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# SHELF-LIFE EXTENSION OF TOMATOES (*Solanumlycopersicum*), OKRA (*Abelmoschus esculentus*) AND EGGPLANT (*Solanum melongena*) USING EDIBLE COATING

**Obiaocha-Nwaogwugwu, C. N., Eruteya, O. C.* and Ahaotu, I.**
Department of Microbiology, Faculty of Science, University of Port Harcourt, Nigeria.

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## ABSTRACT

*Post-harvest losses of fresh farm produce, especially fruits grown in Nigeria is high. This study was aimed at investigating the shelf-life extension of tomatoes, okra and eggplants stored at room temperature (29±2°C), using edible coating involving beeswax, lime and pectin. Five edible films were formulated, namely: Beeswax alone (W), pectin alone (P), beeswax and pectin (WP), beeswax and lime (WL), beeswax, lime and pectin (WLP). Uncoated fruits served as control. The proximate composition of the beeswax and the potential spoilage microorganisms of the selected fruits were determined using standard methods. The results of the proximate composition revealed the following values for carbohydrate (46.25%), lipid (38.50%), moisture (8.85%), protein (5.25%), fibre (1.02%) and ash (0.53%). The WP combination gave the best results, extending the shelf life of the tomatoes, okra and eggplants by more than 23, 21 and 11 days, respectively more than the uncoated fruits. The least effective edible coating was the WLP combination, extending the shelf life of the tomatoes, okra and eggplants by more than 7, 2 and 5 days, respectively compared with the uncoated fruits. The $log_{10}$CFU/g microbial counts for bacterial and fungal isolates revealed the following ranges for tomato (3.50 to 5.89, 3.43 to 5.88), okra (3.47 to 5.86, 3.46 to 5.88) and eggplant (3.50 to 5.89, 3.49 to 5.90), respectively. The predominant bacteria and fungi were Escherichia coli (30.18%) and Aspergillus niger (23.76%), respectively. Overall, the formulations were all effective in reducing spoilage, thereby extending the shelf life of the fruits.*

**Keywords:** Beeswax, Edible Coatings, Fruits, Plant antimicrobials, Shelf-life.

## INTRODUCTION

The preservation of fresh fruit, vegetables and other farm produce has remained a challenge, particularly in developing countries like Nigeria battling with epileptic public power supply. The search for novel post-harvest approaches to prevent spoilage resulting from microbial development in food has continued over the years aimed at preserving food quality, freshness, and safety (Poonia, 2018). The use ofantimicrobial packaging is a quality and safe approach to extend the shelf life of fruit and vegetables and lower the risk of foodborne infections (Poonia, 2018).

Edible coatings are a conventional practice to enhance food appearance and conservation. They are materials made of a mixture of biopolymers (polysaccharides, proteins and lipids) and various additives (plasticizers and chaotropic agents) dispersed in aqueous media that have a thickness of less than 0.3 mm (Embuscado and Huber, 2009; Montalvo et al., 2012; Castro-Muñoz and González-Valdez, 2019; Morales-Jiménez et al., 2020;

Dadzie et al., 2023). Edible coating effectively lessens fruit weight loss during processing, handling, and storage; additionally, they prolong fruit storage life and slow down the deterioration of fruits and vegetables by improving their quality. (Baraiya et al., 2012). Waxes, paraffin and glycerides are examples of lipid edible coatings characterized by their ability to extend shelf-life by preventing drying or dehydration of the edible film providing flexibility (Díaz-Montes and Castro-Muñoz, 2021). According to Quiros-Sauceda (2014) and Singh et al. (2020), coating agents made of lipid-based compounds provide the strongest barrier against water loss, because they are hydrophobic and are generally preferred for keeping vegetables and fruits. The limitation in using non-edible petroleum coating has increased interest in edible coatings made of waxes and lipids found in nature, including honey or beeswax (Rodrigues and Fernandes, 2012).

Fruits and vegetables have a shorter shelf life because they are metabolically active, hence are treated after harvest to reduce significant losses in quality and nutritious components.

