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Combining plant extracts and hot water treatments for the management of postharvest mango anthracnose (*Colletotrichum gloeosporioides*)

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Mango anthracnose (*Colletotrichum gloeosporioides*), is a major postharvest disease that significantly impacts mango production and consumption globally. This study aimed to evaluate the combined effect of plant extracts and hot water treatments on the development of anthracnose and the preservation of postharvest quality in mangoes. In vitro tests using the food poisoning technique assessed the efficacy of acetone, ethyl acetate, hexane, and methanol extracts from nine different medicinal plant species against *C. gloeosporioides*. Among the tested extracts, *Ruta chalepensis* demonstrated the most pronounced effect, inhibiting mycelial growth by more than 85%. Extracts from *Allium sativum* also reduced mycelial growth by 80%. Furthermore, aqueous extracts from three effective plant species identified from the in vitro tests were combined with hot water treatments (HWT) to evaluate their impact on managing postharvest anthracnose. The combined treatments significantly ($p < 0.05$) affected anthracnose development on mangoes. The most substantial reduction in anthracnose severity was observed with treatments involving *R. chalepensis* and *A. sativum* at hot water temperatures of 55 °C and 50 °C. These treatments also significantly improved fruit marketability and maintained postharvest quality. In conclusion, the combined application of *A. sativum* with HWT at 55 °C and 50 °C, as well as *R. chalepensis* extract with HWT at the same temperatures, effectively reduced anthracnose intensity and improved mango fruit marketability by over 80% without affecting postharvest fruit quality indicators such as weight loss, TSS, pH, TA.

Keywords Antifungal, *Allium sativum*, Mango anthracnose, Plant extracts *Ruta chalepensis*

Mango (*Mangifera indica* L.) is a vital fruit crop in tropical and subtropical regions worldwide. It covers approximately 3.7 million hectares globally, ranking second among tropical fruit crops and fifth overall among all fruit crops, following citrus (*citrus* spp.), banana (*Musa* spp), grape (*Vitis vinifera*), and apple (*Malus domestica*)[1]. India leads as the largest producer of mangoes globally, followed by China, Mexico, Thailand, Indonesia, and Pakistan[2].The global mango production volume in 2021 was 58.3 million metric tons, and it is expected to rise by 1.9%, reaching 65 million metric tons by 2026[3].

Mangoes have a significant economic and social influence on people around the globe. They are a crucial part of the diet in numerous subtropical and tropical regions. It is one of the most widely grown among the fruit crops cultivated in Ethiopia preceded only by banana in terms of economic importance[4]. Mangoes are cultivated in various regions across the country, with western and eastern Ethiopia being significant production areas that together contribute over 50% of Ethiopia's total mango output[5]. Benishangul Gumuz Region is one of the potential areas for mango production in Ethiopia. The area has suitable agro ecological condition for production of various tropical and subtropical fruits including mango. The total land area covered by mango in the region during 2019/2020 cropping season is about 1,340.12 hectare with gross annual production of 69,408.47qt[6].

Mangoes are susceptible to various diseases throughout their growth cycle, from seedling in nurseries to storage and transit as mature fruit[7]. Anthracnose, caused by *Colletotrichum gloeosporioides* (teleomorph: *Glomerella cingulata*), is a major threat to global mango production and consumption, affecting productivity

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at both pre and postharvest stages[8,9]. This fungal disease is considered the most critical postharvest issue for mangoes, with potential economic losses ranging from 30 to 60%, and in severe cases, up to 100%, especially in wet or highly humid conditions[10].

Current strategies primarily rely on synthetic fungicides to control fungal decay of fruits. However, their high cost, residual risks to health and the environment, and the development of pathogen resistance due to frequent application are significant concerns[11]. Chitosan/silica nanocomposites (CSNs) have potential for controlling postharvest fruit decay by directly targeting pathogens and indirectly enhancing the host's antioxidant system, but they face challenges related to cost[12,13]. Thus, there is a pressing need to find practical, cost-effective, and environmentally friendly methods to prevent fungal decay in mango fruits. Hence, it is crucial to discover a method with the above features to prevent fungal decay in mangoes.

There is a rising interest in utilizing natural plant extracts as an alternative to synthetic fungicides[14]. This shift aligns with the principles of sustainable agriculture, aiming to reduce or eliminate the use of synthetic fungicides and other chemical treatments[15]. Identifying environmentally safe and biodegradable alternatives for managing postharvest diseases is crucial[16]. This includes exploring natural products, particularly those derived from plants, as substitutes for synthetic fungicides. Moreover, the increasing consumer demand for natural food products has spurred research into alternative antimicrobial agents within the food industry[17].

In recent years, considerable efforts have been directed towards discovering new antifungal compounds from natural sources for food preservation purposes. Several edible coating films from polysaccharides have been documented for postharvest preservation various fruit crops[18]. Numerous studies have also highlighted the antifungal properties of several edible botanical extracts[19]. These extracts have demonstrated effectiveness against various pathogenic fungal species affecting fruit crops. Botanical extracts are of natural origin, biodegradable, and generally do not leave toxic residues or by-products that could harm the environment[20]. The preservative properties of certain plant extracts have been recognized for centuries, prompting renewed interest in the antimicrobial capabilities of extracts from aromatic plants[21].

Plants contain a diverse array of secondary compounds such as phenols, flavonoids, quinones, tannins, essential oils, alkaloids, saponins, and sterols. These plant chemicals have various biological properties that can be harnessed, especially for disease management[22]. The combination of heat treatment and synthetic fungicides has been shown to enhance the efficacy of active ingredients in controlling decay, while reducing the amounts of chemicals required compared to traditional treatments[23]. A similar approach could be applied using plant extracts at elevated temperatures that do not degrade the bioactive compounds or harm the fruit. An integrated disease management strategy has emerged as the optimal approach for post-harvest management of plant diseases. Numerous reports document the combination of hot water treatment (HWT) with other methods, such as plant extracts, salts, bio control agent's organic and inorganic compounds[24–26] Therefore, the aim of this study was to investigate the combined effects of plant extracts and hot water treatment on controlling postharvest anthracnose in mangoes while preserving fruit quality.

Materials and methods

Collection and preparation of plant materials

Samples of plant materials with potential antifungal activity were collected from Assosa zone, Benishangul Gumuz Region, western Ethiopia on the basis of previous reports indicated in Table 1. The collection and identification of these materials followed the World Health Organization (WHO) guidelines on Good Agricultural and Collection Practices (GACP) for medicinal plants[27] and the guideline proposed by Wondafrash[28]. The specimens were submitted to the Benishangul Gumuz Regional state Agriculture Bureau, Biodiversity Conservation and Utilization Directorate for identification, which was conducted by Mr. Guteta Birhanu. The plant materials used in the experiments are preserved at the Biodiversity Conservation and Utilization Directorate's conservation site. The collected plant specimens used for the assays were washed in distilled water to remove surface contaminants and then dried under shade at room temperature (25 ± 2 °C). The dried plant parts were subsequently chopped, ground into a coarse powder, and stored in glass bottles for future use.

Extraction of plant material

Plant samples were individually extracted using maceration techniques. Each plant species' powders were extracted using methanol, acetone, ethyl acetate, and hexane (Loba Chemie, India). Fifty grams of powdered

No	Scientific name	Vernacular name	Plant parts used	Stage of Collection	References
1	*Allium sativum L.*	Garlic	Bulb	Maturity	[29]
2	*Calpurnia aurea* (Ait.) Benth.	Degati	Leaf	Vegetative	[30]
3	*Echinops kebnicho* Mesfin	Kbricho	Root	Maturity	[31]
4	*Justicia schimperiana* (Hochst. ex Nees)	Water willow	Leaf	Vegetative	[32]
5	*Nigella sativa L*	Black cumin	Leaf	Maturity	[33]
6	*Ocimum lamiifolium* Hochst. ex Benth.	Damakase	Leaf	Vegetative	[31]
7	*Oxytenanthera abyssinica* A. Rich Murno	Bamboo	Leaf	Vegetative	[34]
8	*Ruta chalepensis L.*	Rue	Leaf	Vegetative	[35]
9	*Sarcocephalus latifolius* (Sm.) E.A.Bruce	Hoary basil.	Root	Vegetative	[36]

Table 1. List of indigenous medicinal plant species that was assayed against *C. gloeosporioides*.

samples from each plant specimen were mixed with 250 mL of solvent. The extracts were filtered through folded filter papers into a 500 mL round-bottom flask and soaked in each solvent for approximately 48 h, following standard procedures[37]. The plant material suspensions were shaken on a horizontal shaker (Scilogex, United States) and kept at room temperature (25 ± 2 °C) in the dark. Subsequently, the solvent was evaporated to dryness using a rotary evaporator at 40 °C water bath temperature. The crude extracts of the plant species were then stored in the dark at 4 °C for future use.

