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ORIGINAL RESEARCH ARTICLE

Exploring the antifungal potential of *Allium sativum* and *Ocimum gratissimum* against post-harvest fungal pathogens in mango fruits

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ABSTRACT

Post-harvest spoilage of fruits and vegetables caused by fungal pathogens is a serious challenge to fruit production in many parts of the world. The study was conducted to evaluate the sensitivity of fungal pathogens associated with post-harvest rot of mango fruits to crude extracts from two edible plants, *Allium sativum* and *Ocimum gratissimum*, in the study area. Five different fungal isolates were isolated from diseased mango fruits collected from fruit stores in the study area and identified as *Aspergillus* spp. (M_1), *Rhizopus* spp. (M_2), *Fusarium* spp. (M_3), *Penicillium* spp. (M_4), *Fusarium* spp. (M_5), *Penicillium* spp. (M_6), *Aspergillus* spp. (M_7), and *Colletotrichum* spp. (M_8) using radial growth rate and morphological features of the mycelia. A constant concentration of each of the crude extracts was applied to the growth media containing the growing cultures of the fungal isolates. The radial extension of the colonies for each isolate was measured along pre-marked perpendicular axes on the base of the petri dish after 24 h, and this continued for 10–14 days. It was observed that *Rhizopus* spp., *Fusarium* spp., *Penicillium* spp., and *Colletotrichum* spp. had the least growth rate when treated with the extracts.

Keywords: *Allium sativum*; *Ocimum gratissimum*; fungal pathogens; mango fruits

1. Introduction

Mango (*Magifera indica* L.) is the seventh most popular fruit worldwide after oranges and grapes. Global production of mango fruits reaches 55–56 million tons, of which Nigeria is one of the major producers in Africa[1,2]. The fruits are rich sources of several important nutrients for humans; medicinally, the fruit is known to contain essential antioxidants such as polyphenols and mangiferin, which are of supreme health benefit to humans[3,4]. Mango production in Nigeria is seasonal, with certain varieties adapted to specific regions of the country. In commercial production, the fruits are harvested when they have matured prior to ripening, usually 3–4 months after the establishment of fruits[5]. As far as we know, agricultural production is constantly

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threatened by several biotic and abiotic factors, among which fungal and bacterial pathogens are responsible for a significant share of fruit and vegetable losses both on the field and after harvest[6].

Abdulkhair and Alghuthaymi[7] reported that the microbial pathogens causing crop loss are diverse and capable of invading many crops at various parts and growth stages. Poor storage facilities and post-harvest handling available in developing and undeveloped nations are major causes of pathogen invasion of harvested plant parts[8]. Infected materials are unfit for human consumption, resulting in a loss of market value and a reduction in farmers' income[9,10].

Fruits and vegetables are highly susceptible to invasion by microbial pathogens due to their high nutrient composition, moisture level, and location in areas with highly humid and warm climatic conditions[11,12]. Mango fruits prior to ripening undergo a series of physiological and biochemical processes, resulting in their increased susceptibility to post-harvest rot. Pathogens on the field may colonize unripe mango fruits, but symptoms and disease conditions are expressed upon harvest and ripening[11,13]. Fungal pathogens associated with post-harvest spoilage of mango fruits have been documented in several regions of the world[14]. For example, *Aspergillus* spp., *Rhizopus stolonifera*, *Fusarium* spp., *Penicillium* spp., and *Phoma* spp. are spoilage fungi earlier reported on rotten mango fruits in Nigeria[10,15]. Unfortunately, this information and the possibility of control methods using crude extracts from plants have not been satisfactorily documented in the study area. According to Sharma et al.[16], the occurrence of microbial pathogens varies across regions, plant types, seasons, and climatic conditions.

