such as carotenoids, lycopene, ascorbic acid, total sugar, proline, and vitamins A and E, was accessed by thorough investigation. The results show that when oil palm seedlings are exposed to fungal infections, their levels of significant plant compounds and antioxidants significantly decrease. Nevertheless, the utilization of salicylic acid and methyljasmonate exhibits an impressive ability to mitigate these deleterious consequences. In particular, both phytohormones efficiently promote the synthesis of vital biomolecules, preserve or increase antioxidant and vitamin levels, and reduce lipid peroxidation brought by fungal pathogen challenge. These findings highlight the phytohormones' potential as long-term and efficient tools in farming methods meant to lessen the negative effects of fungal infections on crop quality and productivity. Innovative crop protection and management techniques for oil palm farming can be developed with important insights from an understanding of the molecular mechanisms underlying the protective actions of salicylic acid and methyljasmonate.

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INTRODUCTION AND LITERATURE REVIEW

1.1 Introduction

Oil palm (Elaeisguineensis Jacq.) is a perennial crop that originated in West Africa. It was introduced to Brazil and other tropical nations by the Portuguese in the 15th century *(Corley, 1976).* It is cultivated as a resource to be utilized in the international food, home, and industry's goals. In the 1870s, the British brought oil palm as an attractive plant to Malaysia, then known as Malaya. The first commercial oil palm plantation in Malaysian history was established in 1917. Due to its 30% global contribution to the production of edible oils and fats and the fact that it requires the least amount of land to produce, palm oil is rapidly gaining popularity.

Palm oil is very productive and economical on a big scale, with increasing demand. Despite being the second-largest producer of oil palm in the world, Malaysia will likely have to work hard to keep that ranking. Less suitable soil for oil palm cultivation, sustainability concerns, and food safety difficulties are a few of the drawbacks.

Above all, though, the most significant difficulties are keeping an eye on the health of the palm tree and preserving its economic output, which is impacted by pest and disease issues as well as inadequate management techniques. Malaysia has seen a tremendous increase in oil palm hectares over the last four decades as a result of a lack of appropriate land and restricted alluvial soil. This has resulted in a growing trend of oil palm agriculture on tropical peat soils, which are seen to be a suitable medium for oil palm development because of their uniform qualities, steady supply of water, and levelness. Nevertheless, peat soil has a low nutritional content by nature and is prone to diseases, especially the fungus Ganoderma, which can cause significant losses in crop production. There are many who are skeptical about the viability of using peat soil in oil palm plantations.To improve palm oil production, it's important to implement new plantations, regenerate old ones, use high-yielding planting materials, and implement effective pest and disease control measures to close the yield gap between field trials and plantations *(Jalani et al. 2003).* G. boninense's severe infestations on oil palm crops caused significant yield losses due to basal stem rot (BSR). Thompson initially reported the ailment in 1931, when oil palm was introduced in Malaysia. According to a recent BSR survey conducted in 2009-2010, Malaysia's BSR was 3.7%, equating to 59,148 ha and approximately 45.0% of estates *(Idris et al., 2011)*.

The application of Methyljasmonate (MeJA) and salicylic acid (SA) can have various effects on the growth and biochemical parameters of oil palm seedlings, particularly in response to pathogen infestation.Methyljasmonate has been shown to increase the amounts of bioactive substances in oil palm seedlings, thereby enhancing their antioxidant activity. It has also been found to promote the growth and photosynthesis of maize seedlings under saline conditions by enhancing energy metabolism and fatty acid content. Salicylic Acid, on the other hand, can maintain plant growth and development and respond to abiotic stressthe exogenous application of Methyljasmonate and Salicycliic has been shown to increase the amounts of bioactive substances in oil palm seedlings, thereby enhancing their antioxidant activity. The aim of this study is to determine the effects of Methyjasmonate and Salicyclic acid on selected parameters in pathogen- infested oil palm seedlings

1.1.1 Statement of Problem

There's need to improve the growth and biochemical parameters of oil palm seedlings, particularly in response to pathogen infestation. Previous studies have shown that the application of Methyljasmonate and Salicyclic Acid can increase the amounts of bioactive substances in oil palm seedlings, thereby enhancing their antioxidant activity and improving their quality *[Harith-Fadzilah et al., 2021].* However, the specific effects of these hormones on the growth and biochemical parameters of oil palm seedlings in response to pathogen infestation have not been extensively studied. Therefore, there is a need to investigate the effects of Methyljasmonate and Salicyclic Acid on the growth and biochemical parameters of oil palm seedlings under pathogen infestation, with the aim of developing effective strategies for improving the growth and productivity of oil palm seedlings.

1.1.2 Aim and Objective Of The Study

The aim of this study was to conduct a comprehensive investigation to ascertain the effects of salicylic acid and methyljasmonate on particular parameters in oil palm seedlings infected with pathogens.

1.1.3 Specific Objectives of The Study

The study has the following precise aims:

- Determine the effects of Methyl Jasmonate and Salicylic acid on infested oil palm seedlings
- Investigate and compare the defence responses induced by Methyl Jasmonate and Salicylic Acid in oil palm seedlings
- Determine whether treatment with Methyl Jasmonate and Salicylic acid increases plant resistance to fungal infection in oil palm seedlings.

1.2 Literature Review

1.2.1 Description of Oil Palm

Scientifically referred to as Elaeis guineensis, oil palm is a multipurpose plant with substantial global economic significance. The coconut is the only other plant of similar significance, making it the main source of vegetable oil and fat.The oil palm originated in West Africa, but in the late 1800s, it was brought to tropical Southeast Asia and Latin America.Year-round production of oil-rich fruit bunches with 1000–3000 individual fruits is a prolific feature of E. guineensis plants (Corley and Tinker 2015).Traditionally used for cooking in Africa, the fruit's oil has found use in a variety of applications, such as replacing animal fats like butter in baked goods, soaps, and cosmetics, or serving as a foundation for biodiesel*(Martin Russel, 2018).* Many nations today rely heavily on oil palm for their economy, particularly Malaysia and Indonesia, from which huge amounts of oil, meal, and other derivatives are exported *(Murphy 2019)*. Nowadays, oil palm is grown in plantations throughout the humid tropics of Asia, Africa, Caribbean and Americas, where its goods are shipped to markets throughout the world. The two major producers, Malaysia and