Isolation of the target pathogen

The targeted pathogen, *C. gloeosporioides*, was isolated from diseased mango fruit (five fruit per sampling site) using standard procedures[38]. Small sections of symptomatic tissue with black, sunken lesions (measuring 3–5 mm^2), including adjacent healthy tissue, were excised and surface sterilized with 0.1% sodium hypochlorite for 2 min. The samples were then rinsed three times with sterile water. The sterilized tissue pieces were gently dried using sterile filter paper. Subsequently, four pieces of tissue were placed in a Petri dish containing freshly prepared Potato Dextrose Agar (PDA) (HiMedia Lab, India) medium under a laminar flow hood. Then the Petri dishes were then incubated for a week at 25 ± 2 °C. The growth of various fungi was monitored periodically. The culture obtained was further purified using the aerial mycelia tip technique. Identification of *Colletotrichum* spp. was based on morphological, cultural, and pathological characteristics. Pure cultures of the pathogen isolate were maintained in PDA culture tubes at 4 °C and served as stock cultures throughout the study.

Characterization of *Colletotrichum Species* associated with mango anthracnose

A total of 9 isolate namely ASCG1, ASCG2, ASCG3, BACG1, BACG2, BACG3, HOCG1, HOCG2 and HOCG3, representing three districts namely Bambasi, Assosa and Homosha from Assosa zone were characterized using cultural, morphological and pathological characteristics.

Colony growth

Growth rate was assessed by transferring 5 mm mycelial discs from seven-day-old cultures of *Colletotrichum* spp. isolates to the center of PDA plates under aseptic conditions, followed by incubation at 25 °C. The diameter of the colonies was measured in two perpendicular directions on the underside of Petri dishes at two-day intervals post-incubation. Growth rates were calculated after seven days using the formula described by Silva et al.[39].

$$Growthrate = \frac{\text{Growth observed on particular day (mm)} - \text{growth onprevious observation}}{2}$$

Colony color and density

The color of the developing mycelia on the obverse surface was determined using standard RGB (Red, Green and Blue) color charts. The extent of colony elevation above the agar surface for each isolate was evaluated and classified as flat, slightly raised, or raised[40].

Sporulation capacity

Isolates of *Colletotrichum species* were cultured on PDA, malt extract agar (MEA) (HiMedia Lab, India), mango leaf extract agar (MLEA), and Water Agar (WA) media (HiMedia Lab, India), and then incubated at 25 °C in darkness. After ten days, the colonies of each isolate were washed by flooding with 10 mL of sterile-distilled water and gently rubbed from the agar surface using a sterile scalpel. The resulting spore suspension was transferred into a 50 mL sterile beaker and stirred thoroughly for 10 to 15 min using a magnetic stirrer (Scilogex, United States) to extract the spores from the interwoven mycelia. Subsequently, the suspension was filtered into another sterile beaker through a double-layered cheese cloth. The concentration of conidia per milliliter was then counted and determined using a hemocytometer (Bürker, Germany).

Pathogenicity test

The pathogenicity of *Colletotrichum* spp. was tested on apparently healthy detached mango fruit of the Tommy Atkins variety. The fruits were surface-sterilized and placed in a sterile plastic box lined with tissue paper, followed by spraying with 10 mL of sterilized distilled water to maintain at least 95% relative humidity. The humidity was measure using Digital LCD Temperature Humidity Hygrometer with a 1.5 m Probe Cord (Model HTC, China). Inoculation was performed using the wound/drop method, fruits were pierced to a depth of 3 mm with a sterile needle in the middle portion, and then 20 µL of conidial suspension having a concentration of 1×10^5 condia/mL was applied onto the wound[41,42]. Control fruits were inoculated with 20 µl of sterile-distilled water. The inoculated samples were incubated in the plastic containers at 25 °C under controlled conditions. After 48 h, the plastic covers were removed, and the fruits were maintained at the same temperature. Each isolate was tested on a total of three fruits. The causative organisms from diseased parts were re-isolated on PDA, following the methods described for pathogen isolation. The characteristics of the re-isolated pathogens were compared with those of the original isolates.

In vitro evaluation of plant extracts

In vitro assay against radial mycelium growth was carried out to evaluate the antifungal properties of the plant extracts. All the test plant extracts were used to assay for mycelium growth inhibition against the fungal isolate *C. gloeosporioides*, using food poison technique[43]. About 9 mL of molten PDA was poured in sterilized Petri dishes along with 1 mL of plant extracts and plated. After complete solidification of the medium, 5-mm disc of seven-day-old culture of the test organism was transferred to the hole cut at the center of the Petri plate using sterile cork-borer. The plates were then incubated at room temperature (25 ± 2 °C). Control treatment were received the

exact amount of the extracting solvent. The experiment was arranged in completely randomized design (CRD) with three replications. Radial mycelium growth was measured in millimeters on the basis of linear dimensions for each colony when the actively growing tip of mycelial growth of the control Petri dish reaches the edges of the PDA medium and the percentage inhibition of radial growth was calculated following the method employed by Deressa et al.[44] using the formula.

$$Inhibition(\%) = \frac{C - T}{C} \times 100$$

where C is the average increase in mycelial growth in control plate and T is the average increase in mycelial growth in the treatment plate.

In vivo evaluation of plant extracts and hot water treatment

Three plant extracts namely *A. sativum R. chalepensis S. latifolius*, that showed prominent antifungal effect against *C. gloeosporioides.in vitro* were further evaluated in combination with hot water treatment (HWT) to manage the diseases on harvested mango fruits. The experiment used healthy (undamaged) mangoes at physiological maturity (mature green mangoes) of Local (Hindi) Mango cultivar that were uniform in size and color. Mango fruits were surface-sterilized by dipping them in a 1% sodium hypochlorite solution for 3 min, then rinsed three times with sterile distilled water and air-dried at room temperature (24 ± 2 °C) on an alcohol-disinfected shelf. Following disinfestation, the fruits were immersed in a plastic jar containing a *C. gloeosporioides.* spore suspension at a concentration of 1×10^6 conidia/mL for 5 min, the concentrations of the spores were determined using hemocytometer, and then covered with a plastic sheet[45] to maintain high relative humidity. After 15 h of incubation, during which the conidia germinated, the plastic sheets were removed, and the mangoes were submerged for 5 min in 20% (w/v) aqueous plant extracts, which were maintained in a water bath at a temperature of 24 °C, 50 °C, and 55 °C. Mangoes dipped in sterile distilled water at room temperature served as the untreated control. After treatment the fruit we stored at room temperature with a relative humidity of above 85%. Five fruit were used for each treatment, and the experiment was designed as a CRD with factorial combinations of four levels of plant extracts (Three plant extract and one control)and three level dipping temperatures (50 °C, 55 °C and room temperature), replicated three times.

Data collected

Data on disease incidence and severity were recorded to evaluate the effect of combined application plant extracts and hot water treatment antifungal activity of the botanicals under artificial inoculation, with assessments made at 48 h intervals from the appearance of symptoms until the control fruits reached 100% unmarketability. Disease incidence was calculated as the percentage of mango fruits exhibiting any symptoms of anthracnose, using the following formula:

$$\text{Disease incidence (DI)} = \frac{\text{Number of mango fruit showing anthracnose disase symptom}}{\text{Total number of mango fruit assesed}} \times 100$$

Severity of anthracnose on each fruit were scored as approximate percentage of the fruit area covered by lesion falling into a particular category using 1–5 scale according to Corkidi et al.[46]. Where 1 = infected area 0–1% no disease 2 = 1–5%, slight diseases; 3 = 6–9%, moderate disease; 4 = 10–49%, severe disease and 5 = 50–100%; very severe disease.