Chemical fungicides used in post-harvest preservation are effective, reliable, and quick in action. However, overdependence on chemical pesticides may result in pathogen resistance and several humans and animals' health-related problems[17]. Alternatively, post-harvest preservatives that are safe and available with fewer consequences are being evaluated. Plant-based pesticides are naturally accessible, and their potency and mode of action are continuously being studied by several researchers. Currently, long lists of plant species have been reported for their antifungal potency, some of which are medicinal, edible, and other wild plants species[17,18]. Other management methods capable of replacing chemical control methods have been identified, and several others are under evaluation[19].

Garlic (*Allium sativum*) and scent leaves (*Ocimum gratissimum*) are economically important crops from the families Amaryllidaceae and Lamiaceae, respectively. The plants are essential constituents of many delicacies in different areas of the world. Medicinally, health benefits associated with the direct consumption of these plants have been satisfactorily established in several literatures[20]. In addition to their nutritive and medicinal purposes, the plants have been evaluated for their insecticidal and antimicrobial potencies. For instance, oil extracts from *Alium sativum*, *Xylopia aethiopica*, and *Eucalyptus globolus* were significant in the control of field insect pests on leafy amaranths[17,21]. Scent leaf is a vegetable/spicy crop grown alongside other vegetables and horticultural crops in gardens and around homes in the study area, thus making it readily available and accessible. Despite the rich literature on these plants, information on their antifungal potency against post-harvest spoilage fungi of fruits has not been sufficiently documented in the study area. The study was therefore conducted to assess the potential of the crude extract from garlic bulbs and scent leaves to inhibit the growth and sporulation of post-harvest rot-causing fungi isolated from rotten mango fruits in Akwa Ibom State, Nigeria.

2. Materials and methods

2.1. Isolation of pathogenic fungi

Orange fruits showing rot symptoms were collected from fruit stores in Abak Main Market with coordinates of 5°0'11.8296" N and 7°46'27.372" E from March to June 2023. The fruits were surface sterilized following the procedures described by Borisade et al.[22]. About 50 mm of the infected fruit parts were aseptically incised using sterile blades, placed on chloramphenicol-adjusted potato dextrose agar (PDA), and incubated at ambient temperature for 3–5 days. The single spore isolation method[23] was used to maintain pure cultures of fungal isolates on standard agar (PDA + 0.02% chloramphenicol), which were then used for morphological characterization and pathogenicity assays in the study.

2.2. Growth characteristics, sporulation rates, and identification of fungal isolates

A 0.5-cm fungal mat from five-day-old growing plates of each isolate was placed at the center of a 9-mm petri dish containing standard agar (PDA + chloramphenicol) using a cork borer. The plates were sealed with parafilm and incubated at ambient temperature for 10–14 days. The growth rates were assessed by measuring the mycelial growth in two perpendicular directions[24]. X and Y lines were drawn across the center of the plates at 24 h after transferring the fungal mats into the new plates until the plates were covered by the mycelial growth of each isolate. Each fungal isolate was identified using macro- and micromorphological characteristics described in earlier studies by Ezeonuegbu et al.[25].

2.3. Pathogenicity assay

The ability of each fungal isolate to cause rot on healthy fruits was assessed. Clean and healthy orange fruits were inoculated with a 3-mm fungal mat from each isolate and incubated on a partially dampened paper roll in transparent plastic boxes for 5 days. The fruits were assessed for symptoms as highlighted by Ezeonuegbu et al.[25] at 48 h after inoculation (AI) and continued for 5 days. Pathogens that were able to induce fruit spoilage with similar symptoms to the original were re-isolated to demonstrate Koch's postulate.

2.4. Preparation of plant extracts

Exactly 20 g of clean and fresh scent leaves and garlic bulbs were properly washed in 75% ethanol, rinsed repeatedly in tap running water, and spread to dry on sterile absorbent papers. They were then crushed separately using an electronic blender. Each paste was then suspended in 10 mL of distiled water, vortexed for 90 s at a 2 h interval, successively for 6 h, and the filtrate was sieved into sterile storage bottles as crude extracts. Approximately 40 µL of each crude extract was dropped in five aliquots at five positions across the X and Y axes and the center of the plates containing standard agar (PDA + chloramphenicol). The plates were gently swirled against the lab bench to allow for even distribution of the extract prior to solidification. 5 mm-diameter agar discs from each growing isolate were placed at the center of the agar plates (containing the crude extracts), respectively, and incubated at ambient temperature for 10–14 days in triplicate. The growth of each fungal isolate was measured along each axis (X and Y), and the data obtained were subjected to ANOVA using R-statistic version 3.2 and mean separated using LSD.