Indonesia, account for 10% and 5% of their exports, respectively, and hence bear the brunt of the economic and social effects of oil palm farming. In addition to the 4 million workers and smallholders employed by the sector in Indonesia, it supports an additional 11 million people in both Malaysia and Indonesia through indirect employment. In order to support rural development and reduce poverty, the majority of oil palm jobs are located in isolated rural areas with little opportunities for alternative employment. On the other hand, the two biggest producers of oil palm, Indonesia and Malaysia, bear a substantial environmental cost associated with production. Its production still requires a lot of work to become more sustainable. Because palm oil contributes to deforestation and may result in higher total greenhouse gas emissions than fossil fuels, its use in the production of biofuels is especially contentious. The use of palm oil for biofuels would be restricted under changes the European Parliament suggested in January 2018 to the EU's draft directive on renewable energy. The oil palm tree, which is a major source of vegetable oil and fat, begins its life as oil palm seedlings.These seedlings are produced in nurseries, where they are carefully nurtured and prepared for transplantation to plantations. Oil palm seedlings require specific conditions for optimal growth, including an evenly distributed annual rainfall of 2000 mm without a defined dry season, a maximum average temperature of 29-33 degrees, and a minimum average temperature of 22-24 degrees. The oil palm seedling is a small oval-shaped fruit that grows on a hardy and rugged plant, which is a hybrid variety known as Tenera. This variety is highly productive, yielding 500-900% more than the local Nigerian oil palm seedling. Tenera oil palm seedlings are preferred for their high yield, better disease and pest tolerance, and superior oil quality. Oil palm seedlings require careful handling during transplantation to minimize transplanting shock and ensure early establishment. Seedlings should be removed from the main nursery with a spade, and the roots should be carefully cut. It is important to keep the seedlings in the shade and plant them as soon as possible after lifting. Once transplanted, oil palm seedlings require frequent watering, particularly during the hot season. They are heavy feeders and require regular, complete fertilizer to support their growth. Growing seedlings in polythene bags can help promote vigorous growth and minimize transplanting shock.

1.2.2 Scientific Classification of Plant Species

- Kingdom……………………………………………………Plantae
- Phylum...Magndiophyta
- Class..Liliopsida
- Subclass...Arecidae
- Domain……………………………………………………Tracheophytes
- Order………………………………………………………Arecales
- Family……………………………………………………Arecaceae
- Genus………………………………………………………Elaeis
- Specie……………………………………………………E. guineensis

1.2.3 Nomenclature

Scientific Name: *Elaeis guineensis Jacq*.

Common Name(s): African oil palm, oil palm, and macaw-fat

Nigerian Name(s): *Aki naube*(Igbo), *Igiaku*(Yoruba), *Kasa* (Hausa).

1.2.4 Habitat

Oil palm seedlings, particularly the Tenera and Supergene hybrid kinds, thrive in tropical lowland.

1.2.5 Ecological Conditions

Minimum average temperature of 22–24 degrees, highest average temperature of 29–33 degrees, and equally distributed annual rainfall of 2000 mm are necessary for oil palm seedlings without a designated dry season. Deep soil and constant high temperatures (30–32°C/86–89.6°F for at least 80 days) are necessary for these plants to produce at their peak. With pH values ranging from 4.0 to 8.0, they can grow in a range of deep tropical soils. Even though they can tolerate two to three months of dry weather, oil palm seedlings need regular watering all year round. Long-lasting dry weather, however, may lower yields. The best yields of oil palm are produced in areas with no distinct dry season and evenly distributed 2000 mm of yearly rainfall. Furthermore, the places where these seedlings flourish have highest average temperatures of 22-24 degrees Celsius. *(Jing Cui et al, 2020)*

1.2.6 Propagation

Usually, oil palm trees are grown from seed, which is moistened and placed in a tiny box to germinate before planting. Following germination, the seeds are permitted to continue growing in a pre-nursery, which is a bed, tray, or container filled with rich soil. After that, the seedlings are moved to a nursery and given another six to twelve months to flourish before being planted in the field. Nine meters separates field seedlings from trees, and there are another nine meters between rows. Trees take three to four years to start producing fruit. Indirect somatic embryogenesis is another method used to grow the oil palm (Elaeis guineensis Jacq.) in vitro.

The oil palm (Elaeis guineensis Jacq.) is also propagated in vitro by indirect somatic embryogenesis, a process in which somatic cells of an explant of choice are, via an intermediate phase of callus growth, induced to differentiate into somatic embryos. (*Sylvie weckx et al, 2019)*

Figure 1: Showing Oil palm seedling

Source: Google Image

Figure 2: Showing African oil palm *Elaeisguineensis*

Source: Google Image

1.3 Diseases Of Oil Palm

1.3.1 Basal Stem Rot Disease (BST)

The pathogenic fungus Ganoderma boninense is the source of basal stem rot (BSR), a serious disease that affects oil palm plants *(Latiffah Zakaria, 2023).*In Malaysia and Indonesia, where it is predicted to cause significant economic losses of up to USD 500 million annually, this disease is very damaging.The disease is common in coastal Southeast Asian regions, particularly in oil palm farms built on peat soils or on land that was formerly home to coconut or forest trees. It has been shown to affect oil palm trees of various ages, including those as young as one year old. The presence of a white rot at the base of the stem, an abundance of spear leaves, and leaf yellowing are all signs of BSR.if left untreated, the disease can cause the death of up to 80% of plantings halfway to their economic life. An environment that promotes the development of the disease, a susceptible host, and a virulent infection interact to affect the course of the disease (Nur Aliyah et al., 2022).Effective disease management requires early diagnosis of BSR. Kuriya et al. (2022) have suggested that hyper spectral imaging using unmanned aerial vehicles (UAVs) could be an effective technique for identifying BSR in oil palm trees at an early stage. In order to determine spectral signals linked to BSR infection, this technology uses unmanned aerial vehicles (UAVs) fitted with hyperspectral sensors to take high-resolution pictures of oil palm trees. In their study, *(Kurihara et al.2022).* This method involves the use of UAVs equipped with hyper spectral sensors to capture high-resolution images of oil palm trees, which are then analyzed to identify spectral signatures associated with BSR infection. The work by *(Kurihara et al., 2022)* showed the viability of this strategy by employing UAV-based hyper spectral imaging to detect BSR-infected oil palm plants with an accuracy of 92.3%.To effectively manage BSR in oil palm farms, integrated disease management techniques—such as cultural practices, chemical control, and the application of fertilizers or biocontrol agents—along with early detection are crucial (Latiffah Zakaria, 2023).It is still early in the process of developing resistant or tolerant oil palm cultivars, and more investigation is required to pinpoint the genes, gene products, and metabolic pathways that are a part of oil palm defensive mechanisms against G. boninense.