Tomato (*Solanumlycopersicum*) is one of the most significant horticultural vegetable crops; with a global fresh weight production of 80 million tons from a planted area of roughly 3 million hectares (Kalyoncu et al., 2005). In Nigeria, tomatoes remain the most valuable edible vegetable in terms of income from its cultivation and processing (Anyanwu and Komolafe, 2003). Tomato has high levels of carbohydrates, dietary fiber, vital amino acids, vitamins, and minerals. While red tomatoes contain lycopene, an antioxidant that may help protect against carcinogens, yellow tomato varieties contain higher quantity of vitamin A than red tomato fruits (Seymour and Appleton, 2001; Ajayi and Olasehinde, 2009). A fungal plant pathogen can seriously damage tomato fruits (Khan et al., 2008). The presence of harmful toxins and bacterial spores are not usually observed until after a food poisoning outbreak followed by laboratory testing and other forms of investigation. The delay in identifying the pathogen makes bacterial contamination to be deadlier than it would have been if it was done earlier. (Oladipo et al., 2010). Tomato fruit has a very limited shelf life after harvest since 80–90% of its weight is made up of water. If the fruit's cuticle is removed, the water will evaporate, causing significant losses (Vipan et al., 2018). The accelerated rates of transpiration and respiration, fruit deterioration, and active metabolic processes all contribute to the constant alterations in tomato fruit quality after harvest (Pedreschi, 2017). In tropical climates, respiration is the primary factor influencing tomato post-harvest shelf life.

Okra, *Abelmoschus esculentus* (L.) Moench, family Malvaceae, is cultivated for its immature pods, which contain a gum that makes a thick, slimy mucilage used to thicken soups and stews as well as consumed as a vegetable. Okra can be kept for seven to ten days at 12.5$^{o}$C and 90% to 95% relative humidity, however due toits fragility and rapid respiration rate, fresh okra deteriorates swiftly during storage (Hardenburg et al., 1986). Most vegetable commodities like okra are held below 10$^{o}$C to minimize respiration and weight loss; however, okra is susceptible to chilling and develops black ribs, surface pitting, and water-soaked lesions when kept below 10$^{o}$C. (Ilker, 1976).

Garden egg (eggplant), *Solanum melongena* is an important member of the Solanaceae family grown in several African countries but is indigenous to sub-Saharan Africa (Kappel and Mozafarian, 2022). They are highly nutritious and feature prominently in food recipes in the tropics. However, losses in weight, firmness, and nutrients affect the quality of the fruit after harvest. Eggplants have a three-day maximum shelf life when stored at room temperature (Singh et al., 2016). It is susceptible to chilling when stored at temperatures below 7–10 $^{o}$C for an extended period of time (Concellón et al., 2012; Gao et al., 2015).

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The Rutaceae family, which includes citrus fruits, is one of the most widely produced tree crops in the world. *Citrus aurantifolia*(lime), is a medicinal plant that originated in Malaysia. It is also known by the names Mexican or West-Indian lime (Adebayo-Tayo et al., 2016). According to Norman (2015), lime juice consists of naturally occurring bioactive compounds that have antibacterial properties, notably saponins, alkaloids, tannins, phenolics, flavonoids, and terpenoids. Several studies have demonstrated the antibacterial activity of lime juice against *Staphylococcus epidermidis*, *S. aureus*, *Propionibacterium acnes*, *Salmonella* spp., *Bacillus* spp., *Escherichia coli*, *Klebsiella pneumonia*, *Shigella flexneri*, *Proteus* spp., *Pseudomonas* sp., and *Vibrio cholerae* (Onyeagba et al., 2004; Taiwo et al., 2007; Al-Ani et al., 2010; Ali, 2010; Aini et al., 2017).

Pectin is a name that refers to a group of heterogenous heterogeneous, high molecular weight, branched polysaccharides that are found in the cell walls of higher plants (Daoud et al., 2013). According to Perez-Vanquez et al. (2023), pectin is a generally recognized as safe (GRAS) molecule by the Food and Drug Administration (FDA). The initial awareness that pectin possess antibacterial properties was verifiedwas verified by El-Nakeeb and Yousef (1970a). The study expanded knowledge about pectin as far back as 1937 by showing that its antimicrobial properties is broad spectrum (El-Nakeeb and Yousef, 1970b; Men'shikov et al., 1997). The interest of many researchers on the physiological characteristics of this remarkably adaptable polysaccharide widely found in plants and fruits is on the increase. The antibacterial activities of pectin against Gram-negative bacteria (*Shigella vulgaris*, *Salmonella typhi*, *Salmonella paratyphi*, *Salmonella typhimurium*, *Klebsiella aerogenes*, *Escherichia coli*, *Proteus vulgaris*, *Bordetella bronchiseptica*, *Helicobacter pylori* and *Pseudomonas aeruginosa*), Gram-positive bacteria (*Streptococcus pyogenes*, *Staphylococcus lactis* and *Corynebacterium hofmannii*) and fungi (*Candida albicans*, *Saccharomyces cerevisiae*, *Aspergillus niger* and *Penicillium italicum*) are well documented (El-Nakeeb and Yousef, 1970a; Daoud et al., 2013). The aim of this study was to investigate the use of beeswax impregnated with lime and pectin as a potential coating to improve the quality and shelf life of tomatoes, okra and eggplants during storage.

