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Comparative Studies on Bio-Preservation of Fresh-Cut Tomatoes Using Garlic, Ginger and Lemongrass Infused in Coconut Oil

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ABSTRACT
Food spoilage is a major issue in our economy today, especially due to global climate change. Recent climate trends have led to an increased deterioration of freshly cut fruits and vegetables and has resulted in series of food poisoning as well as other health risk. Aside this, food spoilage and deterioration has affected negatively on national economy as from farm to store, supply chains are struggling with a food waste issue. This research aimed at using bio-products in reducing the spoilage rate of fresh-cut tomatoes using lemongrass oil, ginger oil and garlic oil under 28OC and 4OC ambient temperatures. The bio-products (ginger oil, garlic oil and lemon grass oil) were extracted using domestic methods with coconut oil as the base oil. The bio-products were applied on the freshly cut tomatoes once daily. Visual observations on some anatomical features of tomatoes such as cuticle, placenta and pericarp were used to study rate and spoilage duration of tomatoes, while weight of the tomatoes at intervals and microbiological parameters were used in analyzing the effectiveness of the bio- products in reducing tomatoes spoilage rate. After 96 hours of study, the results showed significant ($p<0.05$) reduction in tomatoes spoilage rate in ginger oil assisted tomatoes compared to other bio-products and the control. Signs of pericarp wrinkle, cuticle deterioration and placenta damage were observed in the control from 24 hours. Similarly, significant reduction in tomatoes weight were observed in the control tomatoes which indicated spoilage. High fungal and bacterial count was also observed in the control tomatoes and the microbes observed showed some similarities with previously identified microbes that are associated with fruit spoilage and food poisoning. This research showed that ginger oil, garlic oil and lemon grass oil could be effective in reducing spoilage in freshly cut tomatoes, however future studies should be conducted to investigate the affectivity of other bio- products in reducing fruit spoilage.

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Introduction
Consumer desires for freshly cut fruits and vegetables that are ready to use are growing, necessitating extensive research in this field. Fruit rotting has become more common because of the high temperatures brought on by climate change [1]. The tomato is one of the horticulture goods that customers want since it is a source of vitamin C and minerals, as well as being useful as a vegetable, table fruit, juice, and beverage. One product that is deemed vulnerable to harm is tomatoes (perishable). Regular tomato consumption has been linked to a decreased risk of developing cancer, cardiovascular diseases, and chronic illnesses, including cataracts, asthma, and bronchitis, which have been attributed to the presence of antioxidant compounds [2-4].

Crops' antioxidant capacities may be influenced by pre-harvest elements like genetic make-up and cultural norms. The phenolic content and antioxidant power of fruits are significantly impacted by processing. Ascorbic acid content is reduced during the thermal process of processing tomatoes to produce tomato juice, baked tomatoes, tomato sauce, and tomato soup. However, the capacity of the water-soluble antioxidant system, total phenolic compounds, and lycopene increases [5]. Postharvest losses of tomato fruit can range from up to 35.5%, depending on the distribution chains, and are primarily caused by inappropriate handling [6]. Physical, physiological, mechanical, and microbial harm are all included in postharvest losses. Therefore, it is crucial to make the required steps to preserve fruit quality and lengthen the shelf life of fresh tomatoes.

Consumers have long placed a premium on food quality and safety, and today's food systems must still meet these requirements as a matter of course. Since the inception of the food industry, chemical preservatives (synthetic additives) have been employed to reliably reduce the number of causes that cause fresh-cut fruits and vegetables to deteriorate. However, the majority of these substances do not adhere to the consumer's preferred definitions of "natural" and "healthy," thus the food business must give alternatives [1]. Agro-industries, governments, and consumer organizations all around the world stress the importance of this. Food spoilage is a global agricultural issue and it has been associated to variety of factors.

Extensive work has been done in trying to see the possibilities of reducing food spoilage. For example, a study by deduced that around 30% of food produced for people worldwide is lost at some point along the food supply chain. This is catastrophic, especially because the world's population is expected to grow to around 9.8 billion people by 2050, and we will require a 70% increase in food availability [7,8].

