



Effects of Neem, Moringa, and Synthesized Silver Nanoparticles Coating on Postharvest Shelf Life and Quality Retention of Tomato (*Solanum lycopersicum* L.)

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Authors' contributions

This work was carried out in collaboration among all authors. Authors TSE and OKA designed the study. Authors TSE, OKA, TSO and OAO wrote the protocol. Authors WML, EOF and AOA managed the analyses of the study and literature searches. Authors EOF and OKA wrote the first draft of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

This study aimed to investigate the effects of synthesized silver nanoparticles (AgNPs), *Azadirachta indica* (neem), and *Moringa oleifera* (moringa) leaf extracts on the shelf life and quality retention of tomato (*Solanum lycopersicum* L.) fruits during storage. Thirty-five (35) red, matured tomato fruits were collected, rinsed and grouped for each treatment with AgNPs, neem and moringa coating: Control (n=5), moringa aqueous leaf extract (MALE) (n=5), neem aqueous leaf extract (NALE) (n=5), 1:9 and 6:4 moringa aqueous leaf extract synthesized silver nanoparticles (MALE-AgNPs) (n=5), 1:9 and 6:4 neem aqueous leaf extract synthesized silver nanoparticles (NALE-AgNPs) (n=5), respectively. The firmness, shelf life, and postharvest decay percentage of the tomato fruits were determined. Additionally, fungi associated with the postharvest deterioration of the fruits were isolated and identified using standard procedures. From the results of this study, tomato fruits coated with either neem or moringa crude extract showed the longest shelf life, as compared to the coating with AgNPs. Additionally, two fungi, namely *Aspergillus niger* and *Aspergillus flavus*, were isolated from the decayed tomato fruits. In conclusion, the neem and moringa leaf extracts are effective in the extension of the shelf life and retention of the quality of tomato fruits.

Keywords: *Azadirachta indica*; *Moringa oleifera*; Tomato fruits; Postharvest; shelf life; Preservatives.

1. INTRODUCTION

Crop postharvest shelf life extension and quality retention remain a key global issue. Since food is a crucial part of human life, it is important to prevent postharvest losses and increase the shelf life of food. Tomato (*Solanum lycopersicum* L.) is widely grown and consumed in Nigeria, serving as a valuable source of vitamins and minerals [1]. Tomato is a commonly consumed fruit that is vital for health, and therefore available fresh or in paste form [2]. However, the perishability of tomatoes poses challenges for farmers and consumers, affecting the quality and safety of the fruits. Addressing the shelf life is crucial to meeting consumer demands and ensuring a stable tomato supply [3].

Plant nutraceuticals are sourced from different plant parts as antioxidant and antimicrobial agents in food preservations due to their excellent source of natural bioactive compounds like polyphenols and terpenoids [4]. The extracts of these plants are increasingly considered natural preservatives, potentially replacing synthetic counterparts such as sodium hypochlorite, sodium metabisulphite, and calcium chloride in various applications [5]. Moringa (*Moringa oleifera*) is recognized for its abundance of bioactive compounds, particularly in its leaves, which are rich in vitamins, carotenoids, polyphenols, phenolic acids, flavonoids, alkaloids, glucosinolates, isothiocyanates, tannins, and saponins [6,7]. The embedded bioactive compounds of its leaves contribute to its attributed pharmacological

properties [8-13]. Neem (*Azadirachta indica*) on the other hand, is a widely available plant that contains phytochemicals [14], which can inhibit spoilage-causing micro-organisms in tomatoes, and also preserve its level of nutrients [15]. Various phytoconstituents of its leaves contribute to various attributed biological activities including, antioxidant, antidiabetic, antimicrobial, antifungal, anti-inflammatory, anti-tumor, anti-cancer, and anti-fertility [16].

Most recently, green-synthesized coatings such as plant-derived silver nanoparticles which contain considerable bioactive compounds have been investigated as a potential option to minimize fruit respiration, spoilage, and microbial growth, and thus promote postharvest shelf life and quality retention of fruits [17,18]. Here in this study, the various biological activities associated with the abundant bioactive compounds of neem and moringa have informed and necessitated the investigation into their use for silver nanoparticle synthesis and their effects on shelf life and quality retention of tomato fruits during storage.