Figure 3: Image showing Stem Showing BSR of Oil Palm

Source: Google Image

1.3.2 Bud Rot

When spear leaves in oil palm seedlings develop yellowing, they eventually turn brown due to bud rot. This sign indicates the presence of bud rot in oil palm seedlings. One.In oil palm plantations, bud rot are a serious disease that, if ignored, can kill the palm and result in financial losses. For oil palm seedlings to remain healthy and productive, bud rot must be properly monitored and controlled. *(Kaduch,2008)*

Figure 4: Image showing Bud Rot of Oil Palm

Source: Google Image

1.3.3 Anthracnose

The health and productivity of oil palm farms can be negatively impacted by anthracnose, which is a serious concern. The disease appears as green or greenish-grey spots on leaves that eventually turn yellow, tan, reddish-brown, or brown, frequently with a contrasting color around the lesion. Usually starting in the veins of the leaves, these diseases can spread quickly and cause the leaves to wilt, discolor, and sear. In extreme situations, the disease may destroy the spear leaf and the palm's growth point, which could result in serious consequences and lower plantation yields. A fungal agent is the cause of anthracnose in oil palm, and the disease cycle entails the release of spores from affected tissue, like falling leaves, and the infection of new growth, including fruits, flowers, branches, and leaves *(Ohana costa et al, 2018)*. Plant cells provide the fungus with nutrition, which results in cell death and the development of lesions. When there are extended periods of cool weather and leaf moisture, spores from leaf lesions can re-infect the same leaf or nearby leaves(Alham aji et al ,2013).Oil palm anthracnose is managed by keeping an eye on the plantation, making sure there is adequate airflow, and applying fungicides as needed to lessen the severity and frequency of the illness. The effects of anthracnose can be lessened by planting trees with the proper spacing between them, maintaining the plantation free of weeds, and trimming to increase air circulation within the crown. Although the illness mostly affects the tree's appearance and does not pose a significant threat, in extreme situations it can result in ugly plantations and create producers' concerns. In order to prevent anthracnose and preserve the well-being and output of oil palm plantations, early detection and efficient management techniques are crucial.

1.3.4 Brown Germ

The brown germ of an oil palm seedling is the embryonic axis enclosed by the seed coat, responsible for the seedling's growth. It is the part of the seed that germinates, giving rise to the shoot and root system. Rich in nutrients, the brown germ is protected by the hard endocarp layer formed by the kernel. The oil extracted from the kernel has industrial applications distinct from mesocarp oil*(Cui et al, 2020)*.The germination process of oil palm seeds is a complex metabolic pathway involving the remobilization of seed storage reserves like lipids to support seedling growth. A critical stage in oil palm cultivation, germination is a long and challenging process requiring specific conditions to break dormancy and establish seedlings *(Rokhana et al, 2022)*. Despite its importance, our knowledge of the metabolic pathways and control mechanisms involved in germinating oil palm seed metabolism remains limited. Germination in oil palm seeds includes water imbibitions, activation of metabolic pathways, and the emergence of the radicle piercing the seed envelopes. In palm tree species like oil palm, the first anatomical structure to pierce the seed envelopes is not the radicle but the plumule, the embryonic shoot that emerges from the seed. The plumule then gives rise to the shoot system, while the radicle gives rise to the root system. Various factors influence the germination process in oil palm seeds, such as temperature, moisture, and light. Optimal conditions for germination vary among different oil palm varieties and depend on the specific conditions under which the seeds were produced and stored*(Jing Cui, 2020)*.Enhancing the germination process can be achieved by providing seeds with adequate moisture, maintaining a suitable temperature range, and protecting them from excessive conditions..

Figure 5: Image showing Brown germ of Oil Palm

Source: Google Image

1.3.5 Thielaviopsis Paradoxa

A pathogenic fungus called Thielaviopsis paradoxa causes spear rot in young oil palm trees. Oil palm seedlings can be genetically engineered to contract this disease by introducing a fungus suspension into the base of the stems when the seedling stage is reached. Severity scores are then used to determine the disease's severity *(Álvarez et al., 2012)*. To mimic the symptoms of the disease in a controlled setting, four inoculation techniques have been investigated: direct contact with agar blocks containing mycelia and conidia, drip, scissor cut, and local infiltration. T. paradoxa infections and the onset of illness symptoms were the outcomes of all four techniques. The inoculation technique and dosage were connected with the severity of the disease. The teleomorph Ceratocystis paradoxa (Dade) Moreau was not found in studies aimed at testing Koch's postulates, but T. paradoxa was isolated from disease progression sites in the inoculated trees 4. This indicates that the disease affecting oil palm seedlings is mostly caused by T. paradoxa.In order to evaluate plant pathogenesis, it is often advised to generate the disease in a controlled environment using the local infiltration approach (1 x 106 endoconidia mL−1) and to leave it for 3–7 days*(Gaitán-Chaparro et al., 2021)*

Figure 6: Image showing Thielaviopsis Paradoxa of Oil Palm

Source: Google Image

1.4 BIOTIC STRESS

Attacks by different diseases, including fungi, bacteria, oomycetes, nematodes, and herbivores, are referred to as biotic stress. illnesses brought on by Globally, these diseases are primarily responsible for yield loss. Because they are sessile, plants are unable to avoid these messages from their surroundings. Proficiency in managing these strains is essential for effectively concluding the life cycle. Plants have therefore evolved a variety of defense systems to withstand these attacks and survive in such environments *(Manghwar and Zaman, 2024)*. When they perceive an external stressor, they become activated and produce the necessary biological reactions. These physiological reactions function by transmitting impulses from sensors on the cytoplasmic or cell surface to the transcriptional machinery in the nucleus via a variety of signal transduction channels. Differential transcriptional modifications result from this, making the. Differential transcriptional modifications result from this, strengthening the plant's resistance to the stress. The signaling pathways are essential because they serve as a conduit for the information needed to detect the stressor and produce the proper physiological and biochemical reaction *(Zhu et al., 2002)* Current research employing a proteomics and genomics methodology. Tense numerous potential microbial pathogens, including bacteria, fungus, oomycetes, nematodes, and herbivores, are continuously present in plants. Plants have evolved a multitude of defense mechanisms to protect themselves, many of which are triggered by pathogen invasion. The plant plasma membrane is exposed to the microorganisms during cell wall penetration, and it is here that they come into contact with extracellular surface receptors that are capable of identifying pathogen-associated molecular patterns (PAMPs). When a microorganism is recognized at the cell surface, PAMP-triggered immunity (PTI), which normally stops infection before the pathogen takes hold in the plant, is activated by recognition of a microbe at the cell surface *(Manghwar and Zaman, 2024)*. Nevertheless, pathogenic microorganisms have developed a way to inhibit PTI by secreting specific proteins into the cytosol of plant cells that change resistance signaling or the way resistance responses manifest. These proteins are known as effectors.The interaction between plant defense and dark/light environment activates a signaling cascade, acting as a connecting link between perception of biotic stress, dark/light environment, and generation of an appropriate physiological or biochemical response. The present review highlights molecular responses arising from dark/light fluctuations vis-à-vis elicitation of defense mechanisms in plants. Plants have co-evolved with their parasites for several hundred million years, resulting in the selection of a wide range of plant defenses against microbial pathogens. These defenses include both physical and chemical adaptations, which may be expressed constitutively or activated only in response to attack. For example, utilization of high metal ion concentrations derived from the soil allows plants to reduce the harmful effects of biotic herbivores *(Gull et al., 2019)*