Materials and Methods

This Study Followed the Procedures Below

Samples Preparation

Thirty (30) fresh and undamaged tomatoes was obtained from Ogbeogonogo market (6.128 °N, 6.445 °E) and washed with sterile water. The washed tomatoes were then cut from the epicarp in such that it will be divided into two equal parts. The fresh-cut tomatoes were preserved under two temperature regimes (4 and 28 degrees) and then monitored until spoilage.

Production of Bio-Products

Pure and undiluted coconut oil (trade name: Tropical Sun) was purchased from Okumu oil palm company Plc in Benin City Nigeria. The coconut oil was used as the base oil in the extraction of lemon grass oil, ginger oil and garlic oil following the methods as described below.

Garlic Oil Extraction

The garlic oil was extracted following domestic extraction procedure as described by [9]. About 40 g of fresh garlic was weighed and then chopped finely in a food processor and transferred to a small stainless-steel pot (20 x 10 cm) and 100 ml of the base oil was added (Coconut oil). This set up was heated at low heat (60 to 90 degrees) and stirred constantly to ensure homogeneity. The heated sample was observed for 15 -20 minutes as the colour changes as sign of garlic oil extraction. The extracted garlic was stored in an airtight glass container (5 x 5 cm) in room temperature and used once in two days on the fresh cut tomatoes.

Ginger Oil Extraction

The ginger oil was extracted following domestic extraction procedure as described by Resham and Bhatkhandeb (2016). About 40 g of fresh garlic was weighed and then chopped finely in a food processor and transferred to a small stainless-steel pot (20 x 10 cm) and 100 ml of the base oil was added (Coconut oil). This set up was heated at low heat (60 to 90 degrees) and stirred constantly to ensure homogeneity. The heated sample was observed for 15 -20 minutes as the colour changes as sign of garlic oil extraction. The extracted garlic was stored in an airtight glass container (5 x 5 cm) in room temperature and used once in two days on the fresh cut tomatoes.

Lemon Grass Oil Extraction

The lemon grass oil was extracted following domestic extraction procedure as described by [10]. About 50 g of fresh lemon grass was weighed and then chopped finely in a food processor and transferred to a small stainless-steel pot (20 x 10 cm) and 100 ml of the base oil was added (Coconut oil). This set up was heated at low heat (60 to 90 degrees) and stirred constantly to ensure homogeneity. The heated sample was observed for 20 minutes as the colour changes as sign of garlic oil extraction. The extracted garlic was stored in an airtight glass container (5 x 5 cm) in room temperature and used once in two days on the fresh cut tomatoes.

Application of Bio-Products

The three bio-products were applied on the fresh-cut tomatoes using dropper at each sample of the freshly-cut tomatoes under the two environmental temperature regimes. The three bio- product oils were applied once in two days until signs of spoilage was observed.

Rate and Duration of Spoilage

The rate and duration of spoilage was determined virtually using spoilage-duration plot. To obtain this plot, the tomatoes were observed at every 12 hours interval.

Weight Measurement

Weight of tomatoes was measured on a scale, every 12 hours interval.

Microbiological Analysis

Microbial Population

Microbial population was investigated before the application of the bio-products using serial dilution method. In this method, the culture was poured in a test tube, and six test tubes, each with 9ml of sterile diluent (water) were taken. A sterile pipette was taken and 1ml of properly mixed culture sample/culture was drawn into the pipette. The sample was then added to the first tube to make the total volume of 10 ml. This provides an initial dilution of 10^{-1}. The dilution was thoroughly mixed by emptying and filling the pipette several times. The pipette tip was discarded, and a new pipette tip was attached to the pipette. Now, 1 ml of mixture was taken from the 10^{-1} dilution and is emptied into the second tube. The second tube now has a total dilution factor of 10^{-2}. The same process was then repeated for the remaining tube, taking 1 ml from the previous tube and adding it to the next 9 ml diluents. As six tubes are used, the final dilution for the bacteria/cells will be 10^{-6} (1 in 1,000,000).

Bacterial Enumeration

The standard or viable plate count or colony count method of enumeration was carried out. This is a viable count, not a total cell count. It reveals information related only to viable or live bacteria. Using this method, a small volume (0.1 - 1.0 mL) of the 10^{-2}, 10^{-4} and 10^{-6} dilution was measured into a different sterile petri plates, nutrient agar and MacConkey agar was poured into the plate using creating a "pour plate." The plates were incubated at 37°C for 24 hours. During that time, each individual viable bacterial cell multiplies to form a readily visible colony. The number of colonies were then counted with a colony counted and this number should equal the number of viable bacterial cells in the original volume of sample, which was applied to the plate.