Postharvest quality of mango fruit

Marketability of mango fruit

Data on the proportion of marketable and unmarketable fruits were recorded at the time of disease assessment following the procedure of Dessalegn et al.[26]. Marketable fruits were evaluated at edible stage of the fruit based on criteria such as the extent of visible lesions, shriveling, smoothness, and shininess of the fruit surface. The determination of marketable fruits throughout the experiment was conducted using the following formula:

$$\text{Marketability of mango fruit (\%)} = \frac{\text{Number of marketable fruit}}{\text{Total number of fruit}} \times 100$$

Determination of weight loss (%WL)

The initial weight of the fruits was measured before applying the treatments and storing them at room temperature. Subsequently, the weights of the fruits were recorded before treatment application and when the fruit reach edible stage stored at room temperature (25 ± 2 °C). The calculation of weight loss was conducted according to the standard method described by Jongsri et al.[47], using the following formula:

$$\text{PWL (\%)} = \frac{\text{Average loss in fruit weight}}{\text{Average initial fruit weight}} \times 100$$

where: PWL is the physiological weight loss.

Measurement of total soluble solids (TSS)
Total soluble solids (TSS) were measured using a hand-held refractometer (Hanna Instruments, Italy) with a range of 0 to 32 brix and a resolution of 0.20 brix. The TSS content was determined by placing 1 to 2 drops of clear juice on the prism, following the procedure outlined by Dessalegn et al.[26]. After each measurement, the prism plate of the refractometer was cleaned with distilled water and dried using a soft tissue. Prior to measurements, the refractometer was calibrated using distilled water (0% TSS).

Measurement of titratable acidity (TA)
The titratable acidity (TA) was determined following the procedure outlined by Maul et al.[48]. TA, expressed as percent citric acid, was obtained by titrating 10 mL of mango juice to pH 8.2 using 0.1 M sodium hydroxide (NaOH) (Loba Chemie, India). The TA was then calculated using the following formula:

$$\mathrm{TA} = \frac{\text{titre X 0.1N NaOH X 0.67 x 100}}{1000} \times 100$$

Measurement of pH
The mango juice was extracted from the sample using a juice extractor, and the clear juice obtained was used for the analysis. The pH of the juice was measured using a pH meter (Hanna Instruments, Italy).

Statistical analysis
Data were subjected to analysis of variance (ANOVA) using SAS V 9.0 software package. Percentage data for conidia germination and disease incidence were arc sine transformed prior to analysis. Mean comparisons were conducted using the Least Significant Difference (LSD) test at 5% probability level.

Results
Isolation of the target pathogen
A total of 27 isolates of *C. gloeosporioides* were obtained from symptomatic samples collected from three districts of Assosa Zone, western Ethiopia. Pathogen identification was conducted based on morphological, cultural, and pathological characterization, following the description of *Colletotrichum* species[37]. Isolates of *C. gloeosporioides* exhibited colonies characterized by abundant whitish septate and hyaline aerial mycelium, along with slimy pinkish spore masses that were acervulate. Colony texture was categorized as appressed with sparse aerial mycelium, flocculose with raised and slightly dense aerial mycelium, or floccose with raised and dense aerial mycelium. Colony color varied and was described as white, grey, or pink, while colony shape could be uniform with smooth edges or irregular with rough edges.

Cultural characteristics
Cultural characteristics of *C. gloeosporioides* isolated from diseased mango fruits were observed on PDA, MEA, MLEA, and WA media. Based on colony color, the isolates were categorized into two morphological types. Most isolates exhibited colony colors such as white, white-gray, pink, and gray on both the upper and lower sides of the plates. The reverse side of the plates revealed a range of colors including light orange, light grey, light greenish, light whitish, grey, and dark grey (Fig. 1). Colony shape varied and was described as either uniform with smooth edges, irregular with rough edges, or banded with sectors consisting of thin expansive mycelium.

The study findings indicated that different culture media affect the growth characteristics of *C. gloeosporioides* isolates. All media tested namely PDA, MEA, MLEA, and WA, supported the mycelial growth of all *Colletorichum* spp. isolates. The average colony diameter ranged from 5.51 to 11.5 mm in Potato Dextrose Agar Medium 4.98–13.37 mm in Malt Extract agar Medium, 3.94–10.83 mm in Mango Leaf Extract Agar Medium and 1.65–5.83 mm in water agar (Table 2). Among the *C. gloeosporioides* isolates, ASCG3 exhibited the fastest growth rate across all media. Of the media tested, MLEA showed the highest daily growth rate, averaging 7.94 mm.

Morphological characteristics
The present study reveals that conidial production of *C. gloeosporioides* isolates varied across the tested culture media (Table 3). MEA yielded the highest conidial production, with an average of 3.54×10^7 conidia/mL, followed by Potato Dextrose Agar with 2.51×10^7 conidia/mL. No conidia were produced in Water Agar or Mango Leaf Extract Agar. Notably, the ASCG3 isolate from Assoa District exhibited the highest conidial production, with 5.33×10^7 conidia/ mL on PDA and 4.89×10^7 conidia/mL on MEA. Other isolates produced conidia ranging from 2.42×10^7 to 3.96×10^7 conidia/mL on PDA and from 2.27×10^7 to 8.48×10^7 conidia/mL on MEA medium (Table 3).

Pathogenicity tests
Symptoms characteristic of anthracnose caused by *Colletotrichum* spp. were observed at the inoculation site after 5 days of incubation. The fungi demonstrated infectivity, manifesting as black, sunken lesions on physiologically matured mango fruit (Fig. 2). These symptoms closely resembled those seen in naturally infected fruits. Under optimal moist conditions, the fungus developed acervuli, occasionally with concentric rings, and produced pinkish conidia in large masses. Severely affected fruits typically turned black, either rotting or shriveling and becoming mummified.

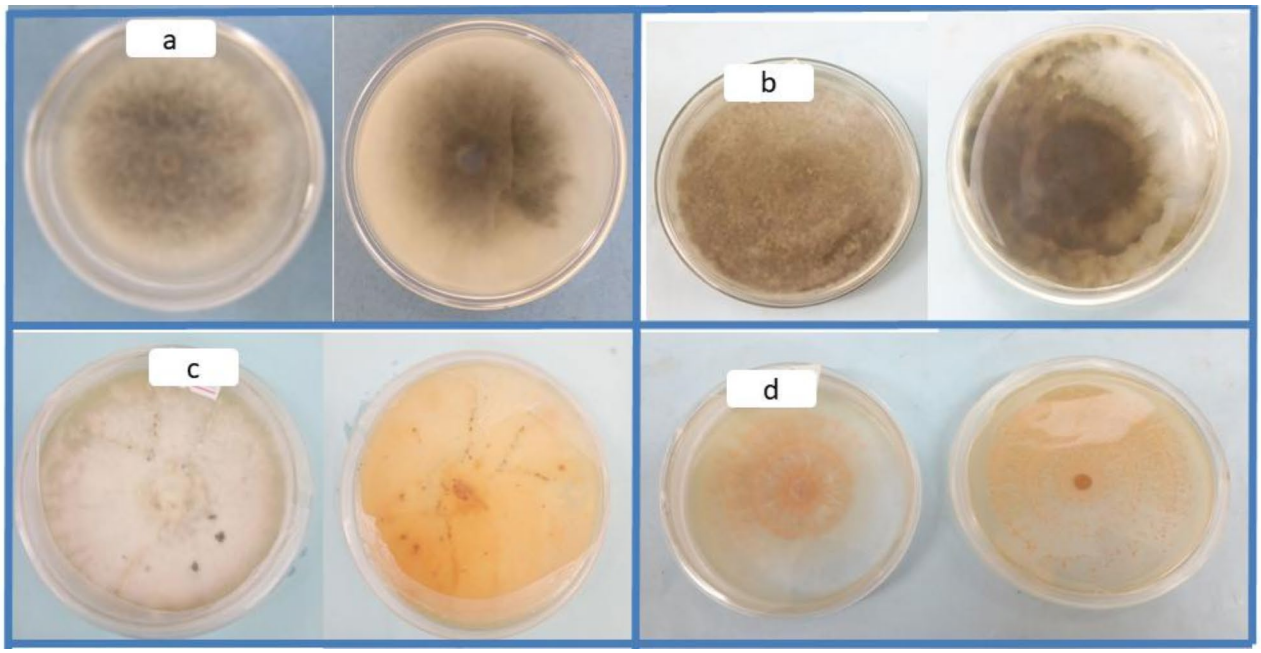

Fig. 1. Colony color of *Colletotrichum* spp. associated with anthracnose of mango (**a**) Light gray color on upper and reverse side of the plate (**b**) gray in the upper and light greenish in the reverse side (**c**) white color in the upper side and light orange in the reverse side (**d**) light orange on both sides with conidia mass.