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1.5 OXIDATIVE STRESS IN PLANTS

Reactive oxygen species (ROS), which are extremely reactive and dangerous chemicals that can cause oxidative cell death, are the source of oxidative stress in plants when there is an imbalance between their production and elimination. Plants produce ROS as a consequence of their aerobic metabolism, and they take part in redox signaling pathways that control the growth, development, and stress adaption of plants. However, high ROS concentrations can damage cellular constituents, resulting in oxidative stress. In plants, the endoplasmic reticulum, mitochondria, chloroplasts, peroxisomes, apoplasts, plasma membranes, and cell walls are the main producers of reactive oxygen species *(Hasanuzzaman et al., 2020)*.ROS generation can be triggered by abiotic stress factors such as heat, cold, drought, salt, ozone depletion, heavy metal toxicity, and UV radiation. These stressors upset the delicate balance within cells, leading to a rise in ROS generation that exceeds the ability of antioxidant defences. Causing an oxidative burst, affecting biomolecules, and upsetting the redox balance within cells. In plants, ROS play two roles. At low concentrations, they act as signalling molecules that help plant cells respond to different stimuli, both biotic and abiotic. On the other hand, they harm cellular constituents in excessive quantities, resulting in oxidative stress *(Sharma et al., 2012)*.Enzymes and tiny antioxidant molecules control the actions of these dual-function entities in both cases. To regulate ROS levels within cells and avoid oxidative stress, plants have both enzymatic and non-enzymatic antioxidant systems. Superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), glutathione reductase (GR), and peroxidase (POX) are the enzymes that provide protection against oxidative stress by neutralizing excess reactive oxygen species (ROS) *(Mhamdi and Van Breusegem, 2018)*. Non-enzymatic antioxidants like vitamins, flavonoids, stilbenes, and carotenoids also contribute to scavenging excess ROS and shielding against oxidative stress. Climate change has the potential to exacerbate oxidative stress in plants by increasing the overall levels of ROS.

1.6 ANTIOXIDATIVE DEFENSE SYSTEM IN PLANT

Plants rely on antioxidant systems to maintain a balance between the production and elimination of reactive oxygen species (ROS). ROS are generated in plants during various processes associated with abiotic stress, such as high light intensity, drought, salinity, and extreme temperatures. These ROS can cause significant damage to important cellular components like carbohydrates, lipids, proteins, DNA, and others, resulting in oxidative stress in plants.The antioxidant defense system in plants consist of low-molecular-weight nonenzymatic antioxidants and antioxidant enzymes *(Kasote et al., 2015)*. The nonenzymatic antioxidants include ascorbic acid (AsA), glutathione (GSH), α-tocopherol, phenolic compounds, flavonoids, alkaloids, and nonprotein amino acids. These antioxidants work together with antioxidant enzymes like superoxide dismutase (SOD), catalase (CAT), peroxidase (POX), polyphenol oxidase (PPO), ascorbate peroxidase (APX), monodehydroascorbate reductase (MDHAR), dehydroascorbate reductase (DHAR), glutathione reductase (GR), glutathione peroxidase (GPX), and glutathione S-transferase (GST) to prevent excessive ROS production. The initial line of defense in the stress response is the enzyme SOD, which transforms superoxide radicals (O2•−) into hydrogen peroxide (H2O2) *(Fujita and Hasanuzzaman, 2022).* Enzymes such as CAT, APX, GPX, or the AsA-GSH cycle can then further convert this H2O2 into water (H2O). The main antioxidant defense mechanism that detoxifies H2O2 in plant cells is the AsA-GSH cycle, sometimes referred to as the Asada-Halliwell cycle. Exogenous antioxidants can be applied to plants to enhance their development and stress tolerance, according to studies. Antioxidants can change the photosynthetic membrane, boost the stability and activity of the enzymes involved in photosynthesis, and raise the quantity and stability of photosystems II (PSII) and I (PSI) *(Hasanuzzaman et al., 2020).* Additionally, exogenous application of antioxidants can lead to their accumulation, thereby increasing the pool of antioxidant molecules and enhancing the reductive metabolism of photosynthesis.

1.7 Non- Enzymatic Antioxidant System In Plants

Ascorbic Acid

Under stressful conditions, plants produce ascorbic acid, also known as vitamin C, which is a potent antioxidant that helps scavenge reactive oxygen species (ROS). Some of the physiological and biochemical functions it performs include photosynthesis, floral induction, fruit enlargement, ROS regulation, and senescence *(Akram et al., 2017).* Several different enzymes and intermediates are used by plant cells to produce it. According to (Bilska et al., 2019).Ascorbic acid can be used as a substrate to detoxify the ROS produced during stress. Lipid peroxidation is reduced, antioxidant enzyme activity is increased, and plant resistance to abiotic stresses is enhanced when ascorbic acid is administered topically. By interacting with plant hormones, it also regulates growth and development *(Akram et al.,2017)*

Tocopherols

Tocopherols, a form of vitamin E, are antioxidants that can only be synthesized by photosynthetic organisms, such as plants, in nature. They are found throughout plant tissues, with higher concentrations in photosynthetic tissues and seeds*(Niu et al., 2022)*. Tocopherols protect plants from oxygen toxicity by neutralizing lipid peroxy radicals, which cause lipid peroxidation. Among tocopherols, α-tocopherol is the most active and crucial in preventing lipid peroxidation. In addition to their antioxidant role in photosynthetic membranes and response to abiotic stresses, tocopherols impact various physiological processes in plants, including germination, growth, and leaf senescence. They may also play a role in signal transduction pathways and gene regulation. However, the subcellular localization and distribution of tocopherols, especially tocotrienols, require further investigation on an evolutionary scale. Tocotrienols, a type of tocochromanols, have gained attention recently, but their functions in plants are not fully understood. It remains uncertain whether tocotrienols accumulate in specific plant parts due to their antioxidant properties or other factors*(Munné-Bosch, 2002)*. Research on the impact of environmental factors on tocotrienol content in plants is also lacking in existing reviews..