3. Results

3.1. The mean growth rates of the fungus isolated from diseased mango fruits in the study

A total of eight fungal isolates were obtained from the diseased mango fruits in this study. The mean growth patterns of the eight fungal isolates labelled M_1, M_2, M_3, M_4, M_5, M_6, M_7, and M_8 isolated from diseased mango fruits are shown in **Table 1**. Isolates M_2, M_1, and M_6 had the highest mean growth rates along both axes (X and Y on plates), followed by isolates M_8, M_7, and M_4. The least mean value of the growth rates was recorded on isolates M_5 and M_3 (**Table 1**).

Table 1. Fungal pathogens isolated from diseased orange and their growth pattern (radial extension) along X and Y-axis on the petri dish.

S/N	Fungal isolates	Radial extension on petri dish along	
		X-axis	Y-axis
1	M_1	5.82 ± 2.68^{ab}	5.40 ± 2.82^{ab}
2	M_2	6.77 ± 2.41^{a}	6.71 ± 2.49^{a}
3	M_3	2.32 ± 1.63^{d}	2.17 ± 1.98^{c}
4	M_4	4.11 ± 2.47^{c}	3.80 ± 2.40^{b}
5	M_5	3.97 ± 2.82^{c}	3.73 ± 2.90^{b}
6	M_6	5.13 ± 2.61^{bc}	5.33 ± 2.69^{ab}
7	M_7	4.60 ± 2.91^{bc}	4.36 ± 3.07^{b}
8	M_8	4.65 ± 2.41^{bc}	4.28 ± 2.58^{b}

Mean values with similar letters are not statistically different from each other at 0.05 df, and mean values with different letters are statistically significant.

The diseased mango and sterilized portions of the diseased fruits are presented in **Figures 1** and **2**.

Figure 1. Diseased mango fruits for isolation.

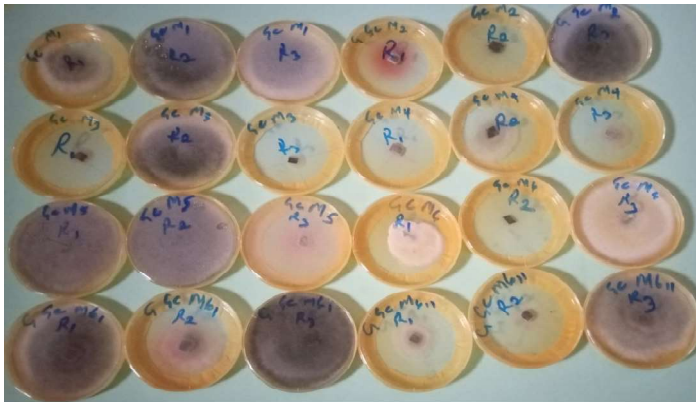

Figure 2. The eight isolated fungal pathogens inoculated with crude extracts of garlic (Gc) in triplicate five days after inoculation (5DAI).

3.2. Identification of the fungal pathogens isolated from the diseased mango fruits

The mean growth rates and morphological data on the nature of mycelial: fluffy, raised, spongy, and colour (**Figures 3, 4** and **5**) were used in the identification of the fungi isolated from the diseased fruits in line with documented literature. The isolated fungi were identified as: *Aspergillus* spp. (M_1), *Rhizopus* spp. (M_2), *Fusarium* spp. (M_3), *Penicillium* spp. (M_4), *Fusarium* spp. (M_5), *Penicillium* spp. (M_6), *Aspergillus* spp. (M_7), and *Colletotrichum* spp. (M_8), as described by Mailafia et al.[10].