Isolates	Culture media			
	PDA	MEA	MLEA	WA
ASCG1	9.08 ± 0.13^{b}	8.26 ± 0.19^{b}	$8.45 \pm 0.06^{b-d}$	1.73 ± 0.04^{d}
ASCG2	7.77 ± 0.17^{cd}	7.05 ± 0.17^{d}	8.77 ± 0.26^{bc}	2.48 ± 0.09^{c}
ASCG3	11.5 ± 0.22^{a}	13.37 ± 0.13^{a}	10.83 ± 0.37^{a}	5.83 ± 0.32^{a}
BACG4	6.84 ± 0.08^{de}	4.98 ± 0.31^{e}	$7.84 \pm 0.25^{c-e}$	1.84 ± 0.16^{d}
BACG5	5.51 ± 0.29^{f}	5.38 ± 0.32^{e}	3.94 ± 0.78^{f}	1.65 ± 0.03^{d}
BACG6	6.29 ± 0.17^{ef}	7.47 ± 0.19^{cd}	7.43 ± 0.10^{de}	1.75 ± 0.05^{d}
HOCG7	6.87 ± 0.49^{de}	7.05 ± 0.32^{d}	7.53 ± 0.30^{de}	5.44 ± 0.11^{b}
HOCG8	8.00 ± 0.72^{c}	7.94 ± 0.19^{bc}	7.14 ± 0.36^{e}	1.76 ± 0.08^{d}
HOCG9	$7.27 \pm 0.18^{c-e}$	8.17 ± 0.38^{bc}	9.53 ± 0.34^{b}	1.70 ± 0.06^{d}
LSD (0.05)	0.99	0.77	1.09	0.39
CV (%)	7.52	5.80	8.04	8.55

Table 2. Effect of different growth solid media on average daily mycelia growth diameter (mm) of *Colletotrichum* spp isolates. Means within the same column followed by the same letter are not significantly different at $p < 0.05$ according to Fisher's least significant difference (LSD) test.

In vitro antifungal activity of extracts from selected medicinal plant species against *C. gloeosporeoides*

All of the plant extracts significantly ($p < 0.05$) affect the growth *C. gloeosporioides* compared to the control treatment (Table 4). Methanol extracts from *R. chalepensis* and *A sativum* showed prominent inhibition of growth in *C. gloeosporioides*, with a inhibition percentage of 83.7% and 78.14%, respectively, compared to the control treatment i.e. methanol only (Table 4). Similarly, acetone, ethyl acetate, and hexane extracts from these plants significantly reduced the growth rate of the test fungi by 80%. Extracts from *S. latifolius* and *N. sativa* also exhibited notable antifungal activity against *C. gloeosporioides*, inhibiting growth to below 27 mm, with the ethyl acetate extract from *S. latifolius* reducing growth to below 19 mm.

Other plant extracts also demonstrated good antifungal effects, with mean growth ranges of 30 mm to 43 mm for methanol extracts, 30.67 mm to 40.67 mm for acetone extracts, 26.00 to 36.67 mm for ethyl acetate extracts, and 28.67 mm to 41 mm for hexane extracts Based on the combined mean inhibitory effects of these plant extracts, they can be ranked in decreasing order of efficacy as follows: *R. chalepensis* > *A. sativum* > *S. latifolius* > *C. aurea* > *J. schimperiana* > *E. kebericho* > *O. abyssinica* > *O. lamiifolium*. Three plant species namely

Isolates	Culture media			
	PDA	MEA	MLEA	WA
ASCG1	2.42×10^7	8.48×10^7	–	–
ASCG2	3.96×10^7	4.47×10^7	–	–
ASCG3	5.33×10^7	4.89×10^7	–	–
BACG4	2.66×10^7	2.27×10^7	–	–
BACG5	2.02×10^7	1.55×10^7	–	–
BACG6	–	–	–	–
HOCG7	–	2.56×10^7	–	–
HOCG8	3.45×10^7	4.22×10^7	–	–
HOCG9	2.74×10^7	3.40×10^7	–	–
Mean	2.51×10^7	3.54×10^7	–	–

Table 3. Sporulation capacity of *C. Gloeosporioides* isolates under different culture media. *PDA* Potato Dextrose Agar, *MEA* Malt extract Agar, *MLEA* Mango Leaf Extract Agar, *WA* Water Agar.

R. chalepensis, *A. sativum* and *S. latifolius* were further evaluated in combination with hot water treatment for evaluating their antifungal effect on harvested mango fruit.

Effect of plant extracts and hot water treatment on anthracnose development

Disease incidence

The effect of combined application of plant extract and hot water treatment on the development of anthracnose on artificially infected mango fruit is presented in Fig. 3; Table 5. The combined application of plant extracts and hot water treatment significantly ($p < 0.05$) decreased anthracnose incidence. The most substantial reduction was observed in fruits immersed in *A. sativum* extracts at 55 °C, resulting in a mean incidence of 26.67% (Table 5).

Extracts from *A. sativum* at 50 °C and *R. chalepensis* at 50 °C also exhibited notable antifungal effects, reducing disease incidence to below 34%, compared to 100% in the control fruit after 12 days incubation from inoculation. In contrast, extracts from *S. latifolius* showed relatively modest effectiveness in reducing anthracnose incidence on harvested fruit.

The treatments also significantly ($p < 0.05$) influenced the onset of disease symptoms on artificially inoculated mango fruit (Fig. 3). Disease symptoms appeared 4 days after inoculation in the control treatment, but this onset was significantly delayed to 6 days by dipping the fruit in *R. chalepensis* at 55 °C, *R. chalepensis* at 50 °C, *A. sativum* at 55 °C, and *A. sativum* at 50 °C. All the treatments also demonstrated reduction on the development of anthracnose on harvested mango fruit as compared to the untreated control (Table 5).

Disease severity

The integrated application of plant extracts and hot water treatment significantly ($P < 0.05$) influenced the development of anthracnose disease on mango (Fig. 4; Table 6). The most substantial reduction in anthracnose severity, to 1.33, was observed when the fruit was treated with *R. chalepensis* and *A. sativum*, both at a hot water temperature of 55 °C. Treating the fruit with *A. sativum* at 50 °C also reduced severity to 1.6, with no statistical difference compared to the previous treatment. Additionally, the extract from *R. chalepensis* at 50 °C showed effective reduction of anthracnose levels to 1.8 in artificially inoculated mango fruit. In contrast, the extract from *S. latifolius* had a relatively modest effect (Severity of 2.27–3.13) on reducing anthracnose development on harvested fruit.

Overall, treating the fruit with a combined application of plant extracts and hot water significantly reduced both the onset and progression of anthracnose on mangoes. Our study also indicates that the water temperature during treatment influenced the effectiveness of the extracts. Generally, higher temperatures, particularly 55 °C, resulted in the highest reduction of anthracnose severity when combined with all plant extracts (Table 6).

Effect of integrated application of plant extracts and hot water treatment on maintaining the postharvest quality of mango fruit

Fruit marketability

This experiment indicates that combined application of plant extracts and hot water treatment had significant ($p < 0.05$) effect on marketability of mango fruit compared to control treatment (Fig. 5; Table 7). All of the fruit (100%) in the control treatment were unmarketable; 12 days after inoculation, this level was significantly improved by treatment of the fruit with application of plant extracts and hot water treatment to harvested mango fruits. The maximum marketable (86.67%) fruits were recorded with application of *A. sativum* at 55 °C Similarly postharvest dipping of the mango fruit with *R. chalepensis* at 55 °C and 50 °C, and *A. sativum* extract at 50 °C also improves the marketability to 80.00–83.33%, which was statistically at par with the former treatment. The experiment indicates that the combined application of hot water treatment and plant extracts significantly improved the marketability percentage of the mangoes compared to the control group that did not receive any treatment.