2. MATERIALS AND METHODS

2.1 Coating Materials Preparation and Fruit Samples Collection

Fresh leaves of neem and moringa were collected at the back of the Lagos State University sports center and a residential area at Adexson, Lagos State, respectively, and were identified and authenticated at the Herbarium of the Department of Botany, Lagos State

University, Ojo, Lagos State, Nigeria. The dried leaves were blended to get the powder. The powder was then sieved and kept in separate air-tight conical flasks.

Thirty-five (35) red matured, firm, smooth, and healthy tomato fruits were obtained from a local food and fruits market, Iyana Iba market, Ojo, Lagos State, Nigeria. The tomatoes were divided into five for each treatment and control group. The tomatoes were procured based on their firmness and reddish matured color before being stored at room temperature. The tomato fruits were washed under running tap water and air-dried at room temperature. Neem/moringa aqueous leaf extracts were prepared by dissolving 70g of neem/ moringa leaf powder in 350 mL of distilled water separately.

2.2 Silver Nanoparticles (AgNPs) Synthesis

The leaf powder of neem and moringa (100 g each) was separately dissolved in 1000 mL distilled water, filtered, and stored. The silver nanoparticles were prepared according to the methods of [19,20] with some modifications. A freshly prepared 2 mM silver nitrate solution was mixed with neem and moringa aqueous leaf extract separately in ratios 1:9 and 6:4, respectively. The color change indicated silver nanoparticle synthesis and was further confirmed by observing the absorption peak between 400 – 450 nm using a UV-visible spectrophotometer.

2.3 Treatment of Tomato with NALE, MALE, NALE-AgNPs and MALE-AgNPs

The tomato samples were immersed in neem and moringa aqueous leaf extract separately before being arranged in a clean container and kept at room temperature on the Laboratory table. Changes were observed and data were recorded for the 15 days of treatment to ascertain the effects of the extracts. Conversely, another set of tomatoes was immersed in each AgNP solution for 2 hours before being placed in a clean container in the laboratory at 25°C. Changes were also observed and data were recorded for the 15 days of treatment to ascertain the effects of the synthesized AgNPs.

2.4 Data Collection

The post-harvest decay percentage (PDP), marketability, shelf-life, and firmness of the tomato fruits were calculated [21] using:

$$\text{Post-harvest decay percentage (PDP)} = \frac{\text{number of decaying fruits}}{\text{total number of fruits}} \times 100$$

$$\text{Marketability} = \frac{\text{number of Marketable fruits}}{\text{total number of fruits}} \times 100$$

Firmness = rating scale 1 - 5

Where 1 is very poor, 2 is poor, 3 is acceptable, 4 is good, and 5 is excellent.

2.5 Isolation and Identification of Fungi Causing Spoilage of Tomato Fruits During Storage

Potato dextrose agar was used for the isolation of fungi from the tomato fruits and the preparation of pure culture. Thirty-nine grams (39g) of potato dextrose agar was dissolved in 1000 mL of distilled water in a sterile conical flask covered with cotton wool and aluminum foil paper. The mixture was shaken thoroughly and autoclaved at 121°C for 15 minutes under a pressure of 15 pounds per square inch (15lb/inch²). The medium was cooled after autoclaving to 45°C and then dispensed aseptically into a sterile Petri dish. Chloramphenicol was added to the medium to prevent the growth medium. The workbench was disinfected, and a sterilized cork borer was used to extract pieces from a diseased tomato, which were placed into the medium. After 5 days of incubation at 37°C, mixed cultures were re-isolated until obtaining a pure culture. Identification was based on morphological features and microscopic examination using lactophenol cotton blue solution, following the modified procedures of [22].

2.6 Statistical Analysis

The daily weight of the coated tomatoes was recorded in triplicates and the data were subjected to univariate statistical analysis such as mean and standard deviation (SD) using Statistix 10 software. The means were separated using analysis of variance and comparisons were made Least Significance Difference (LSD) at 95% confidence level.

3. RESULTS

The 15-days experiment showed that the control group's tomatoes spoiled by the 8th day, losing their firmness from the 5th day. However, tomatoes coated with neem aqueous leaf extract synthesized silver nanoparticles (NALE-AgNPs) solution (6:4) lasted 12 days, with significant weight loss from day 6. Another variant (1:9) lasted 14 days before significant weight loss led to complete deterioration by day 15. Tomatoes coated with neem aqueous leaf extract had the longest shelf life, losing firmness at day 8 and deteriorating completely by day 15.