## MATERIALS AND METHODS

### Source of the Samples

A total of one hundred and eighty (180) produce, comprising 60 each of tomatoes, okra and garden egg (eggplant) sourced from Rumuokoro market in Obio/Akpor Local Government Area, Rivers State, were employed in this study. All the samples were collected with a clean sterile polyethylene bag and taken to the laboratory. They were identified at the Herbarium of the Department of Plant Science and Biotechnology, University of Port Harcourt. Beeswax was obtained from local honey sellers from Taraba State and transported to Port Harcourt.

### Proximate analysis of beeswax

Proximate analysis of honey wax was carried out. The percentage of total carbohydrate, moisture, crude fat, crude protein, fibre and ash content were determined following the methods of AOAC (2005).

### Preparation of samples

The method described by Afedzi et al. (2022) was adopted with modification. The fruits were selected from uniform sizes and shapes, free from mechanical injury and spoilage signs. They were all washed with water that was boiled up to 100 ºC and allowed to cool, rinsed in sterile water and disinfected with 70% alcohol using sterile cotton wool. Beeswax weighing 2.2 kg was weighed and dissolved and filtered. Five treatments were employed in this study as follows: Wax (W) only (100%); pectin (P) only (100%); wax and lime (WL), (90:10%); wax and pectin (WP), (90:10%) and wax, lime and pectin

(WLP), (90:5:5%). Coating was carried out by dipping fruit in the gel for 5 min and leaving to dry under room temperature (29±2 °C) for 1 h. Uncoated fruits served as control. All treated samples and control were stored at ambient temperature (29±2°C). The fruits were monitored and analysis conducted in duplicates at 2-days intervals.

## Microbiological analysis

### Enumeration of bacteria and fungi

A sterile knife was used to cut each fruit and 10 g crushed in a sterilized mortar. The crushed fruit inside a mortal was transferred into a conical flask containing 90ml of sterile normal saline. The content of the conical flask was mixed properly before carrying out serial dilution up to $10^{-6}$. Thereafter, 0.1ml from $10^{-2}$, $10^{-3}$ and $10^{-4}$was then seeded on nutrient agar (NA) and potato dextrose agar (PDA). Incubation was done at room temperature for 24 h for bacteria isolation and 48-72 h for fungi. The colonies were counted and purified by sub-culturing onto freshly prepared sterile NA and PDA. The pure cultures obtained were stored on NA and PDA slant for identification purpose.

### Identification of the bacterial isolates

Bacterial isolates were characterized based on cultural morphology, physiological (Gram's staining) and biochemical characteristics [catalase, oxidase, indole, methyl-red, Voges Proskauer, citrate utilization, motility, coagulase, spore formation, TSIA ($H_2S$, gas and acid production) and utilization of glucose, lactose and sucrose] and compared with Bergey's Manual of Systematic Bacteriology (Cowan and Steel,1985; Holt et al., 2004).

### Identification of the Fungal isolates

This was based on the macroscopic and microscopic characteristics. Lactophenol cotton blue mount was carried out on fungal isolates for full identification of the exact fungi based on the size, shape, surface feature the conidia and hyphae arrangement (Barnett and Hunter, 1972; Larone, 1986).