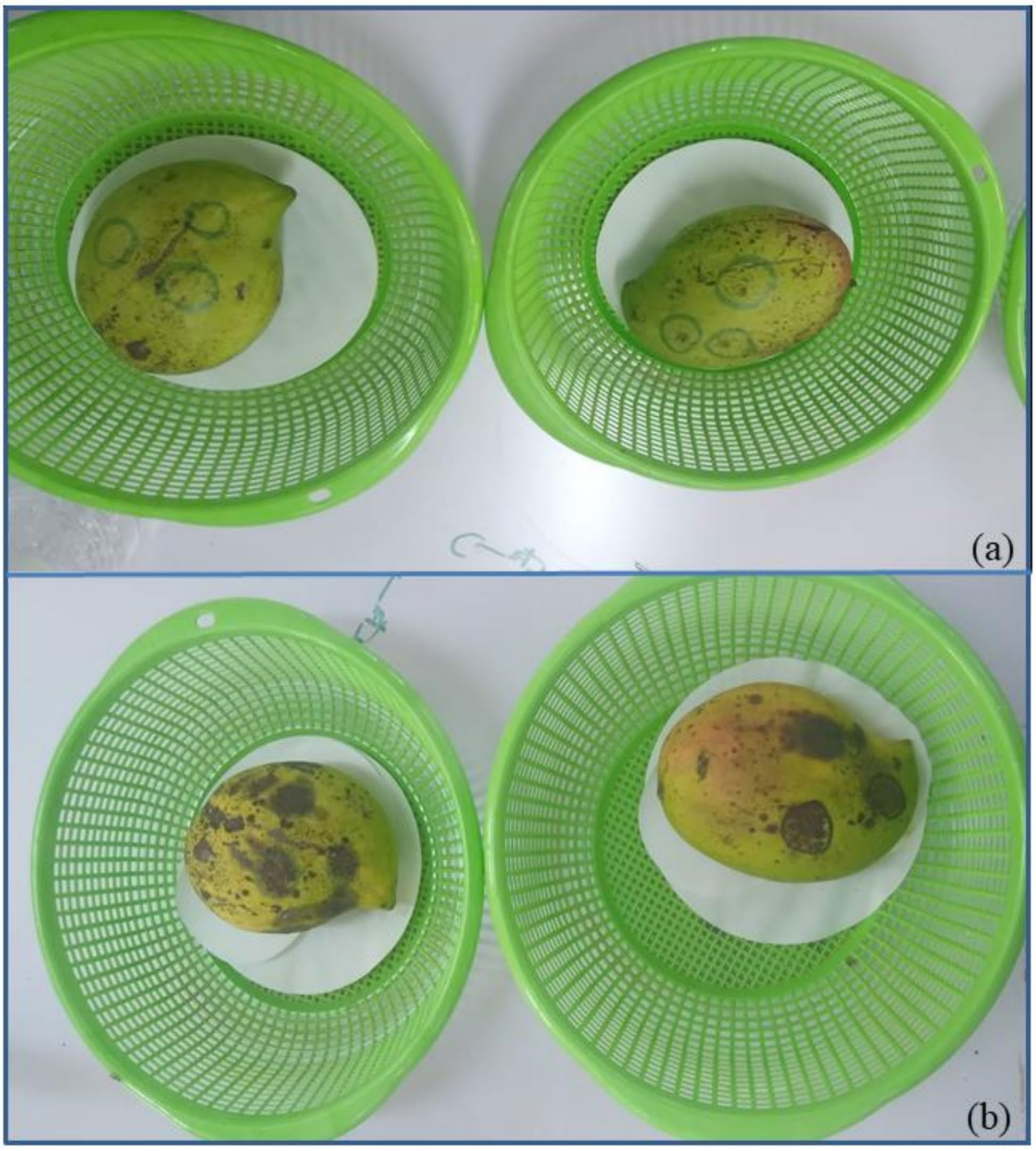

Fig. 2. Pathogenicity of *C. gloeosporioides* isolates on artificially inoculated mango fruit (a) fruit up on inoculation (b) fruits incubated for 10 days after inoculation.

Weight loss of mango fruit

The integrated application of plant extracts and hot water treatment significant difference at ($p < 0.05$) There were more differences in the weight loss of untreated mango (control) than other treated mango as shown in (Table 8).

The treatments showed a significant effect ($p < 0.05$) on reducing postharvest weight loss of mango fruit. The control sample exhibited a faster weight loss compared to the others. After 12 days of storage, *A. sativum* combined with 55 °C recorded the lowest weight loss at 3.67 g, followed by *R. chalepensis* combined with 55 °C, which had a weight loss of 4.33 g, compared to the control's 18.33 g. Additionally, *S. latifolius* at 55 °C, *S. latifolius* combined with 50 °C and 24 °C, as well as *R. chalepensis* and *A. sativum* combined with 24 °C, also experienced higher weight loss compared to the other treatments.

Plant species	Extracting solvent							
	Methanol		Acetone		Ethyl acetate		Hexane	
	Colony growth	%Inhibition	Colony growth	%Inhibition	Colony growth	%Inhibition	Colony growth	%Inhibition
A. sativum	19.67 ± 0.33^{i}	78.14 ± 0.37^{b}	17.67 ± 0.33^{i}	80.37 ± 0.37^{b}	13.33 ± 0.33^{g}	85.19 ± 0.37^{b}	15.33 ± 0.67^{i}	82.97 ± 0.74^{b}
R. chalepensis	14.67 ± 0.33^{j}	83.70 ± 0.37^{a}	12.67 ± 0.33^{j}	85.92 ± 0.37^{a}	10.00 ± 0.00^{h}	88.89 ± 0.00^{a}	12.33 ± 0.33^{j}	86.30 ± 0.37^{a}
S. latifolius	24.00 ± 0.58^{h}	73.33 ± 0.64^{c}	22.00 ± 1.00^{h}	75.56 ± 1.11^{c}	18.67 ± 0.33^{f}	79.26 ± 0.37^{c}	20.67 ± 0.33^{h}	77.03 ± 0.37^{c}
N. sativa	27.00 ± 0.00^{g}	70.00 ± 0.00^{d}	25.00 ± 0.00^{g}	72.22 ± 0.00^{d}	23.67 ± 0.33^{e}	73.70 ± 0.37^{d}	25.00 ± 0.00^{g}	72.22 ± 0.00^{d}
E. kebericho	37.33 ± 0.33^{d}	58.52 ± 0.37^{g}	34.33 ± 0.33^{d}	61.86 ± 0.37^{g}	29.33 ± 0.33^{c}	67.41 ± 0.37^{f}	31.33 ± 0.33^{d}	65.19 ± 0.37^{g}
C. aurea	34.67 ± 0.33^{e}	61.48 ± 0.37^{f}	30.67 ± 0.33^{f}	65.92 ± 0.37^{e}	26.00 ± 0.00^{d}	71.11 ± 0.00^{e}	28.67 ± 0.33^{f}	68.14 ± 0.37^{e}
J. schimpeniana	30.33 ± 0.33^{f}	66.30 ± 0.37^{e}	32.67 ± 0.33^{e}	63.70 ± 0.37^{f}	28.67 ± 0.33^{c}	68.14 ± 0.37^{f}	30.00 ± 0.00^{e}	66.67 ± 0.00^{f}
O. abyssinica	40.67 ± 0.33^{c}	54.81 ± 0.37^{h}	37.00 ± 0.00^{c}	58.89 ± 0.00^{h}	34.67 ± 0.33^{b}	61.48 ± 0.37^{g}	36.67 ± 0.33^{c}	59.26 ± 0.37^{h}
O. lamiifolium	43.00 ± 0.00^{b}	52.22 ± 0.00^{i}	40.67 ± 0.33^{b}	54.81 ± 0.37^{i}	36.67 ± 2.03^{b}	59.26 ± 2.25^{g}	41.00 ± 0.58^{b}	54.44 ± 0.64^{i}
Control	90.00 ± 0.00^{a}	0.00 ± 0.00^{j}	90.00 ± 0.00^{a}	0.00 ± 0.00^{j}	90.00 ± 0.00^{a}	0.00 ± 0.00^{h}	90.00 ± 0.00^{a}	0.00 ± 0.00^{j}
LSD (0.05)	0.93	1.04	1.20	1.34	2.04	2.26	1.08	1.19
CV	1.52	1.02	2.06	1.27	3.85	2.03	1.91	1.12

Table 4. Antifungal effects of extracts from selected medicinal plant species on the radial growth of *Colletotrichum gloeosporioides*. Means within the same column followed by the same letter are not significantly different at $p < 0.05$ according to Fisher's least significant difference (LSD) test.