The 15-days experiment revealed that all the tomato fruits deteriorated by the 15th day, with observable decay starting from the 5th day. White mold appeared from the 3rd day before decay. Tomato fruits coated with moringa aqueous leaf extracts deteriorated from the 8th day, but some lasted until the 15th day. However, those coated with moringa aqueous leaf extract synthesized silver nanoparticles (MALE-AgNPs) showed preservation until the 8th day (in a 6:4) and some lasting until the 10th day (in a 1:9). Significant differences in weight loss were observed on specific days between moringa-coated and control fruits, as well as between silver nanoparticle-coated and control fruits.

Table 1. Effects of NALE and NALE-AgNPs coatings on the weight of *S. lycopersicum*

Groups/Days	Control	NALE	Nano 1	Nano 2
IW1	91.50 ± 4.64 ^a	74.57 ± 12.26 ^b	80.87 ± 4.92 ^c	83.23 ± 8.14 ^c
IW2	91.50 ± 4.64 ^a	84.3 ⁰ ± 12.50 ^b	81.97 ± 4.88 ^b	84.80 ± 8.41 ^b
DAT 1	89.60 ± 4.32 ^a	79.03 ± 12.08 ^b	80.83 ± 3.85 ^b	83.00 ± 8.29 ^b
DAT 2	88.07 ± 4.16 ^a	76.70 ± 12.08 ^b	78.83 ± 3.23 ^b	81.47 ± 8.11 ^b
DAT 3	82.33 ± 3.61 ^a	75.33 ± 13.25 ^b	76.13 ± 4.37 ^b	80.23 ± 7.88 ^a
DAT 4	79.30 ± 6.67 ^a	75.30 ± 11.98 ^b	72.57 ± 5.50 ^b	78.37 ± 7.04 ^a
DAT 5	75.80 ± 6.05 ^a	73.37 ± 13.20 ^a	73.30 ± 4.03 ^a	78.83 ± 8.34 ^b
DAT 6	70.30 ± 4.21 ^a	72.60 ± 13.10 ^a	71.10 ± 4.33 ^a	76.50 ± 7.02 ^b
DAT 7	66.57 ± 4.10 ^a	71.87 ± 12.64 ^b	68.57 ± 5.31 ^a	74.27 ± 6.12 ^b
DAT 8	61.93 ± 4.73 ^a	71.40 ± 12.47 ^b	67.27 ± 5.14 ^c	69.20 ± 4.50 ^c
DAT 9	0.00 ± 0.00 ^a	70.93 ± 12.14 ^b	65.97 ± 4.97 ^c	67.40 ± 2.94 ^c
DAT 10	0.00 ± 0.00 ^a	67.60 ± 11.00 ^b	64.73 ± 4.73 ^c	65.00 ± 1.39 ^c
DAT 11	0.00 ± 0.00 ^a	64.33 ± 11.75 ^b	63.83 ± 4.39 ^b	57.33 ± 0.75 ^c
DAT 12	0.00 ± 0.00 ^a	62.87 ± 12.24 ^b	62.87 ± 4.27 ^b	49.40 ± 0.35 ^c

Mean ± SEM values with the same alphabet in the same row are not significantly different from each other at $p < 0.05$, where, NALE= Neem aqueous leaf extract, Nano 1 = Neem aqueous leaf extract dissolved in AgNO₃ at 1:9, and Nano 2 = Neem aqueous leaf extract dissolved in AgNO₃ at 6:4, IW1= Initial weight before coating, IW2= Initial weight after coating. DAT= Days after treatment

Table 2. Effects of MALE and MALE-AgNPs coatings on the weight of *S. lycopersicum*