## RESULTS AND DISCUSSION

### Proximate composition of beeswax

The results of the percentage proximate composition of the beeswax presented in Table 1 reveals the highest carbohydrate content (46.25%), followed by lipid (38.50%) and the least was ash (0.53%). To the best of our knowledge, there are no reports on the proximate composition of beeswax in available literature which focused only on honey. What is however, consistent is that the carbohydrate was highest in both the beeswax and honey. The lipid content of beeswax is far higher than what was obtained in honey. A number of authors have reported percentage proximate composition ranges for carbohydrate (76.44±0.64 – 86.89±0.93), protein (0.46±0.09 – 3.14±0.20), ash (0.107 – 1.73±0.74), lipid (0.013 – 3.44±0.42), fibre (0.002 – 2.03±0.08) and moisture (9.39±0.05 – 20.14±0.04) (Wasagu et al., 2013; Buba et al., 2013; Adeniyi et al., 2014; Bamaiyi et al., 2019; Aneni et al., 2023). The nutritional composition of beeswax may promote the growth of microorganisms with the film reducing transpiration.

Table 1. Percentage proximate composition of beeswax

| Component | Concentration (%) |
|---|---|
| Carbohydrate | 46.25 |
| Lipid | 38.50 |
| Moisture | 8.45 |
| Protein | 5.25 |
| Fibre | 1.02 |
| Ash | 0.53 |

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**Total bacterial count and total fungal count of coated and uncoated fruits during storage.**

The total bacterial count (TBC) of coated tomatoes + uncoated sample (control), okra + uncoated sample (control) and eggplants + uncoated sample (control) are presented in Figures 1, 2 and 3 while the corresponding total fungal count (TFC) are depicted in Figure, 4, 5 and 6, respectively.

Generally, the TBC and TFC of the control samples were very high prior deterioration of all the fruits. For the coated tomatoes (Fig 1) and okra (Fig. 2), bacterial growth at day 20 was highest in samples coated with the wax + lime + pectin, followed by wax + lime while the least was wax + pectin. The highest bacterial growth for the eggplant (Fig. 3) was observed jointly in wax + lime + pectin, wax only and wax + pectin and the least growth was recorded in the samples coated with only pectin.

The TFC of coated tomatoes in day 20 in coated tomatoes (Fig. 4) was highest in wax + lime + pectin followed by pectin only and the least was wax + pectin. For coated okra the fungal growth at day 20 was highest jointly in wax + lime + pectin, wax only and pectin alone, followed by wax + lime and least was wax + pectin (Fig. 5). In coated eggplant, fungal growth was highest in samples coated with wax + lime + pectin, followed by wax + pectin and least growth was observed in samples coated with pectin (Fig. 6).

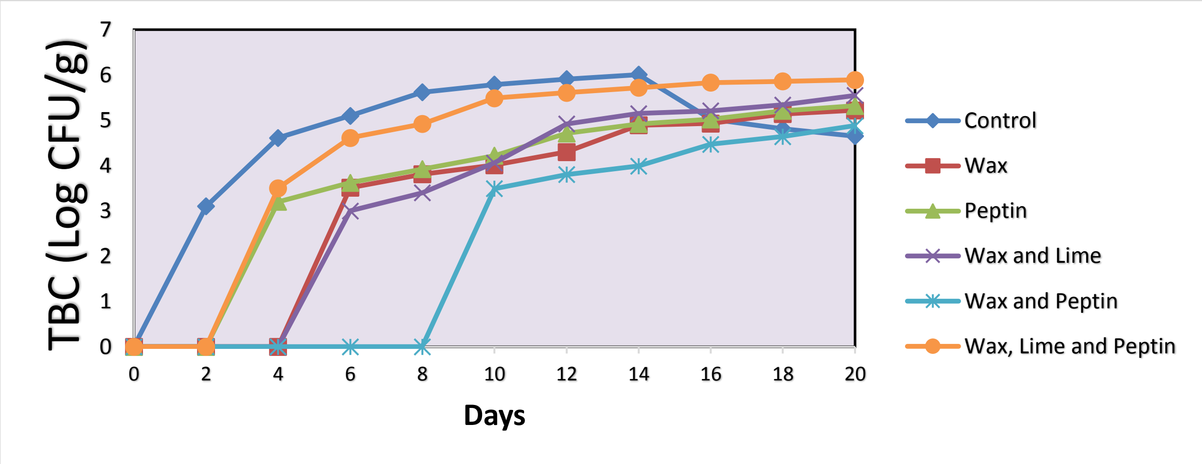


Figure 1: Total bacterial count of coated and uncoated tomatoes stored for 20 days at room temperature