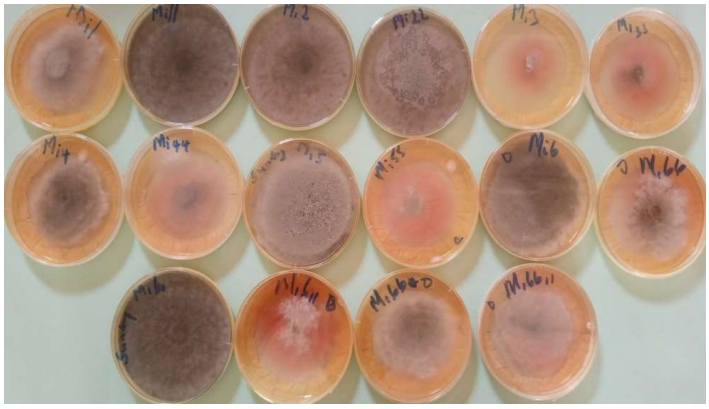

Figure 3. Eight pure isolates of the fungal pathogens isolated from the diseased mango fruits in two replicates labelled as (M_1, M_2, M_3, M_4, M_5, M_6, M_{6i}, and M_{6ii}).

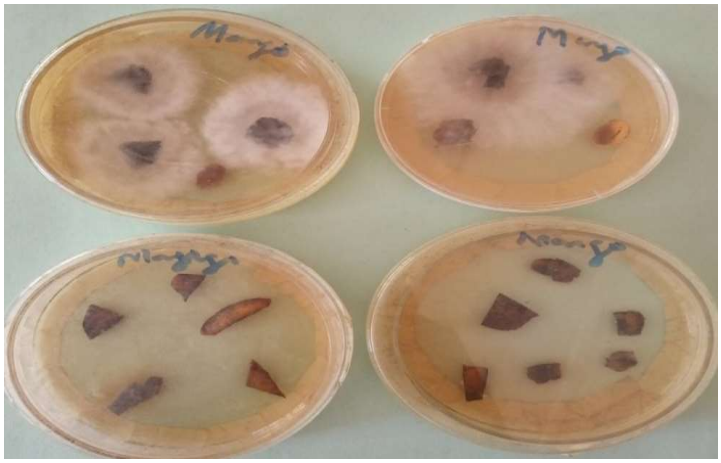

Figure 4. Early growth of fungal pathogen on sterilised portion of the diseased mango fruits inoculated on PDA agar.

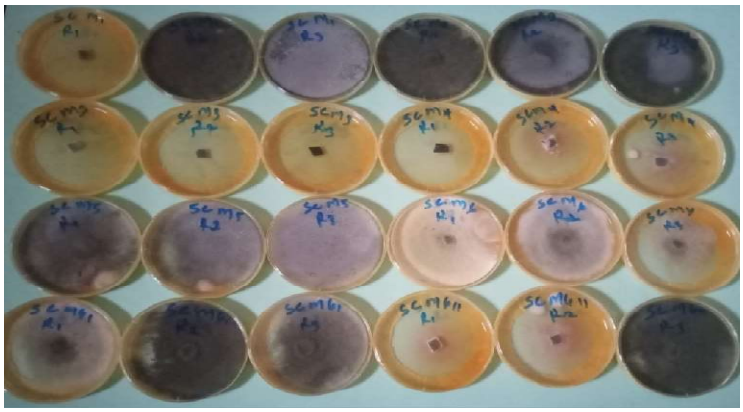

Figure 5. The eight isolated fungal pathogens inoculated with crude extracts of Scent leaf (Sc) leaves in triplicate at five days after inoculation (5DAI).