Carotenoids

Carotenoids are lipophilic antioxidants that react with free radicals and singlet oxygen during photooxidative processes in plants. Carotenoids are also associated with lowering cancer risk, particularly skin and lung cancer. In humans, carotenoids damage cell membranes and protect cells from free radicals. Increasing carotenoids through diet can increase antioxidants and protective cells, which is beneficial for cancer prevention. Carotenoids are pigments that are produced by photosynthetic bacteria, algae, and plants that give bright colors to fruits, vegetables, and other plants. They are categorized into two groups: carotenes and xanthophylls, and some of these pigments have the ability to be converted into vitamin A, which is vital for human development and health (Stahl and Sies, 2003).

1.8 Methyljasmonate

Methyl jasmonate, or MeJA, is a plant hormone that is generated from jasmonic acid and regulates several physiological processes in plants, such as growth, development, secondary metabolism, and defense mechanisms against biotic and abiotic stresses. MeJA is a stress hormone that controls gene expression, photosynthetic activity, and growth. It is made from the acid α-linolenic *(Wasternack and Song, 2016).* It is an active form of jasmonic acid that operates in plants as a signaling molecule, affecting ripening of fruit, senescence of leaves, trichome and tuber formation, and growth of roots, shoots, and leaves as well as secondary metabolites. MeJA affects growth metrics, photosynthetic efficiency, gene expression, nitrogen homeostasis, and growth metrics *(Rodrigues Magalhães et al., 2023).* MeJA treatment has been shown to reduce photosynthetic efficiency in barley seedlings, change the expression of the aquaporin gene, and reduce the amount of nitrogen in plant leaves. Furthermore, MeJA treatment in germinated maize kernels can improve proline accumulation, carotenoid accumulation, and radical scavenging activity *(Wasternack and Song, 2016).*

1.9 Salicyclic Acid

One essential plant hormone that supports plant defense against pathogen invasion is salicylic acid (SA). It is made from isochorismate (IC), which is generated in Arabidopsis by the enzyme IC synthase (ICS). Recent research has determined the enzymes that convert IC to SA(*Mishra and Baek, 2021)*. Through interaction with the growth-regulating hormone auxin, SA represses growth-regulating genes while inducing defense genes against biotrophic pathogens. Auxin breakdown or plant immunity activation result from SA's regulation of auxin production, transport, and signaling. Plant immunity is positively regulated by the GH3.5 gene, which modifies SA-auxin crosstalk. PBS3, another GH3 protein, is likewise an IC-glutamate synthase, indicating that more GH3 family members might possibly be involved in SA biosynthesis*(Lefevere et al., 2020)*. The GH3.5 protein, which conjugates benzoic acid more effectively than SA, is used to create SA from benzoic acid. The production and regulation of SA must be understood in order to create plant disease management plans that work.

CHAPTER TWO

MATERIALS AND METHODS

2.1 Materials

2.1.1 Plant Sample

Fresh leaves of *Oil palm seedling (Elaeisguineensis Jacq.)* Collected in March, 2024 from a fallowed farm at Oluku, Ovia North East, Edo State.

2.1.2 Reagents

Ammonium Hydroxide(NH_4OH)

Acqueous Acetone Solution

Distilled Water

Carotenoids Standard

Lycopene Standard

Vitamin A Standard

Vitamin E Standard

Ascorbic Acid

Thiobarbituric acid

Trichloroacetic acid

Proline

Sodium chloride

Ethanol

Copper sulphate

Folin-Ciocalteau

Potassium sodium tartate

Sdium Dioxonitrate(III)

Auminium chloride (III)

Methanol

Phosphorus acid

2.1.3 Equipment/ Apparatus

The apparatus used in this study include:

Electrical weighing balance

Spectrophotometer

Waer bath

Centrifuge

High performance liquide chromatography

Pestle and mortar

Whatman filter paper

2.2 Methods

2.2.1 Preparation of Samples

The Oil palm seedling of six (6) months old in groups was used without washing and drying. 1g of the plant samples were weighed and grounded using a mortar and pestle with the use of 2ml of normal saline for easy removal and grinding process. After which the sample was poured into the test tube. The samples were then centrifuged and decanted using a pipette into different tubes for spectrophotometric readings.

2.2.2 Innoculation Of Oil Palm Seedling

Thielaviopsis Paradoxa was injected into seedlings using four different techniques in this study: cutting, infiltration, drip, and direct contact with cultured agar blocks. For fifteen days, the seedlings were kept in controlled environments using a thermohygrometer used to track humidity and temperature *(Gaitán-Chaparro et al., 2021)*. Mycelium blocks were positioned at the base of the third leaf, 0.1 milliliters of endoconidial solution was infiltrated, the leaf was cut using scissors dipped in the suspension, and one milliliter of endoconidial suspension was applied to the base as part of the inoculation procedure. At different concentrations of endoconidia, the illness was evaluated at 15 days following fungal inoculation, with each seedling being regarded as a duplicate. Three times through, the entire experiment was conducted. At different time points post-infection (hpi), such as 0, 24, 48, 72, 96, 120, 240, and 360 hours after the infection, the development of symptoms was evaluated. The purpose of the study was to assess T. paradoxa's effects on seedlings *(Gaitán-Chaparro et al., 2021)*

2.2.3 Experimental Design

A total of four(4) groups of oil palm seedling of 6 months old was used to determine the effect of methyljasmonate on infested oil palm seedling. These groups include;

Group 1: The normal control, these group was used as standard. These group is made up of oil palm seedling(*Elaeis guineensis*) which has not been infested or inoculate with the fungus *Thielaviopsis paradoxa*.

Group 2: The oil palm seedling(*Elaeis guineensis*) in this group has been infested with the pathogen(*Thielaviopsis paradoxa*). It is therefore referred to as the negative control.

Group 3: These group of oil palm seedling (*Elaeis guineensis*), is called the positive control. The elicitor (Methyljasmonate) is added to the oil palm seedling which has not been infested with the pathogen (*Thielaviopsis paradoxa*).

Group 4: This group is called the treatment group, it contains oil palm seedling (*Elaeis guineensis*) infested with the pathogen (*Thielaviopsis paradoxa*) and exposed to the elicitor methyljasmonate.

Group 4: These group of oil palm seedling (*Elaeis guineensis*), is called the positive control. The elicitor (Salicyclic Acid) is added to the oil palm seedling which has not been infested with the pathogen (*Thielaviopsis paradoxa*).