Groups/Days	Control	MALE	Nano 3	Nano 4
IW1	91.50 ± 4.64 ^a	81.73 ± 3.90 ^b	93.77 ± 2.64 ^a	94.07 ± 6.31 ^a
IW2	91.50 ± 4.64 ^a	95.50 ± 5.24 ^b	99.30 ± 5.27 ^c	100.73 ± 7.94 ^c
DAT 1	89.53 ± 4.21 ^a	86.93 ± 10.47 ^a	97.50 ± 5.15 ^b	98.67 ± 7.72 ^b
DAT 2	88.07 ± 4.16 ^a	82.90 ± 10.12 ^b	95.03 ± 4.73 ^c	95.80 ± 7.01 ^c
DAT 3	82.33 ± 3.61 ^a	83.83 ± 11.07 ^a	89.13 ± 7.90 ^b	92.77 ± 6.92 ^b
DAT 4	80.80 ± 4.08 ^a	80.73 ± 9.64 ^a	87.77 ± 7.42 ^b	91.50 ± 6.29 ^b
DAT 5	75.80 ± 6.05 ^a	77.53 ± 7.75 ^a	86.57 ± 6.75 ^b	90.17 ± 5.71 ^b
DAT 6	72.77 ± 4.25 ^a	74.13 ± 5.81 ^a	87.87 ± 6.12 ^b	89.43 ± 6.84 ^b
DAT 7	68.93 ± 4.19 ^a	70.67 ± 7.11 ^a	85.30 ± 4.85 ^b	88.00 ± 6.76 ^b
DAT 8	64.67 ± 4.73 ^a	73.87 ± 6.12 ^b	82.60 ± 3.12 ^c	85.47 ± 5.77 ^c
DAT 9	0.00 ± 0.00 ^a	65.33 ± 11.61 ^b	69.97 ± 4.79 ^c	77.97 ± 7.08 ^d
DAT 10	0.00 ± 0.00 ^a	64.83 ± 11.56 ^b	62.13 ± 7.04 ^b	74.47 ± 8.31 ^c
DAT 11	0.00 ± 0.00 ^a	64.33 ± 11.51 ^b	49.73 ± 13.28 ^c	68.07 ± 16.17 ^b
DAT 12	0.00 ± 0.00 ^a	63.20 ± 11.01 ^b	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a

Mean \pm SEM values with the same alphabet in the same row are not significantly different from each other at $p < 0.05$, where, MALE= Moringa aqueous leaf extract, Nano 3 = Moringa aqueous leaf extract dissolved in AgNO₃ at 1:9, and Nano 4 = Moringa aqueous leaf extract dissolved in AgNO₃ at 6:4, IW1= Initial weight before coating, IW2= Initial weight after coating. DAT= Days after treatment

Table 3. Post-harvest decay percentage of tomato fruits coated with NALE, MALE, and their synthesized silver nanoparticles at different concentrations

	Day 3	Day 5	Day 7	Day 9	Day 11	Day 13	Day 15
Control	0	20	60	100	100	100	100
NALE	0	0	20	40	40	60	80
MALE	0	0	40	40	40	60	80
Nano 1	0	0	80	80	80	80	100
Nano 2	0	20	60	60	60	100	100
Nano 3	0	0	40	40	*	*	*
Nano 4	0	0	60	60	*	*	*

* Denotes null set (totally decayed sample)