3.3. The mean growth rates of the isolated fungus inoculated with the crude extract of a fresh bulb of *Allium sativum* and leaves of *Ocimum gratissimum* plant

Upon inoculation of the isolated fungi with the extract, it was observed that *Aspergilus* (5.63 ± 2.92) and *Fusarium* (5.38 ± 3.02) had the highest mean growth rate on both axes, followed by *Penicillium* spp. (3.95 ± 3.02) and *Colletotrichum* spp. (3.83 ± 3.00) (**Table 2**). The lowest growth rates were recorded on *Rhizopus* spp. (2.66 ± 2.98) throughout the inoculation period (14 DAI), as shown in **Table 2**.

Table 2. The mean growth rates (radial extension) of the isolated fungus inoculated with crude extract of *Allium sativum* bulb along X and Y-axis on the petri dish.

S/N	Fungal isolates	Radial extension on petri dish along	
		X-axis	Y-axis
1	*Aspergillus* spp.	5.47 ± 2.95^{a}	5.70 ± 2.86^{a}
2	*Rhizopus* spp.	2.66 ± 2.98^{b}	2.75 ± 2.93^{c}
3	*Fusarium* spp.	3.53 ± 2.73^{b}	3.56 ± 2.66^{bc}
4	*Penicillium* spp.	3.20 ± 2.95^{b}	3.21 ± 2.62^{bc}
5	*Fusarium* spp.	5.38 ± 3.02^{a}	5.59 ± 2.93^{bc}
6	*Penicillium* spp.	3.95 ± 3.02^{b}	4.14 ± 2.99^{b}
7	*Aspergillus* spp.	5.63 ± 2.92^{a}	5.68 ± 2.91^{a}
8	*Colletotrichum* spp.	3.83 ± 3.00^{b}	3.84 ± 3.01^{bc}

Mean values with similar letters are not statistically different from each other at 0.05 df, and mean values with different letters are statistically significant.

It was also observed that *Rhizopus* spp., *Fusarium* spp., and *Aspergillus* spp. (6.84 ± 2.51, 6.35 ± 2.73, and 6.20 ± 2.70) inoculated with crude extract of the scent leaf plant had the highest radial growth rate throughout the sampling period, followed by *Penicillium* spp., *Aspergillus* spp., and *Colletotrichum* spp. (5.55 ± 2.86, 4.83 ± 3.54, and 4.49 ± 2.98), respectively (**Table 3**).

However, the least growth rate was observed on *Fusarium* and *Penicillium* (M_4) and *Fusarium* spp. (M_3) (**Table 3**).

Table 3. The mean growth rates of the isolated fungus inoculated with crude extract of fresh leaves of scent leaf (*Ocimum gratissimum*) plant.

S/N	Fungal isolates	Radial extension on petri dish along	
		X-axis	Y-axis
1	*Aspergillus* spp.	4.83 ± 3.54^{c}	5.14 ± 3.22^{bc}
2	*Rhizopus* spp.	6.84 ± 2.51^{a}	6.89 ± 2.40^{a}
3	*Fusarium* spp.	1.29 ± 0.52^{e}	1.40 ± 0.56^{e}
4	*Penicillium* spp.	3.01 ± 2.23^{d}	2.95 ± 2.11^{d}
5	*Fusarium* spp.	6.35 ± 2.73^{ab}	6.49 ± 2.59^{a}
6	*Penicillium* spp.	5.55 ± 2.86^{bc}	5.70 ± 2.76^{abc}
7	*Aspergillus* spp.	6.20 ± 2.70^{ab}	6.02 ± 2.64^{ab}
8	*Colletotrichum* spp.	4.49 ± 2.98^{c}	4.65 ± 2.93^{c}

Mean values with similar letters are not statistically different from each other at 0.05 df, and mean values with different letters are statistically significant.