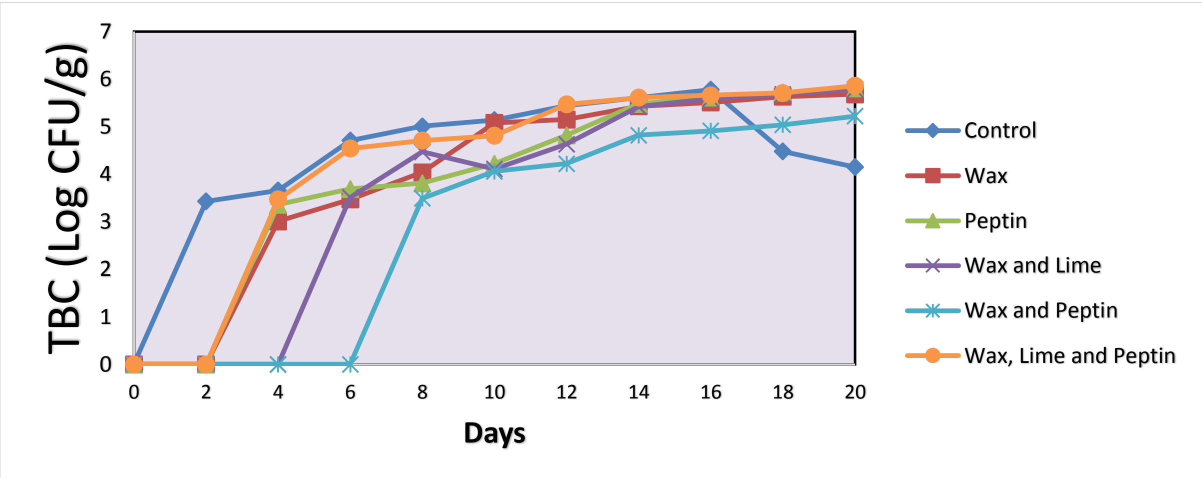


Figure 2: Total bacterial count of coated and uncoated okra stored for 20 days at room temperature

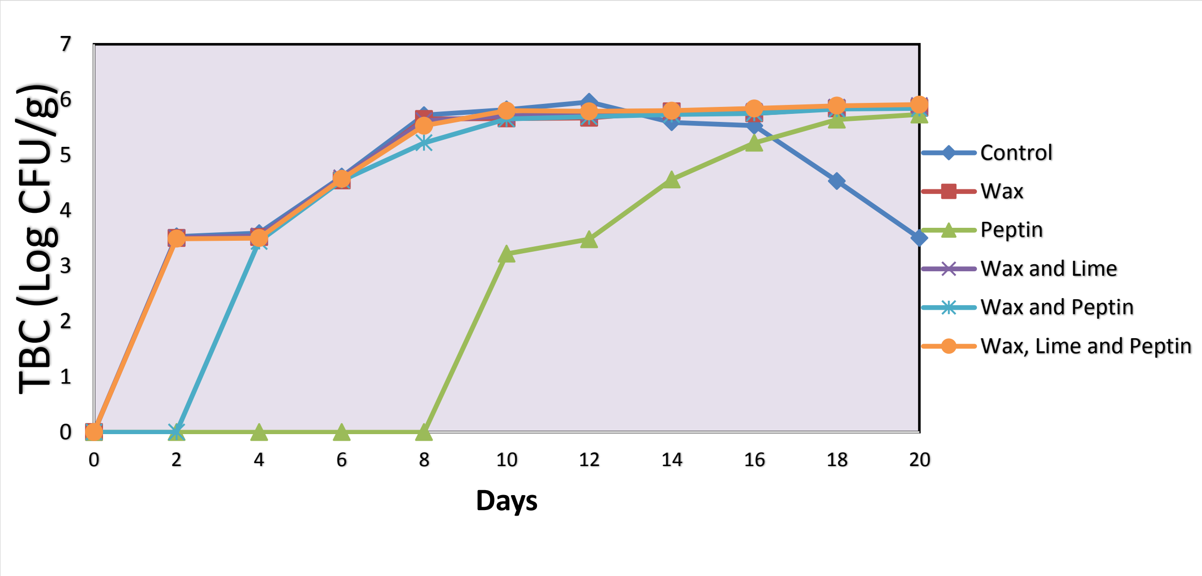


Figure 3: Total bacterial count of coated and uncoated eggplant stored for 20 days at room temperature