Group 5: This group is called the treatment group, it contains oil palm seedling (*Elaeis guineensis*) infested with the pathogen (*Thielaviopsis paradoxa*) and exposed to the elicitor Salicyclic Acid

S/N	GROUPS	NAMES	CONSTITUENTS
1	Group 1	Normal/Control	Oil Palm Seedling
2	Group 2	Negative Control	Oil Palm Seedling + Pathogen
3	Group 3	Positive Control	Oil Palm Seedling + Metja
4	Group 4	Treatment Group	Oil Palm Seedling+ Pathogen +Metj
5	Group 5	Posiive Control	Oil Palm Seedling + SA
6	Group 6	Treatment Control	Oil Palm Seedling+ Pathogen +SA

2.2. Quantfication Analysis

Carotenoids

Principle: Oil palm seedlings are treated with a suitable solvent (hexane or acetone) to extract the carotenoids. The content of carotenoids in the extract is then measured using spectrophotometer or high-performance liquid chromatography (HPLC). The assay relies on the measurement of carotenoids through their absorption of light at particular wavelengths. Oil palm seedlings are treated with a suitable solvent (hexane or acetone) to extract the carotenoids. The content of carotenoids in the extract is then measured using spectrophotometer or high-performance liquid chromatography (HPLC). The assay's foundation is carotenoids' ability to absorb light at particular wavelengths, which allows Oil palm seedlings to be treated with a suitable solvent (hexane or acetone) to extract the carotenoids. The content of carotenoids in the extract is then measured using spectrophotometer or high-performance liquid chromatography (HPLC). Carotenoids' ability to absorb light at particular wavelengths makes it possible to quantify them, which is the basis of the assay.

Procedures:

Carotenoids were extracted from oil palm seedlings using supercritical fluid chromatography or spectrophotometric analysis,the effects of different parameters like stationary phase, co-solvent, pressure, temperature, and flow rate were evaluated to optimize carotenoids seperationons.Carotenoids standards solutions were prepared for calibration.Afterwards, spectrophotometric method was utilized to quantify the carotenoids content in the extracted samples

Lycopene

Principle: An approach for figuring out how much of the carotenoid pigment lycopene is present in oil palm seedlings is the lycopene test. The assay's basic idea is based on lycopene's ability to absorb light at a particular wavelength, usually about 470 nm. Using an appropriate solvent, such as hexane or acetone, the lycopene is extracted from the seedling tissue. A spectrophotometer is then used to measure the extract's absorbance at the desired wavelength. After that, the lycopene concentration is determined using a standard curve that has been created by calculating the absorbance of lycopene concentrations that are known.

Procedure:

Lycopene was extracted from oil palm seedlings using supercritical fluid or spectrophotmetric analysis,the effects of different parameters like stationary phase, co-solvent, pressure, temperature, and flow rate were evaluated to optimize lycopene procedure.The lycopene standard solutions were prepared ranging from 0.04 to 1mM afterwards a reaction mix of ninhydrin in acetic and ethanol was utilized for lycopene quatification, the reaction mix was protected from light to prevent degradation of lycopene

Ascorbic Acid

Principle:

An approach for figuring out ascorbic acid content in oil palm seedlings is the ascorbic acid test. The assay's basic idea is that ascorbic acid reduces 2, 6-dichlorophenol in dophenol dye, causing the dye to change color from colorless to pink. The amount of ascorbic acid present in the sample is reflected in the intensity of the pink color.

Procedure:

One part of the oil palm seedling sample was mixed with four parts of 6% metaphosphoric acid (MPA) acidify the sample and stabilize the ascorbate ,the mixture was then frozen at -7- degrees until analysis,the frozen sample was then thawed at room temperature and centrifuged at 3,000 rpm.Afterwards, the supernatant was decanted and containing trisodium phosphate and dithiothreitol phosphate (DTT) to reduce dehydroascorbate to ascorbate, an internal standard(10-methyl uric acid) was added and the mixture wa re-acidified with 40% MPA to stabilize the ascorbate. The sample was filtered to remove insoluble material

Carotenes

Principle: Many fruits and vegetables have yellow, orange, and red hues due to a class of pigments called carotenes. Because they are precursors to vitamin A, which is necessary for immune system, growth, and vision, they are also significant for human health. The fruit of the oil palm plant contains large amounts of carotenes, which are crucial constituents of palm oil. The basic idea behind the carotene assay is that the molecules of carotene absorb light. The amount of carotenes present in the sample is directly correlated with the extract's absorbance. Using the aforementioned formula, one can determine the carotene content by measuring the absorbance at a particular wavelength (450 nm).

Procedures:

Collect fresh leaves from oil palm seedlings, the leaves should be healthy and free from any signs of disease or damage. About 100 mg of the fresh leaves were weighed and grouded into a fine powder in a mortar and pestle. Then 5 mL of acetone was added to the mortar and pestle and grouded until the mixture was homogeneous.The mixture was trasfered to a centrifuge tube and centrifuged at 3000 rpm for 5 minutes.Afterwards, supernatant was collected and transfered to a new tubes steps 4 and 5 was repeated twice to ensure complete extraction of the carotenes. The supernatants were combined and make up the volume to 10 mL with acetone.

Calculations.

$$Carotene\ content\ (mg/100\ g)\ =\ (A\ \times\ V\ \times\ 1000)\ /\ (W\ \times\ 100\ \times\ D)$$

Where; A is the absorbance of the extract, V is the volume of the extract (10 mL), W is the weight of the fresh leaves (100 mg), and D is the path length of the cuvette (1 cm).

Proline

Principle: One important osmolyte that builds up in plants under water stress is proline. It is essential for both osmotic correction and stress tolerance. Proline assays work on the basis of measuring the amount of proline present in plant tissues, especially when the plants are stressed by drought. Glutamate is used to make proline, which accumulates as a sign of stress in plants.

Procedures:

Oil palm seedlings were heated for 20 minutes, proline was extracted together with total amino acids, pigments, and souuble sugars by heating plant mateial twice with 80% ethanol and once with 50% ethanol.Ater that 20-50mg of extract was mixed with 70:30 ethanol: water mixture and left overnight at 4 degrees. The mixure was then centrifuged at

14000g for 5 minutes

Vitamin E

Principle: A class of fat-soluble antioxidants known as vitamins E is essential for shielding cells from oxidative damage. Vitamin E is necessary for several physiological functions and stress reactions in oil palm seedlings. The basic idea of vitamin E tests is to quantify the concentrations of various vitamin E forms, such as tocopherols and tocotrienols, which are strong antioxidants essential to human health.s method is considered the standard for analyzing protein content in various organic materials.

Procedure:

A gram (1g) of the initial material was weighed, and ground after which it was put in a test tube for 10 minutes with 20ml of nhexane, then centrifuged for 10 minutes. After the solution was filtered, 3ml of the filtrate were placed in duplicate dry test tubes and dried in a boiling water bath. A water bath was then used to boil 2ml of 0.5N alcoholic potassium hydroxide for 30 minutes after that, 3ml of n-hexane were added and given a good shake.After being moved into a different set of test tubes, the n-hexane evaporated until it was completely dry. Two milliliters (ml) of ethanol were added to the leftover.1ml of 0.2% ferric chloride in ethanol was added to another volume, one milliliter of 0.5% 1-dipyridyl in ethanol was then added, and then one milliliter of ethanol to bring the total to five milliliters. Absorbance was measured at 520 nm against the blank after the solution was mixed.