4. Discussion

Fungal pathogens associated with post-harvest rots of mango fruits in the study area were *Aspergillus* spp., *Colletotrichum* spp., *Fusarium* spp., *Penicillium* spp., and *Rhizopus* spp. The fungi were identified using macro- and micro-morphological features of the mycelial earlier reported[10]. The results of the study were in conformity with findings by Mailafia et al.[10], who reported that *Aspergillus* spp., *Fusarium* spp., *Penicillium* spp., *Rhizopus* spp., and yeast are phytopathogenic fungi with a wide host range, including fruits and vegetable crops. *Aspergillus niger* and *Fusarium avenaceum* have been reported with a prevalence level of 65%–70% on several spoilt fruits in the north-central part of Nigeria[10]. Similarly, the anthracnose disease of mango

caused by *Collectotricum* spp. has been reported in several parts of the world, with prevalence and severity levels of over 70% and 40%, respectively[26]. According to Afsah-Hejri et al.[27], *Aspergillus niger*, other than causing post-harvest rots, produces mycotoxins and other toxic metabolites that can be harmful to humans and animals globally.

The biodiversity of fungal pathogens associated with post-harvest spoilage of fruits is increasing and varies across regions[16], and they are capable of invading several host crops[7] concomitantly, especially fruits with mechanical injuries. In the study area, post-harvest spoilage of fruits is increasingly high, as most fruits are imported from neighboring regions such as Benue, Taraba, and Nassarawa, where they may be exposed to mechanical injuries during transportation and handling. Going by this trend, post-harvest losses of fruits in the study area and in other areas sharing similar conditions may be higher than reported. In addition, fruit selling is a small business practiced by private individuals with little or no idea about post-harvest management practices that can reduce fruit losses. It is therefore necessary to present updated information on post-harvest spoilage fungi associated with major fruits such as mango in the study area[7,16].

Fusarium and *Aspergillus* spp. isolated in this study had the highest growth rate along both axes on the plates, which was suspected to be a contributing factor to their level of prevalence and resultant ability to cause rot of the fruit as observed in the pathogenicity test. According to Borisade et al.[28], sporulation rates and lag-time, as components of growth behavior, are important factors in the characterization of the virulence level of entomopathogenic fungi.

The biological method of using botanical fungicides in the management of fruit rot diseases caused by phyto-pathogenic fungi has been an increasing area of research interest for many researchers in recent times[29]. The two botanical extracts used in the study had varying inhibitory effects on the radial growth of the fungal isolates. The least growth rate was recorded on *Fusarium* isolate treated with extract from scent leaves and *Rhizopus* spp. treated with extract from garlic, respectively. This result indicated that crude extract from the scent leaf and garlic bulb could inhibit the sporulation rate of *Fusarium* and *Rhizopus* spp. associated with soft rot of mango fruits, respectively, in the study area, and the process of extraction was simple and could be adopted by fruit sellers upon further evaluations. The potency of garlic extracts in the control of phytopathogenic fungi has been reported in several literatures[30,31]. The findings from this study and further studies to investigate the effect of these extracts at varying concentrations on post-harvest rot-causing fungi associated with mango will form essential components to be incorporated into an integrated disease management program.

5. Conclusion

The study revealed that post-harvest spoilage of mango fruits is caused by an array of fungal pathogens, and their sensitivity to the plant's extracts used in the study was dependent on the level of susceptibility of the fungus to the extract. The result of these findings was baseline information for standardization of application rates and susceptibility assays that can be incorporated into integrated disease management strategies in the study area.

Author contributions

Conceptualization, OGO; methodology, UYI; formal analysis, UEA; investigation, UYI and AR; resources, MSZ; data curation, AR and HHA; writing—original draft preparation, OGO; writing—review and editing, OGO and UYI; visualization, UEA and HHA; supervision, OGO and HHA; funding acquisition, OGO. All authors have read and agreed to the published version of the manuscript.

Conflict of interest

The authors declare no conflict of interest.