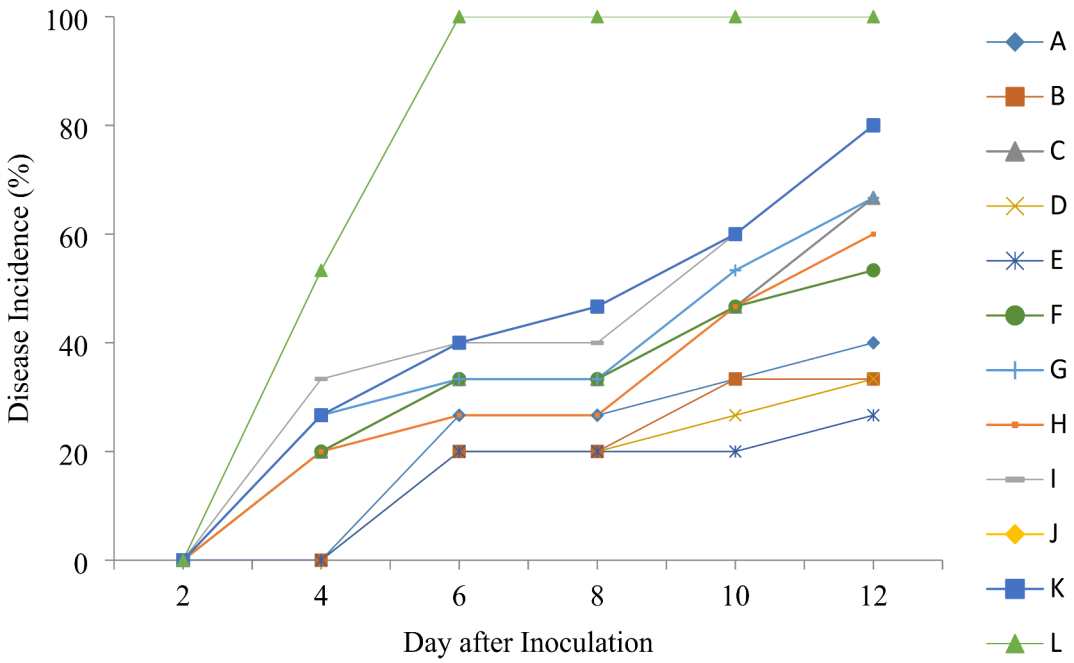

Fig. 3. Effect of combined application of plant extract and hot water treatment on anthracnose severity on artificially inoculated mango fruit. Where A = *Ruta chalepensis* + 50 °C, B = *R. chalepensis* + 55 °C, C = *R. chalepensis* + 24 °C, D = *Allium sativum L* + 50 °C, E = *A. sativum L* + 55 °C, F = *A. sativum L* + 24 °C, G = *Sarcocephalus latifolius* + 50 °C, H = *S. latifolius* + 55 °C, I = *S. latifolius* + 24 °C, J = Control + 50°C, K = Control + 55 °C, L = Control + 24 °C.

Total soluble solid (TSS °Brix) of mango fruit

The combined application of plant extracts and hot water temperature treatments had a significant effect ($p < 0.05$) on the total soluble solids (TSS) of mango fruit (Table 9).

The combination of *R. chalepensis* with 55 °C HWT and *A. sativum* at 55 °C HWT resulted in the lowest TSS of 9.33 °Brix, followed by *A. sativum* at 50 °C with a TSS of 9.67 °Brix. In contrast, the untreated control treatment had the highest TSS at 11.33 °Brix. Other treatments yielded TSS values ranging from 10.00 to 10.67 °Brix.

Plant extract	Hot water treatment			Mean
	55 °C	50 °C	24 °C	
R. chalepensis	33.33 ± 6.67	40.00 ± 0.00	66.67 ± 6.67	37.78 ± 10.19^{d}
A. sativum	26.67 ± 6.67	33.33 ± 6.67	53.33 ± 6.67	46.78 ± 8.01^{c}
S. latifolius	60.00 ± 0.00	66.67 ± 6.67	80.00 ± 0.00	68.89 ± 5.88^{b}
Control	80.00 ± 0.00	80.00 ± 0.00	100 ± 0.00	86.67 ± 6.67^{a}
Mean	50.00 ± 12.32^{a}	55.00 ± 11.01^{b}	75.00 ± 9.95^{c}	
CV (%)				13.60

Table 5. Effect of combined application of plant extract and hot water treatment on anthracnose incidence (%) on artificially inoculated mango fruit. Mean followed by the same letter are not statistically significant at $p < 0.05$ according to LSD; CV = Coefficient of variation.

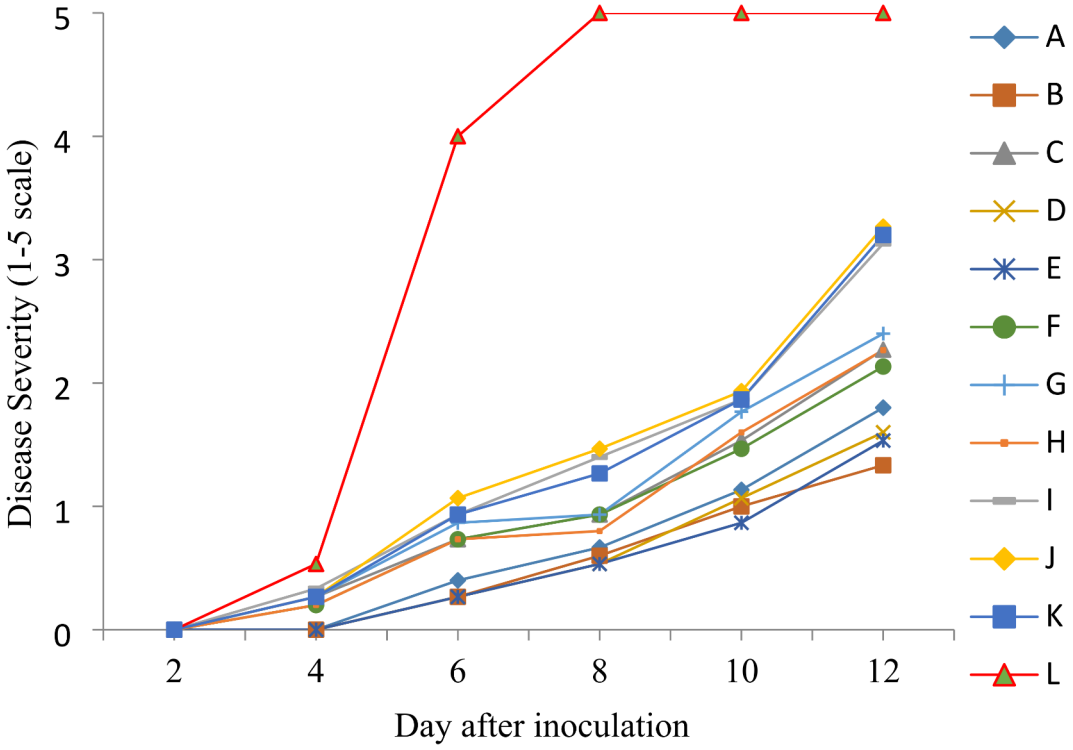

Fig. 4. Effect of combined application of plant extract and hot water treatment on anthracnose severity on artificially inoculated mango fruit. Where A = *Ruta chalepensis* + 50 °C, B = *R. chalepensis* + 55 °C, C = *R. chalepensis* + 24 °C, D = *Allium sativum L* + 50 °C; E = *A. sativum* + 55 °C, F = *A. sativum* + 24 °C, G = *Sarcocephalus latifolius* + 50 °C, H = *S. latifolius* + 55 °C =,I = *S. latifolius* + 24 °C, J = Control + 50°C, K = Control + 55 °C, L = Control + 24 °C.

Effect of the combination of plant extracts and hot water on pH of mango fruit

The combined application of plant extracts and hot water treatments did not significantly affect the pH of mango fruits. However, the use of plant extracts and hot water treatments individually showed significant ($P < 0.05$) differences (Table 10). Among the treatments, *Allium sativum* at 55 °C and 50 °C hot water had the lowest pH values, ranging from 4.19 to 4.21, while *Ruta chalepensis* combined with 55 °C and 50 °C hot water had pH values ranging from 4.20 to 4.32. In this experiment, the pH values varied between 4.19 and 4.77. Overall, the treatments showed significant differences compared to the control.