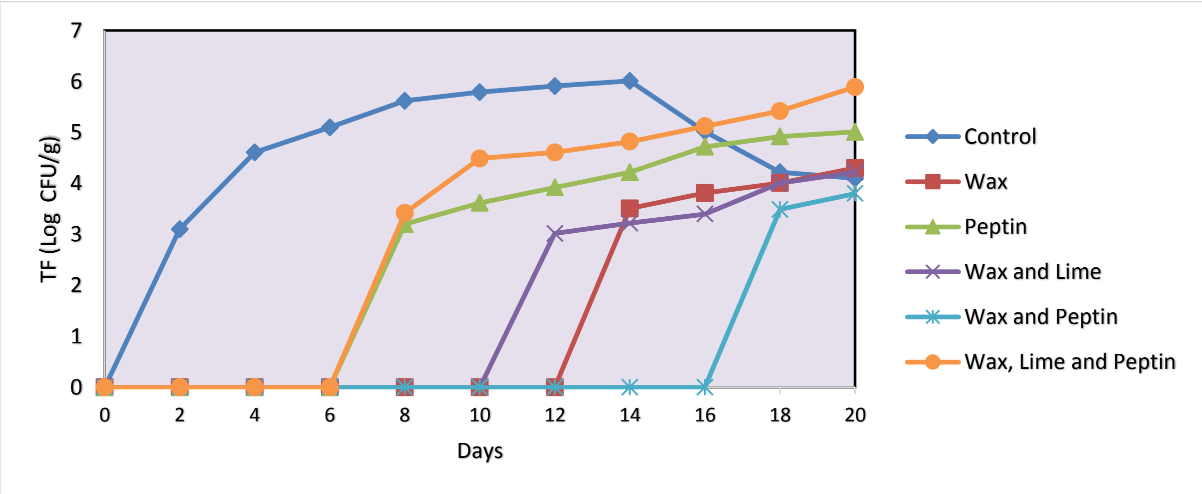


Figure 4: Total fungal count of coated and uncoated tomatoes stored for 20 days at room temperature.

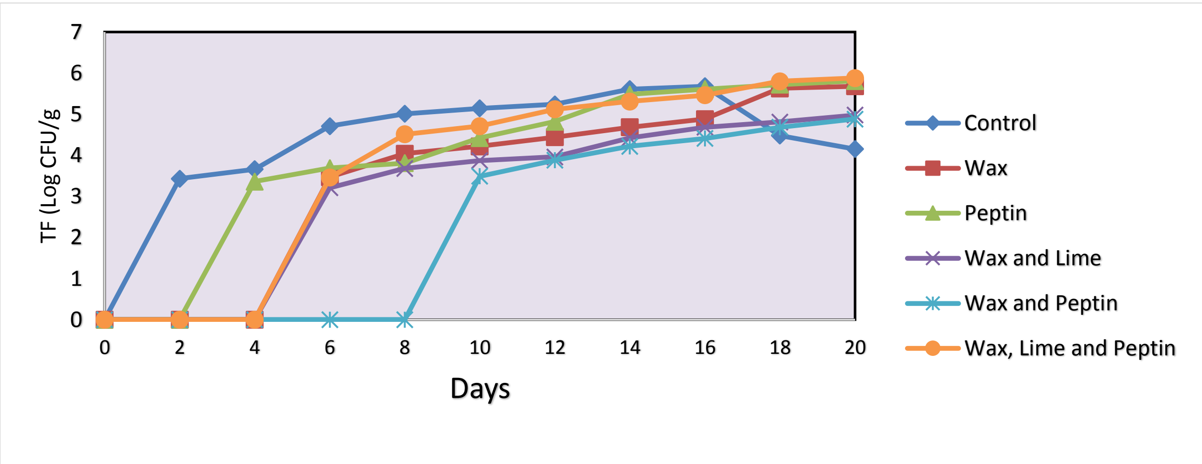


Figure 5: Total fungal count of coated and uncoated okra stored for 20 days at room temperature.