References

1. Food and Agriculture Organization of the United Nations. Crops and livestock products. Available online: http://www.fao.org/faostat/en/#data/QCL (accessed on 30 September 2023).
2. Yadav D, Singh SP. Mango: History origin and distribution. *Journal of Pharmacognosy and Phytochemistry* 2017; 6(6): 1257–1262.
3. Kim H, Castellon-Chicas MJ, Arbizu S, et al. Mango (*Mangifera indica* L.) polyphenols: Anti-inflammatory intestinal microbial health benefits, and associated mechanisms of actions. *Molecules* 2021; 26(9): 2732. doi: 10.3390/molecules26092732
4. Gold-Smith F, Fernandez A, Bishop K. Mangiferin and cancer: Mechanisms of action. *Nutrients* 2016; 8(7): 396. doi: 10.3390/nu8070396
5. Yahia EHM. *Postharvest Handling of Mango: Technical Report*. Agricultural Technology Utilization and Transfer/RONCO; 1999.
6. Yahaya SM, Mardiyya AY. Review of post-harvest losses of fruits and vegetables. *Biomedical Journal of Scientific & Technical Research* 2019; 13(4): 10192–10200. doi: 10.26717/BJSTR.2019.13.002448
7. Abdulkhair WM, Alghuthaymi MA. Plant pathogens. In: Rigobelo EC (editor). *Plant Growth*. IntechOpen; 2016. pp. 49–59. doi: 10.5772/65325
8. Ladaniya M. *Citrus Fruit: Biology, Technology and Evaluation*, 1st ed. Academic Press; 2010. 886p.
9. Fatima N, Batool H, Sultana V, et al. Prevalence of post-harvest rot of vegetables and fruits in Karachi, Pakistan. *Pakistan Journal of Botany* 2009; 41(6): 3185–3190.
10. Mailafia S, Olabode HO, Osanupin R. Isolation and identification of fungi associated with spoilt fruits vended in Gwagwalada market, Abuja, Nigeria. *Veterinary World* 2017; 10(4): 393–397. doi: 10.14202/vetworld.2017.393-397
11. Karunanayake LC, Sinniah GD, Adikaram NK, Abayasekara CL. Alternatives to synthetic fungicides in controlling postharvest anthracnose and stem-end rot in mango. In: Proceedings of the III International Symposium on Postharvest Pathology: Using Science to Increase Food Availability; 7–11 June 2015; Bari, Italy. pp. 453–460. doi: 10.17660/ActaHortic.2016.1144.67
12. Prusky D, Shalom Y, Kobiler I, et al. The level of quiescent infection of Alternaria alternata in mango fruits at harvest determines the postharvest treatment applied for the control of rots during storage. *Postharvest Biology and Technology* 2002; 25(3): 339–347. doi: 10.1016/S0925-5214(01)00169-7
13. Pujari KH, Joshi MS, Shedge MS. Management of postharvest fruit rot of mango caused by *Colletotrichum gloeosporiodes*. In: Golding JB, Heyes JA, Toivonen PMA (editors). *XXIX International Horticultural Congress on Horticulture: Sustaining Lives, Livelihoods and Landscapes (IHC2014)*, Proceedings of International Symposia on Postharvest Knowledge for the Future and Consumer and Sensory Driven Improvements to Fruits and Nuts; 17–22 August 2014; Brisbane, Australia. International Society for Horticultural Science; 2016. pp. 215–218.
14. Goudarzi A, Samavi S, Amiri Mazraie M, Majidi Z. Fungal pathogens associated with pre- and post-harvest fruit rots of mango in southern Iran. *Journal of Phytopathology* 2021; 169(9): 545–555. doi: 10.1111/jph.13027
15. Chukunda FA, Baraka RE, Azubuike P. Post-harvest diseases of mango (*Mangifera indica* L.) fruits in port Harcourt, Nigeria. *Nigerian Journal of Mycology* 2020; 12(2): 162–173.
16. Sharma B, Singh BN, Dwivedi P, Rajawat MV. Interference of climate change on plant-microbe interaction: Present and future prospects. *Frontiers in Agronomy* 2022; 3. doi: 10.3389/fagro.2021.725804
17. Chen J, Shen Y, Chen C, Wan C. Inhibition of key citrus postharvest fungal strains by plant extracts in vitro and in vivo: A review. *Plants* 2019; 8(2): 26. doi: 10.3390/plants8020026
18. Oyetayo VO, Ogundare AO. Antifungal property of selected Nigerian medicinal plants. In: Razzaghi-Abyaneh M, Rai M (editors). *Antifungal Metabolites from Plants*. Springer; 2013. pp. 59–77. doi: 10.1007/978-3-642-38076-1_3
19. Kahramanoğlu İ, Nisar MF, Chen C, et al. Light: An alternative method for physical control of postharvest rotting caused by fungi of citrus fruit. *Journal of Food Quality* 2020; 2020: 8821346. doi: 10.1155/2020/8821346
20. Ugbogu OC, Emmanuel O, Agi GO, et al. A review on the traditional uses, phytochemistry, and pharmacological activities of clove basil (*Ocimum gratissimum* L.). *Heliyon* 2021; 7(11): e08404. doi: 10.1016/j.heliyon.2021.e08404
21. Borisade OA, Awodele SO, Uwaidem YI. Insect pest profile of leaf amaranth (*Amaranthus hybridus* L) and prevention herbivory using oil-based extracts of *Alium sativum* L, *Xylopia aethiopica* Dunal and *Eucalyptus globolus* L. *International Journal of Plant & Soil Science* 2019; 28(6): 1–9. doi: 10.9734/IJPSS/2019/v28i630130