Effect of the combination of plant extracts and hot water on TA of mango fruit

The combined application of plant extracts and hot water treatments significantly affected the maintenance of postharvest titratable acidity (TA) in mango fruits. *A. sativum* L at 55 °C and *R. chalepensis* at 55 °C recorded the highest TA, with values of 0.84 and 0.83, respectively, compared to the untreated control, which had a TA of 0.35. The TA for other treatments ranged from 0.50 to 0.64. In the present study extrats from *Sarcocephalus latifolius* showed modest effect on TA of mango fruit when its applied in combination with hot water treatment at different temperature (Table 11).

Plant extract	Hot water treat	Disease severity (1–5 scale)*
R. chalepensis	55 °C	1.33 ± 0.24^{e}
	50 °C	$1.80 \pm 0.00^{c-e}$
	24 °C	2.27 ± 0.18^{c}
A. sativum	55 °C	1.33 ± 0.24^{e}
	50 °C	1.60 ± 0.31^{de}
	24 °C	2.13 ± 0.27^{cd}
S. latifolius	55 °C	2.27 ± 0.07^{c}
	50 °C	2.40 ± 0.20^{c}
	24 °C	3.13 ± 0.13^{b}
Control	55 °C	3.20 ± 0.12^{b}
	50 °C	3.27 ± 0.07^{b}
	24 °C	5.00 ± 0.00^{a}
CV (%)		12.95

Table 6. Effect of combined application of plant extract and hot water treatment on anthracnose severity on artificially inoculated mango fruit. *Disease Severity scored on 1–5 scale, Where 1 = infected area 0–1% no disease 2 = 1–5%, slight diseases; 3 = 6–9%, moderate disease; 4 = 10–49%, severe disease and 5 = 50–100%; very severe disease. Mean followed with the same latter(s) are not statistically different at $p < 0.05$ according to LSD.

Fig. 5. Effect of combined application of plant extract and hot water treatment on anthracnose intensity on artificially inoculated mango fruit. Where a = *Ruta chalepensis* + HWT at 55 °C, b = *R. chalepensis* + HWT at 50 °C, c = *R. chalepensis* + HWT at 24 °C, d = *Allium sativum* + HWT at 55 °C, e = *A. sativum* + HWT at 50 °C, f = *A. sativum* + HWT at 24 °C, g = *Sarcocephalus latifolius* + HWT at 55 °C, h = *S. latifolius* + HWT at 50 °C, i = *S. latifolius* + HWT at 24 °C, j = No extract + HWT at 55 °C, k = No extract + HWT at 50 °C, l = No extract + HWT at 24 °C (Untreated control).

Plant extracts	Hot water treatment			Mean
	55 °C	50 °C	24 °C	
R. chalepensis	83.33 ± 3.33^{a}	80.00 ± 0.00^{a}	73.33 ± 6.67^{a}	78.89^{A}
A. sativum	86.67 ± 3.33^{a}	83.33 ± 3.33^{a}	73.33 ± 6.67^{a}	81.11^{A}
S. latifolius	73.33 ± 6.67^{a}	73.33 ± 6.67^{a}	53.33 ± 6.67^{b}	66.67^{B}
Control	53.33 ± 6.67^{b}	46.67 ± 6.67^{b}	0.00 ± 0.00^{c}	33.33^{C}
Mean	74.17^{Z}	70.83^{Z}	50.00^{Y}	65.00
CV (%)				14.27

Table 7. Effect of combined application of plant extract and hot water treatment marketability on artificially inoculated mango fruit. Mean followed with the same latter(s) are not statistically different at $p < 0.05$ according to LSD.

Plant extract	Hot water treatment			Mean
	55 °C	50 °C	24 °C	
R. chalepensis	4.33 ± 0.33^{fg}	5.50 ± 0.50^{f}	7.50 ± 0.50^{de}	5.78^{c}
A. sativum	3.67 ± 0.33^{g}	4.33 ± 0.33^{fg}	7.33 ± 0.33^{e}	5.11^{c}
S. latifolius	9.00 ± 0.58^{c}	8.67 ± 0.33^{cd}	10.33 ± 0.33^{b}	9.33^{b}
Control	10.33 ± 0.33^{b}	10.67 ± 0.33^{b}	18.33 ± 0.67^{a}	13.11^{a}
Mean	6.83^{y}	7.29^{y}	10.87^{z}	8.33
CV (%)				8.83

Table 8. Effect of combined application of plant extract and hot water treatment on WL of artificially inoculated mango fruit. Values (mean of three replications) followed with the same latter(s) are not statistically different at $p < 0.05$.

Plant extracts	Hot water treatment			Mean
	55 °C	50 °C	24 °C	
R. chalepensis	9.33 ± 0.33	10.00 ± 0.00	10.00 ± 0.00	9.78^{b}
A. sativum	9.33 ± 0.33	9.67 ± 0.33	10.00 ± 0.00	9.67^{b}
S. latifolius	10.67 ± 0.33	10.33 ± 0.33	10.67 ± 0.33	10.56^{a}
Control	10.33 ± 0.33	10.33 ± 0.33	11.33 ± 0.33	10.67^{a}
Mean	9.92^{y}	10.08^{yz}	10.50^{z}	10.17
CV (%)				4.91

Table 9. Effect of combined application of plant extract and hot water treatment on TSS on artificially inoculated mango fruit. Values (mean of three replications) followed with the same latter(s) are not statistically different at $p < 0.05$.

Plant extract	Hot water treatment			Mean
	55 °C	50 °C	24 °C	
R. chalepensis	4.20 ± 0.04	4.32 ± 0.06	4.57 ± 0.05	4.37^{B}
A. sativum	4.19 ± 0.02	4.21 ± 0.04	4.55 ± 0.06	4.32^{B}
S. latifolius	4.45 ± 0.03	4.50 ± 0.02	4.66 ± 0.03	4.54^{A}
Control	4.45 ± 0.03	4.52 ± 0.02	4.77 ± 0.06	4.58^{A}
Mean	4.32^{X}	4.39^{Y}	4.64^{Z}	4.45
CV(%)				1.65

Table 10. Effect of combined application of plant extract and hot water treatment pH on artificially inoculated mango fruit pH. Values (mean of three replications) followed with the same latter(s) are not statistically different at $p < 0.05$.

Plant extract	Hot water treatment			Mean
	55 °C	50 °C	24 °C	
R. chalepensis	0.83 ± 0.01	0.81 ± 0.06	0.62 ± 0.02	0.75^{A}
A. sativum L	0.84 ± 0.04	0.82 ± 0.01	0.64 ± 0.01	0.77^{A}
S. latifolius	0.59 ± 0.03	0.54 ± 0.02	0.52 ± 0.02	0.55^{B}
Control	0.50 ± 0.02	0.50 ± 0.02	0.35 ± 0.03	0.45^{C}
Mean	0.69^{X}	0.67^{X}	0.53^{Z}	0.6^{2}
CV (%)				7.69

Table 11. Effect of combined application of plant extract and hot water treatment on TA of artificially inoculated mango fruit. Values (mean of three replications) followed with the same latter(s) are not statistically different at $p < 0.05$.

Discussion

Postharvest diseases are the primary contributors to food loss in the fresh produce supply chain[49]. Mango anthracnose, predominantly caused by *C. gloeosporioides*, is recognized as the most significant postharvest disease impacting mango production and consumption worldwide. Although various species within the *Colletotrichum* complex have been reported to be associated with anthracnose in mango, *C. gloeosporioides*, which is characterized by black, expanding lesions on mango plant parts including fruits, leaves, flowers, petioles, twigs, and stems remains the predominant cause of this disease[8,38]. This study was initiated to develop safe, effective and sustainable strategies to manage postharvest mango anthracnose disease based on the biology of the pathogen.

The study characterized nine *C. gloeosporioides* isolates collected from diseased mangoes across three districts: Assosa, Bambasi, and Homosha, using cultural, morphological, and pathological traits. The cultural characteristics of the isolates were consistent with the known descriptions of *C. gloeosporioides*[42,49], displaying a variety of colony colors, including white, gray, pink, and others on different plate surfaces. The observed color diversity among the colonies may be due to phenotypic variations influenced by environmental conditions[39].