Vitamin A

Principle: Retinol, another name for vitamin A, is a substance that is vital to human health, especially for growth and vision. Vitamin A is found in oil palm seedlings as carotenoids, which are pigments that give the fruit its distinctive orange hue. β-carotene, the most prevalent carotenoid found in oil palm, has the ability to transform into vitamin A within the human body.

The basic idea behind vitamin A assays for oil palm seedlings are to quantify the amount of carotenoids, specifically β-carotene, present in the tissues of the seedlings. High-performance liquid chromatography (HPLC), which separates and measures the various carotenoids contained in the sample, is usually used for this.

Procedure:

One gram of the sample was weighed and ground after which it was put in a test tube for ten minutes with 20ml of hexane, after that, 3ml of the higher hexane extract were divided into two dry test tubes and allowed to evaporate until they were completely dry. Then, 2ml of 50% trichloroacetic acid (TCA) in chloroform were added, along with 0.2 ml of acetic anhydride chloroform reagent,at 620 nm, the absorbance was measured every 30 and 15 seconds.

2.3 BIOCHEMICAL ANALYSIS

1. Determination of Total sugar and Total Starch

Principle: The process of extracting and measuring the amount of soluble sugars in the tissues is the basis for the total sugar test used on oil palm seedlings. Usually, to do this, the sugars are extracted with a solvent (water or ethanol) and the sugar concentration in the extract is measured. The two most used techniques for determining sugar content are high-performance liquid chromatography (HPLC), which separates and quantifies the constituent sugars in the extract, and spectrophotometry, which gauges the light absorption by the sugar solution *(A. Sirait et al., 2020)*.The carbohydrate condition of oil palm seedlings is ascertained by testing them for total sugar, which is a crucial sign of their vitality and overall health. Numerous factors, including the stage of development, environmental circumstances, and cultural techniques, might influence the sugar content of the seedlings. Researchers can evaluate the nutritional health and stress tolerance of the seedlings by measuring the total sugar content *(Chalermpol et al., 2013)*.

Procedure:

Simple sugar extraction

Weighing a 5g sample, 20 mL of 80% ethanol was added. After refluxing for one to two hours, the material was filtered via extraction thimbles or filter paper. The first step was carried out twice more, extracting the carbohydrates for 30 minutes each time using 10 mL of 80% ethanol, a rotary evaporator was used to evaporate the alcohol.Aterwards, a portion of the aqueous was transferred to a 100 mL volumetric flask and diluted with distilled water to volume. filter paper was the used to filter the sample.Using distilled water as the extraction solvent, Steps 1 through 5 were repeated.

Total sugar content determination

Standard glucose stock solution was diluted in test tubes at the following volumes: 0, 200, 400, 600, 800, and 1000 µL. (Kindly see Table 1). Run the standard in duplicates,to provide each test tube a capacity of 1000 µL, distilled water was added, to make sure the fluid is evenly distributed, each test tube was shook. Pipetted and well shaken, 1000 µL of phenol solution (5%) was placed into each test tube. Afterwards, Each test tube was filled with 5.00 mL of strong sulfuric acid, and it was well shaken, after standing for 15 to 30 minutes, the optical density at 490 nm was determined using spectrophotometry.A typical calibration curve was created, plotting optical density against glucose concentration.

NB: The phenol-sulphuric method is also used for carbohydrate determination and starch determination.

2.Determination of Thiobabituric Acid

Principle: T-bars often referred to as thiobarbituric acid reactive substances (TBARS), are employed as a marker for lipid peroxidation, indicating oxidative stress in plant tissues. The amount of glucose is quantified using an appropriate technique, such as spectrophotometry. The T-bars test's basic idea is that thiobarbituric acid (TBA) and malondialdehyde (MDA), a consequence of lipid peroxidation, react to produce a colored complex that can be measured spectrophotometrically. Liquid chromatography with high performance (HPLC) *(Zolfagharnassab et al., 2022)*

Procedure:

Fresh leaves or other tissues were collected from oil palm seedlings, the plant tissues were grounded into a fine powder using a mortar and pestle.Approximately 1 g of the powdered plant material was weighed and transferred to a 25 mL test tube, 5 mL of the extraction solvent was added, which was either:100% glacial acetic acid (AA), 50% glacial acetic acid in water (AW) 0.01% butylated hydroxytoluene (BHT) was added to the solvent to prevent further oxidation,the mixture was shaken for 1 hour to extract the TBARS.The extract was filtered to remove any solid particles. Afterwards, the filtrate was centrifuged, if necessary, to obtain a clear supernatant. A 4.0 mM thiobarbituric acid (TBA) solution was freshly prepared in glacial acetic acid.From the MDA stock solution, a series of standard solutions with concentrations ranging from 0.1 to 1.0 mM were prepared.

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CHAPTER THREE

RESULTS

3.1 Results

Effect of methyljasmonate and salicylic acid on oil palm seedlings exposed to fungal pathogen

As shown in Tables 3.1 – 3.3, exposure of oil palm seedlings to fungal pathogen significantly reduced the levels of important plant molecules. However, application of Methyljasmonate and Salicylic acid enhanced the synthesis of important plant molecules ($p < 0.05$).

Table 3.1: Effect of methyljasmonate and salicylic acid on the concentrations of important biomolecules

Group	Total Sugar (mg/mL)	Proline (µg/mL)	Ascorbic Acid (mg/mL)
Normal Control	12.39 ± 0.68^a	40.14 ± 2.53^a	10.43 ± 0.62^a
Negative Control	5.19 ± 0.53^b	10.61 ± 0.81^b	2.84 ± 0.09^b
Methyljasmonate	8.31 ± 0.72^a	39.82 ± 1.52^a	11.16 ± 0.91^a
Fungi + Methyljasmonate	7.93 ± 0.86^a	28.96 ± 1.40^c	4.86 ± 0.52^c
Salicylic Acid	11.70 ± 0.91^a	41.16 ± 2.07^a	7.08 ± 0.59^a
Fungi + Salicylic Acid	9.80 ± 0.64^a	26.07 ± 1.06^c	6.84 ± 0.73^a

Data are concentrations of some plant biomolecules, and are expressed as mean ± SEM.

Values with different superscripts are significantly different ($p < 0.05$)

Values with the same superscripts are not significantly different ($p > 0.05$)

Table 3.2: Effect of methyljasmonate and salicylic acid on the concentrations of vitamins A and E.