22. Borisade OA, Uwaidem YI, Salami AE. Preliminary report on *Fusarium oxysporum* f. sp. *lycopersici* (Sensu lato) from some tomato producing agroecological areas in Southwestern Nigeria and susceptibility of F1-resistant tomato hybrid (F1-Lindo) to infection. *Annual Research & Review in Biology* 2017; 18(2): 1–9. doi: 10.9734/arrb/2017/34626
23. Choi YW, Hyde KD, Ho WWH. Single spore isolation of fungi. *Fungal Diversity* 1999; 3: 29–38.
24. Zhang CQ, Liu YH, Wu HM, et al. Baseline sensitivity of *Pestalotiopsis microspora*, which causes black spot disease on Chinese hickory (*Carya cathayensis*), to pyraclostrobin. *Crop Protection* 2012; 42: 256–259. doi: 10.1016/j.cropro.2012.07.018
25. Ezeonuegbu BA, Abdullahi MD, Whong CMZ, et al. Characterization and phylogeny of fungi isolated from industrial wastewater using multiple genes. *Scientific Reports* 2022; 12(1): 2094. doi: 10.1038/s41598-022-05820-9
26. Chala A, Getahun M, Alemayehu S, Tadesse M. Survey of mango anthracnose in southern Ethiopia and in-vitro screening of some essential oils against *Colletotrichum gloeosporioides*. *International Journal of Fruit Science* 2014; 14(2): 157–173.
27. Afsah-Hejri L, Jinap S, Hajeb P, et al. A review on mycotoxins in food and feed: Malaysia case study. *Comprehensive Reviews in Food Science and Food Safety* 2013; 12(6): 629–651. doi: 10.1111/1541-4337.12029
28. Borisade OA, Oso AA, Falade MJ. Interactions of some registered agrochemicals in Nigerian farming systems with entomopathogenic fungi, *Metarhizium anisopliae* and *Isaria farinose*. *Ife Journal of Science* 2016; 18(4): 949–961. doi: 10.1111/1541-4337.12029
29. Ladaniya MS. Commercial fresh citrus cultivars and producing countries. In: *Citrus Fruit: Biology, Technology and Evaluation*. Academic Press; 2008. pp. 13–65.
30. Mnayer D, Fabiano-Tixier AS, Petitcolas E, et al. Chemical composition, antibacterial and antioxidant activities of six essentials oils from the Alliaceae family. *Molecules* 2014; 19(12): 20034–20053. doi: 10.3390/molecules191220034
31. Satyal P, Craft JD, Dosoky NS, Setzer WN. The chemical compositions of the volatile oils of garlic (*Allium sativum*) and wild garlic (*Allium vineale*). *Foods* 2017; 6(8): 63. doi: 10.3390/foods6080063