The colony growth rate of *C. gloeosporioides* isolates varied across the different culture media. All four media supported mycelial growth for each isolate. The average daily colony growth rates were 7.68 mm on Potato Dextrose Agar (PDA), 7.74 mm on Malt Extract Agar (MEA), and 7.94 mm on Mango Leaf Extract Agar (MLEA). Conidial production also varied among isolates and across different media. Variation in colony growth, topology, and sporulation capacity among *C. gloeosporioides* across different culture media has been documented in previous studies[42,51]. This variation may be attributed to differences in the nutritional content of the media. In the current study, Mango Leaf Extract Agar proved to be the most effective for colony growth, likely due to the nutrients in mango leaf extract that promote fungal development[52]. Conversely, Malt Extract Agar (MEA) provided optimal conditions for sporulation of the isolates. Devi et al.[53] similarly reported that MEA was highly effective for inducing heavy sporulation in various fungal species. In the present study, pathogenicity tests confirmed that the *C. gloeosporioides* isolates were pathogenic to harvested mango fruit. Abera et al.[42] identified pathogenicity toward mature fruit as a distinctive characteristic of *C. gloeosporioides*. The infected areas showed black, sunken lesions across the fruit's surface. Similarly, Dofuor et al.[8] reported that black, water-soaked lesions are typical features of mango anthracnose.

The present study also evaluates the antifungal activity of plant extracts against *C. gloeosporioides*. The use of natural products for controlling fungal diseases in plants is considered an interesting alternative to synthetic fungicides due to their specificity, biodegradability, low toxicity, minimal residual effects in the ecosystem, and lower negative impacts on the environment[54]. In vitro evaluation of extracts from nine different plant species demonstrated a significant reduction in the mycelial growth of *C. gloeosporioides*. The antifungal activity observed can likely be attributed to either individual compounds or synergistic effects of multiple compounds[55]. Methanol extracts of *R. chalepensis* and *A. sativum* exhibited prominent inhibition of *C. gloeosporioides* growth, with reductions of 83.7% and 78.14%, respectively, compared to the untreated control. Additionally, ethyl acetate extracts from these plants also significantly inhibited mycelial growth, with reductions of 88.89% and 85.19%, respectively. These findings align with studies by Alemu et al.,[35] and Imtiaj et al.[56], which reported that extracts of *R. chalepensis* and *A. sativum* possess antibiotic and antifungal properties, effectively suppressing the growth and conidial germination of *C. gloeosporioides*. Extracts from *S. latifolius* and *N. sativa* also demonstrated notable antifungal activity against *C. gloeosporioides*, inhibiting growth by over 80%, with the ethyl acetate extract from *S. latifolius* reducing growth by 79.26%. previously the effectiveness of *S. latifolius* and *N. sativa* against *C. gloeosporioides* has been well documented[57,58].

In the present study, extracts from *A. sativum*, *R. chalepensis*, and *S. latifolius* which showed potential in in vitro tests, were further evaluated in vivo in combination with hot water treatment for managing anthracnose disease on artificially inoculated mango fruit. The results indicated that the combined application of plant extracts and hot water treatment significantly ($p < 0.05$) reduced the incidence of postharvest mango anthracnose. The most substantial reduction was observed in fruits immersed in *A. sativum* extract at 55 °C, with a mean incidence of 26.67%. This treatment significantly influenced the development of anthracnose. Moreover, the greatest reduction in anthracnose severity, to a level of 1.33, was achieved with treatments involving *R. chalepensis* and *A. sativum*, both applied at a hot water temperature of 55 °C. The reduction in anthracnose development in

artificially infected mangoes suggests that biologically active plant-derived products could play a significant role in crop protection strategies. Plant extracts are considered non-phytotoxic and may serve as effective natural pesticides for crop protection[59]. The antifungal activity of these plant extracts is likely due to their secondary metabolites. Cragg et al.[60] also noted that plants contain thousands of compounds that could be valuable sources of new, biologically active molecules. The presence of bioactive compounds such as caffeic, p-coumaric, ferulic, and di-ferulic acids, along with various flavonoids and phenolic compounds in *A. sativum*[61,62], as well as linalyl acetate, β-linalool, and other bioactive compounds in *R. chalepensis*[63], contributes to their antifungal activities. Additionally, hot water treatment may alter the chemical environment of the fruit peel, activating antimicrobial compounds present on it[55] in addition to inhibiting the germination and growth of fungal pathogens[64]. The notable reduction in disease development could be attributed to the synergistic effects of the plant extract and hot water treatments[45].

This experiment indicates that integrated application of plant extracts and hot water treatment had significant ($P<0.05$) effect on marketability of mango fruit compared to control treatment. Treatment of the fruit with combined application *A. sativum* at and *R. chalepensis* with hot water treatment improve the marketability by more than 80%. Desiccation and decay are key factors that shorten the commercial lifespan of fruits, typically due to postharvest diseases and physiological disorders. This study found that fruits treated with different plant extracts showed enhanced marketability. This improvement is likely because the plant extracts inhibit microbial growth that causes rotting and lower the fruit's metabolic rate, reducing weight loss from respiration[65,66]. Moreover, plant extracts have been shown to act as anti-senescent agents, slowing down metabolic breakdown and deterioration caused by biochemical processes in the fruit[66]. The observed effectiveness of plant leaf extracts in reducing physiological loss and prolonging shelf life aligns with previous findings on various fruits[66].

The combined use of plant extracts and hot water treatment significantly ($p<0.05$) reduced fruit weight loss. In contrast, the control sample experienced a faster rate of weight loss. Bautista-Baños et al.[66] noted that high infection rates can lead to increased respiration and subsequent weight loss in fruits. Gidado et al.[67] indicate that mango with high respiration rates during storage tends to have shorter shelf life. Additionally, Vilaplana et al.[68] reported that applying postharvest treatments can mitigate weight loss and reduce disease in mangoes. The combined application of plant extracts and hot water temperature treatments showed a significant effect on TSS for the study. The extracts showed significant effect in maintain fruit TSS. The high TSS content of mango fruits were recorded could be due to the high level of anthracnose severity, which could have accelerated ripening, thus, resulting in increment of sugar level of the fruits before the other treatments. The low TSS may be the attribute of the low ripening process as a result of low level of anthracnose infection on the fruits. The application of plant extracts and hot water temperature alone showed significant ($p<0.05$) differences on PH of mango fruits. It was suggested that the change in pH is associated with the effect of treatment on the respiration and metabolic activity of the fruits[69]. In this study, it seems that fruits with high rate of disease incidence and severity had higher rates of respiration which would raise pH of the fruit juice as ripening advances[70]. The combined application of plant extracts and hot water temperature showed non-significant difference on titerable acidity (TA) of mango fruits. The reduced TA may be the attribute of anthracnose disease, which enhance ripening and senescence of fruit; and thus reduced TA possibly through the use substrate for respiration[69].

Conclusion

The present study demonstrates that combining plant extracts with HWT significantly ($p<0.05$) reduces anthracnose disease development in artificially inoculated mangoes. Specifically, using *R. chalepensis* and *A. sativum* combined with hot water treatments at 55 °C and 50 °C notably lowered anthracnose disease intensity and enhanced fruit marketability by over 80%. This combined approach also showed promise in reducing weight loss and preserving postharvest fruit quality, including TSS, TA, and pH. These findings suggest that integrated application of plant extract and hot water treatment could be an effective, environmentally friendly, and sustainable strategy for managing postharvest mango anthracnose. Further research is needed to identify the biologically active compounds in the plant extracts and to elucidate the mechanisms by which these treatments work.

Data availability

The corresponding author is able to supply the datasets utilized and/or analyzed for this study upon reasonable request.

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Author contributions

S.W., conceived and designed the study, collected the data, interpreted the data and wrote the manuscript. K.A. designed the study, analyzed and interpreted the data, wrote and edited the manuscript; B.T. wrote and edited the manuscript. All authors contributed as members of the research team, read, and approved the M.S.

Declarations

Ethics approval and consent to participate

The plant materials used for this experiment were collected after getting consent from the local authority.

Competing interests

The authors declare no competing interests.

Additional information

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