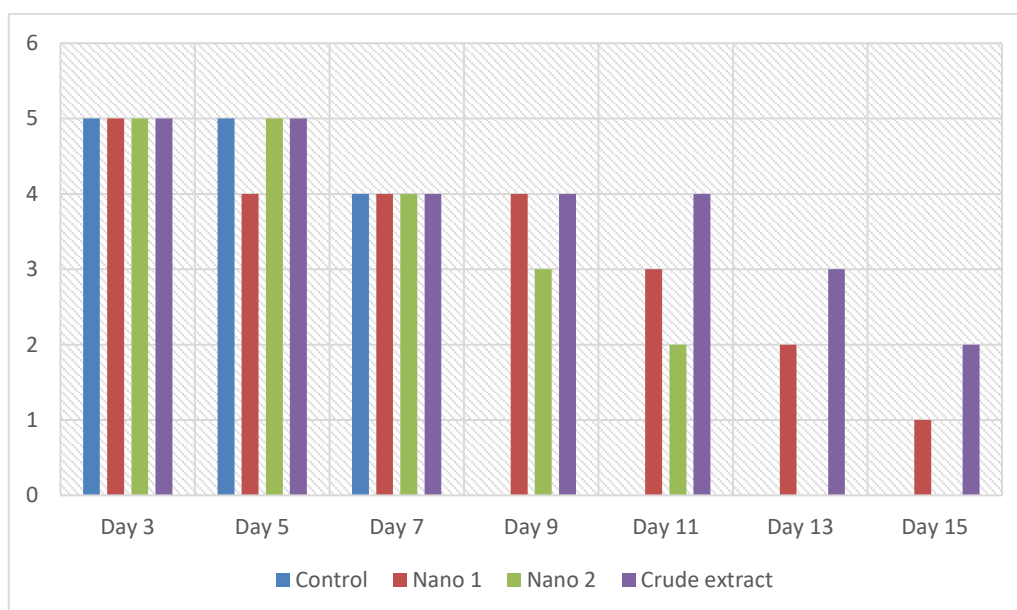


Fig. 1. Firmness of Tomato coated with NALE crude and NALE-AgNPs during storage

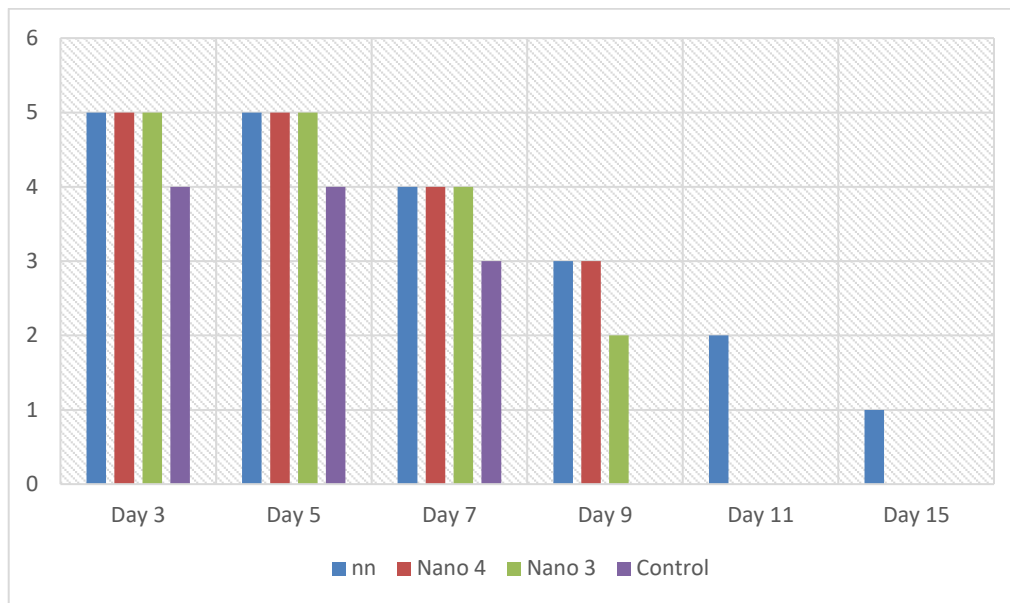


Fig. 2. Firmness of Tomato coated with MALE crude (nn) and MALE-AgNPs during storage

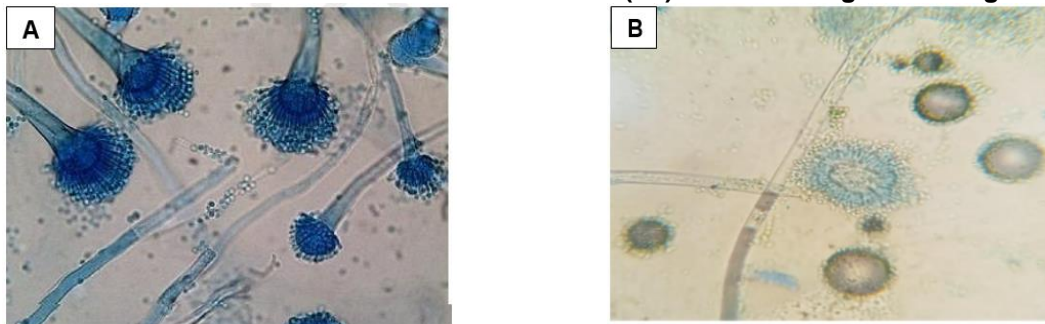


Fig. 3. Photomicrograph of (a) *Aspergillus niger* (X400) (b) *Aspergillus flavus* (X400) isolated from deteriorated tomato fruits

Table 3 illustrates the postharvest decay of tomato fruits during the storage period of 15 days. The percentage of decay was observed to increase as the days increased. The deterioration of tomato fruits started on the 5th day with only 20 percent of both control and NALE-AgNPs (6:4) observed. Meanwhile, it was observed that, the different concentrations of MALE-AgNPs (1:9 and 6:4) delayed decay up to the 15th day.

Figs. 1 and 2 illustrate the firmness of the tomato fruits. It was observed that the tomatoes coated with the aqueous leaf extracts of neem and moringa have the longest shelf life of 15 days compared with 8 days of moringa and neem silver nanoparticles and the control.