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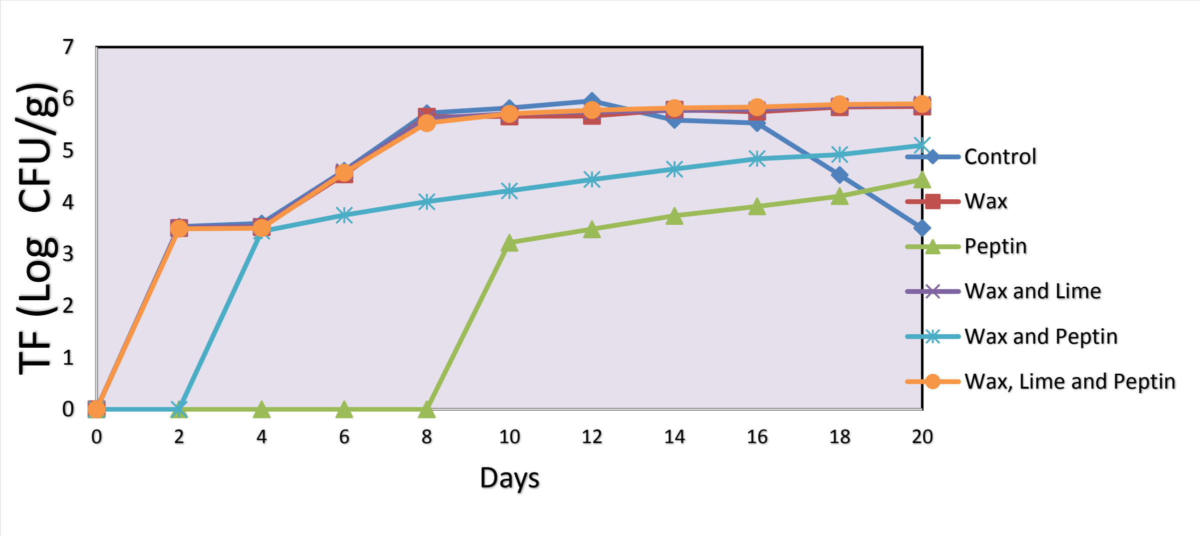


Figure 6: Total fungal count of coated and uncoated eggplant stored for 20 days at room temperature.

Generally, edible coatings are used to improve the overall look of fruits, vegetables, and minimally processed fruits during storage. In addition, it reduces moisture and scent loss, and delay colour changes. (Antunes et al., 2012). The results of the keeping time (days) of the control (uncoated fruits) and coated fruits is presented in Table 2. The wax + pectin combination gave the best result for all fruits, followed by pectin only and the least was wax + lime + pectin. Overall, the treatments were all effective in extending the shelf life of the fruits in comparison with the control samples, particularly tomato. The finding of this study is in agreement with the report by Alvarez et al. (2022), that pectin-beeswax coating incorporated eugenol might be a profitable commercial method to avoid deterioration and maintain the quality of citrus fruit. The results of the effectiveness of pectin alone in the extension of the shelf life of fruits in this present study is a corroboration of previous report by Plesoianu and Nour (2022) that the best control of weight loss throughout storage was obtained for the samples coated with pectin alone of fresh-cut pears during 15-day storage at 8$^oC$.In another study that involved the use of pectin-based edible coating mixed with corn flour and beetroot powder in different proportions incorporated into tomatoes and stored for 30 days at 25 $^oC$, Sucheta et al. (2019) reported that the coating formulated with 50% pectin and 50% corn flour (PCF) achieved the best weight loss, decay percentage, respiration rate, and biochemical quality. Furthermore, the researchers reported that 100% pectin coating could maintain maximum glossiness and minimum shrinkage of the tomato's pericarp without causing off-flavours. Breceda-Hernandez et al. (2020) reported that pectin edible coating with lemon essential oil has potential to reduce post-harvest losses of Red Globe grapes during their storage at 4°C because it prolonged the grapes' shelf life by 35 days. The application of a pectin-based coating containing a lemon byproduct extract was reported to preserve the physiological parameters in carrots, reduced their physiological activity and, thus, delayed senescence for up to 14 days (Imeneo et al., 2022). The number of days that WL (Wax + Lime) formulation preserved the three fruits is in agreement with the report by Pudjiastuti et al. (2021) which stated that addition of lime extract as an antibacterial agent to edible film made from cassava starch did not significantly affect the film's physical or chemical qualities, therefore, it was a viable option for food packing. Fisher and Phillips (2008) posited that citrus oils can be used as antimicrobials in the food business since they exhibit inhibitory action against a variety of Gram-positive and Gram-negative bacteria.