Group	Vitamin A (µg/mL)	Vitamin E (mg/dL)
Normal Control	7.91 ± 0.51^a	50.05 ± 1.27^a
Negative Control	5.08 ± 0.31^b	31.02 ± 2.00^b
Methyljasmonate	9.52 ± 0.93^a	48.41 ± 0.96^a
Fungi + Methyljasmonate	8.14 ± 0.64^a	44.62 ± 1.58^a
Salicylic Acid	8.63 ± 0.00^a	47.00 ± 3.08^a
Fungi + Salicylic Acid	8.50 ± 0.42^a	59.12 ± 1.00^a

Data are concentrations of vitamins A and E, and are expressed as mean ± SEM.

Values with different superscripts are significantly different ($p < 0.05$)

Values with the same superscripts are not significantly different ($p > 0.05$)

Table 3.3: Effect of methyljasmonate and salicylic acid on the concentrations of carotenoids, lycopene and TBARS

Group	**Carotenoids (mg/g Extract)**	**Lycopene (mg/g Extract)**	**TBARS (%)**
Normal Control	102.21 ± 3.62^a	50.48 ± 2.11^a	60.17 ± 2.06^a
Negative Control	48.90 ± 1.09^b	20.95 ± 1.06^b	30.05 ± 1.08^b
Methyljasmonate	96.39 ± 2.16^a	50.31 ± 1.88^a	43.95 ± 2.72^a
Fungi + Methyljasmonate	75.64 ± 1.84^a	39.65 ± 1.62^a	55.73 ± 3.11^a
Salicylic Acid	109.35 ± 4.08^a	56.39 ± 2.01^a	51.94 ± 2.62^a
Fungi + Salicylic Acid	82.21 ± 2.73^a	36.93 ± 1.75^a	49.58 ± 1.96^a

Data are concentrations of carotenoids, lycopene and TBARS, and are expressed as mean ± SEM.

Values with different superscripts are significantly different ($p < 0.05$)

Values with the same superscripts are not significantly different ($p > 0.05$)

CHAPTER FOUR

DISCUSSION AND CONCLUSION

4.1 Discussion

This study investigated the effects of salicylic acid and methyljasmonate on fungal pathogen-exposed oil palm seedlings. Important plant biomolecules, vitamins A and E, carotenoids, lycopene, and TBARS (Thiobarbituric Acid Reactive Substances), which are markers of oxidative stress and lipid peroxidation, were the main focus of the study. The effects of salicylic acid and methyljasmonate on the amounts of important biomolecules are shown in Table 3.1. The negative control group shows the reduction in these biomolecules brought on by exposure to fungal pathogens, while the normal control group represents the baseline values in healthy seedlings. Comparing the methyljasmonate and salicylic acid treatments to the negative control, there is a trend of either preserving or raising the levels of proline, ascorbic acid, and total sugar, suggesting that these biomolecules were shielded aginst the pathogen-induced decline. The methyljasmonate-treated group exhibited a considerable increase in total sugar and proline concentrations when compared to the negative control group. This finding raises the possibility that methyljasmonate has a role in improving stress tolerance in oil palm seedlings.

The effects of salicylic acid and methyljasmonate on vitamin A and E concentrations are shown in Table 3.2. Both hormone treatments either maintain or increase the levels of these vitamins relative to the negative control, which is consistent with the trend seen in Table 3.1 and suggests a protective effect against fungal pathogen-induced depletion. Interestingly, compared to the negative control, the salicylic acid treatment group shows a considerable increase in vitamin E concentration, indicating a specialized role for salicylic acid in bolstering antioxidant defense systems in oil palm seedlings.

The effects of salicylic acid and methyljasmonate on TBARS, carotenoids, and lycopene levels are shown in Table 3.3. Both of the hormone treatments lessen the reduction in carotenoids and lycopene brought by exposure to fungal pathogens, which is in keeping with the results from earlier tables. Furthermore, there is a tendency for both therapies to lower TBARS levels, suggesting a defense against oxidative stress and lipid peroxidation. All things considered, the findings imply that salicylic acid and methyljasmonate therapies benefit oil palm seedlings exposed to fungal infections by boosting the synthesis of critical macromolecules, preserving antioxidant levels, and mitigating lipid peroxidation These results highlight the potential of salicylic acid and methyljasmonate as preventative measures against the harmful effects of fungal infections on the productivity and health of oil palms. The underlying molecular mechanisms of action of these hormones and their possible use in agricultural techniques to improve crop resilience against fungal infections could be the subject of future research.

A study on the application of methyl jasmonate and salicylic acid to mitigate drought-induced oxidative damages in French bean plants found that drought-stressed plants experienced a significant drop in their non-destructive chlorophyll index (SPAD) value. However, drought-stressed plants showed an increase in SPAD value with both single and combined administrations of MeJA and SA. The amounts of chlorophyll a, chlorophyll b, total chlorophyll, and carotenoids in leaves were reduced by 53%, 51%, 50%, and 48% under drought stress. The exogenous applications of MeJA and SA reduced the detrimental effects of drought stress, but the combined application significantly enhanced the amounts of carotenoid, total chlorophyll, chlorophyll a, and chlorophyll b *(Mohi-Ud-Din et al., 2021).* Hormone biosynthesis in oil palm roots is affected by SA and JA, with SA regulating ethylene biosynthesis and ROS production. Oil palm competes with G. boninense for disease prevention, whereas endophytic T. harzianum enhances nutrient status and transport in host plants *(Ho et al., 2016).* A study of temporal patterns and intercorrelations between physical antioxidant attributes and enzyme activities in apricot fruit reveals that

MeJA and SA improve plant defense against brown rot by reducing fungal growth, improving physical and antioxidant attributes, and increasing defense-related enzyme activity in apricot fruits during shelf-life storage conditions *(Ezzat et al., 2021).*These findings are comparable with those of the oil palm study, in which the use of MeJA and SA helped preserve levels of essential macromolecules, vitamins, carotenoids, and lowered oxidative stress markers in fungal pathogen-exposed seedlings. Both findings demonstrate the potential of these plant hormones.

4.2 **CONCLUSION**

Salicylic acid and methyljasmonate are found to be effective in protecting oil palm seedlings from fungal infections. These hormones can improve macromolecule production and preserve or increase antioxidant and vitamin levels, reducing the drop in proline, ascorbic acid, and total sugar concentrations caused by fungal pathogen exposure. They also support the preservation or elevation of vitamin A and E levels, assisting the antioxidant defense systems of oil palm seedlings. Additionally, they maintain carotenoids and lycopene concentrations while lowering lipid peroxidation. Understanding these processes can influence sustainable disease control techniques for oil palm farming

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