A total of two fungi, *Aspergillus* species, were isolated, identified, and characterized from the

deteriorated tomato fruits. These were *A. niger* and *A. flavus*.

The conidia of *A. niger* were dark brown to black and spherical having a sporulated surface growth on the culture media, with visible aseptate hyphae (without cross-wall) while, the conidia of *A. flavus* were smooth with green dispersed spores, and septate (cross-walled) hyphae were present with phialides.

4. DISCUSSION

The prevention of fruit spoilage by pathogenic fungi and the preservation of fruit freshness poses a serious challenge in the fruit industry. Concerns over the use of synthetic preservatives have led to a shift towards exploring plant-based alternatives. The effectiveness of neem and moringa aqueous leaf extracts in reducing tomato decay observed in this study suggests

that it could be a viable alternative for combating pathogen-related decay in tomatoes. This observation aligns with a study reported by [23], who reported that treating various kinds of fruits with chitosan and guava leaf extract significantly increased the shelf life of the fruits. Similarly, our findings are consistent with the report of [24], who highlighted the effectiveness of extracts from medicinal plants like *Allium sativum*, *Azadirachta indica*, *Mentha arvensis*, and *Psoralea corlylifolia* in preserving fruits from pathogenic and environmental factors. Moreover, in this study, neem aqueous leaf extract increased the shelf life of the fruits, possibly by reducing the fungal and bacterial spoilage during storage.

Tomatoes coated with moringa and neem aqueous leaf extracts exhibited reduced post-harvest decay, as reflected in the reduced number of decayed fruits compared to the control, NALE, and MALE-AgNPs. These plants also showed higher marketability with a greater number of marketable fruits in both categories. While control fruits lasted for only 8 days, most of the coated tomato fruits still maintained and retained their colour and number but became completely rotten on the 15th day. The tomato fruits coated with MALE-AgNPs at different concentrations had lower postharvest decay percentages. This implied that coating with MALE-AgNPs could help tomato fruits resist environmental and pathogenic attacks better than other treatments. The observed progressive weight loss in neem-coated tomatoes and control aligns with the report of [25], who stated that post-harvest weight change in fruits is typically linked to temperature and storage time, often attributed to water loss through transpiration. Thus, the higher the temperature, the higher the respiratory rate of the fruits and the higher its metabolic activity, which may lead to an increase in weight loss during storage. The higher decrease in the firmness of the control tomato fruits compared to the treated fruits may be attributed to a higher rate of metabolic activities and activity of cell wall degrading enzymes that loosens the fruit skin which result in higher permeability of the cell for higher rate of moisture loss.

Another possible reason why the incorporation of silver nanoparticles showed lower preservative effects on tomatoes compared to the leaf extracts of neem and moringa could be attributed to the complex interactions between silver nanoparticles and the tomato surface, as well as

the distinct antimicrobial properties of the leaf extracts. AgNPs exert antimicrobial activity primarily through the release of silver ions that disrupt microbial cell membranes and inhibit cellular processes [26], the antimicrobial mechanisms of neem and moringa leaf extracts are multifaceted and may involve the inhibition of microbial enzymes and disruption of cell membranes, which interferes with microbial metabolism [27]. This diversity in antimicrobial mechanisms could contribute to the superior preservation effects of the leaf extracts on tomatoes compared to AgNPs.

In addition, the findings of this study also revealed some of the fungi associated with the post-harvest decay of the tomato fruits in storage. These fungi are *Aspergillus niger* and *A. flavus*, which have previously been reported as pathogens of tomato fruits [28,29]. They have also been found in other crops including orange, sour-sop, and garri (fried mashed fermented cassava) [3]. Association of these fungi with these fruits or foods may suggest their omnipresent, non-host-specific, and non-geographical-specific nature.

5. CONCLUSION

Our findings from this study demonstrated that neem (*Azadirachta indica*) and moringa (*Moringa oleifera*) leaf powder and their synthesized silver nanoparticles can effectively prolong the shelf life and also preserve the quality of tomato fruits beyond their typical limits. This offers valuable information on plant leaves' potential in postharvest preservation in addition to their known nutraceutical properties. Future studies may explore the phytochemical composition and *in-vitro* and *in-vivo* potentials of the leaf extracts of the plants in preventing disease development in tomato fruits, which may explicate their postharvest shelf life and quality retention potentials on tomato fruits.

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COMPETING INTERESTS

The authors have declared that no competing interests exist.

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