Table 2: Shelf-life of the coated and control samples

| Treatment | Spoilage time (days) | | |
|---|---|---|---|
| | Tomatoes | Okra | Eggplant |
| Control | 7 | 7 | 4 |
| Wax | 17 | 12 | 10 |
| Pectin | 14 | 12 | 18 |
| Wax + lime | 16 | 10 | 10 |
| Wax + pectin | 30 | 28 | 15 |
| Wax + lime + pectin | 14 | 9 | 9 |

In this study 2.2 % beeswax extended the shelf life of eggplant stored at room temperature (29±2ºC) for 10 days. This is comparable to the shelf life of eggplant stored at 10ºC for 17 days using3% beeswax reported by Dadzie et al. (2023). The researchers reported that the coating significantly minimized weight loss, preserved firmness, and delayed colour changes.

**Microorganisms isolated from the coated and uncoated fruits**

The $log_{10}$CFU/g microbial counts for bacteria and fungi revealed the following ranges for tomato (3.50 to 5.89, 3.43 to 5.88), okra (3.47 to 5.86, 3.46 to 5.88) and eggplant (3.50 to 5.89, 3.49 to 5.90), respectively. The microorganisms isolated from the fruits employed for this study in course of the storage period are presented in Table 3 and 4, respectively for bacteria and fungi. The predominant bacteria and fungi were *Escherichia coli* (30.18%) and *Aspergillus niger* (23.76%), respectively. Nasiru and Dalhatu (2020) reported predominant occurrence of *Aspergillus niger* and *Staphylococcus aureus* in eggplants sold in Sokoto metropolis, Nigeria. A number of authors have reported the occurrence of these bacteria and fungi amongst several others from tomato (*Bacillus cereus*, *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas* spp., *Klebsiella* spp., *Staphylococcus aureus*, *Aspergillus* spp., *Rhizopus* spp., *Candida* spp., *Mucor mucido*, *Penicillium* spp. And *Fusarium* spp.) in Ghana, India and Nigeria (Ghosh, 2009; Aernan et al., 2014; S. Mbajiuka and Enya, 2014; Obong et al., 2018; Danaski et al., 2022); okra (*Bacillus subtilis. Staphylococcus aureus*, *Clostridium* spp., *Pseudomonas aeruginosa*, *Escherichia coli. Klebsiella pneumuniae, Rhizopus stolonifera*, *Aspergillus niger, A. fumigatus, A. flavus, Penicillium*spp.*, Mucor pusillus* and *Candida crusei*) in India and Nigeria (Sharma et al., 2013; Adegbehingbe, 2014; Ibrahim et al., 2021; Ogunoye et al., 2023) and eggplants (*Escherichia coli*, *Salmonella* spp., *Staphylococcus aureus*, *Bacillus* spp., *Clostridium botulinum*, *Aspergillus* spp., *Candida tropicalis*, *Mucor* spp., *Fusarium spp.* and*Rhizopus*) in India and Nigeria (Jidda and Musa, 2016; Yaji et al., 2016; Kothadiya, 2019; MikeAnosike et al., 2019, Nasiru and Dalhatu, 2020).

Table 3: Prevalence of bacterial isolate from the tomatoes, okra and eggplant

| Bacteria | No of Isolate Obtained | % Isolate Obtained |
|---|---|---|
| *Escherichia coli* | 144 | 30.18 |
| *Pseudomonas* spp. | 48 | 10.06 |
| *Salmonella* spp. | 42 | 8.80 |
| *Bacillus* spp. | 90 | 18.86 |
| *Staphylococcus aureus* | 126 | 26.41 |
| *Clostridium* spp. | 24 | 5.03 |
| *Klebsiella* spp. | 3 | 0.62 |
| Total | 477 | 100 |

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Table 4: Prevalence of fungi isolate from the tomatoes, okra and eggplant

| Fungi | No of Isolate Obtained | % Isolate Obtained |
|---|---|---|
| *Aspergillus niger* | 144 | 23.76 |
| *Fusarium* spp. | 108 | 17.82 |
| *Mucor* spp. | 90 | 14.85 |
| *Penicillium* spp. | 84 | 13.86 |
| *Rhizopus* spp. | 72 | 11.88 |
| *Candida* | 60 | 9.90 |
| *Aspergillus* spp. | 40 | 6.60 |
| Total | 606 | 100 |

## CONCLUSION

The findings of this study revealed that beeswax and pectin or lime edible coating provides a promising solution in extending the shelf life and quality of fresh produce. The wax and pectin combination were more effective during the storage of all three fruits.

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