Fungal Isolation

1ml of 10^{-2}, 10^{-4} and 10^{-6} of the serials dilutes were poured into labelled Petri plates using respective pipettes. Molten, cooled (45°C) Potato dextrose agar medium was poured into the petri plates and the plates were rotated gently to ensure uniform distribution of cells in the medium. The plates were then allowed to solidify. The inoculated plates were incubated at 37°C for 3-5 days and examined.

Enumeration of Fungal Isolates (TFC)

The discrete colonies of viable fungi cells on PDA medium were counted after 72hr. The total fungal colony (TFC) was determined using the formula below;

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$$\text{TFC (cfu/g)} = \frac{\text{No. of colonies on plate} \times \text{Dilution factor}}{\text{Volume of aliquot}}$$

Identification of Fungal Isolate

The isolated fungi colony were identified using the morphological characteristics (macroscopy) of the colony. The presence or absence of aerial mycelium, the colour, wrinkles, furrows and any other pigment and the macro morphological characters were evaluated [11].

Differential Media

Culture/sample were inoculated with MacConkey agar using sterile swab and was then incubated at 37°C for 18 hours. Microbial colonies were differentiated after incubation.

Physiological Examination of Bacteria Colony

Different bacterial colonies were distinguished based on physical observation.

Results

Rate and Duration of Tomato Spoilage

The table below shows the rate and duration of spoilage in tomatoes throughout the period of study. This result was based on some selected anatomical features of tomatoes that has been associated with spoilage. At the first 24 hours, it was observed that all the assayed anatomical features of the tomatoes were intact at both 4°C and 28°C, with exception of the control where the pericarp and placenta started showing signs of wrinkling at 12 hours. However, at 36 hours, it was observed that the tomatoes cuticle, placenta and pericarp in the lemon grass oil assisted tomatoes at 28°C showed signs of wrinkling. Similar result was observed in the garlic oil assisted tomatoes. The ginger oil assisted tomatoes under 4°C was observed to still show intact pericarp, placenta and cuticle even at 72 hours. But all the tomatoes samples spoilt at 96 hours.

Table 1: Rate and Duration of Tomato Spoilage

Sample s	Ambient temperature (°C)	12 hours	24 hours	36 hours	48 hours	60 hours	72 hours	84 hours	96 hours
LG tomatoes	4	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUW, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW
	28	CUI, PLI, PEI	CUI, PLI, PEI	CUW, PLW. PEW	CUW, PLW, PEW	CUW, PLW. PEW	CUW, PLW. PEW	CUW, PLW, PEW	CUW, PLW, PEW
GR tomatoes	4	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLW, PEW	CUW, PLW. PEW	CUW, PLW, PEW	CUW, PLW, PEW
	28	CUI, PLI, PEI	CUI, PLI, PEI	CUW, PLW. PEW	CUW, PLW. PEW	CUW, PLW. PEW	CUW, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW
GIN tomatoes	4	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI. PEI	CUW, PLW, PEW	CUW, PLW, PEW
	28	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUW, PLW. PEW	CUW, PLW, PEW	CUW, PLW, PEW
Control	4	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUI, PLI, PEI	CUW, PLW. PEW	CUW, PLW. PEW	CUW, PLW, PEW	CUW, PLW, PEW
	28	CUI, PLI, PEI	CUI, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW	CUW, PLW, PEW

CUI= Cuticle is Intact, CUW=Cuticle is Wrinkled, PLI = Placenta is Intact, PLW=Placenta is Wrinkled, PEI = Pericarp is Intact, PEW= Pericarp is Wrinkled.

This table below shows the weight of tomatoes throughout the 96 hours period of study. At 4 °C ambient temperature, it was observed that the tomatoes treated with ginger oil maintained its original weight (7 g) up to 72 hours however, the weight started diminishing at 84 hours after the application of bio-product. Meanwhile for the control at 28°C, the weight of tomatoes started diminishing right from 24 hours.

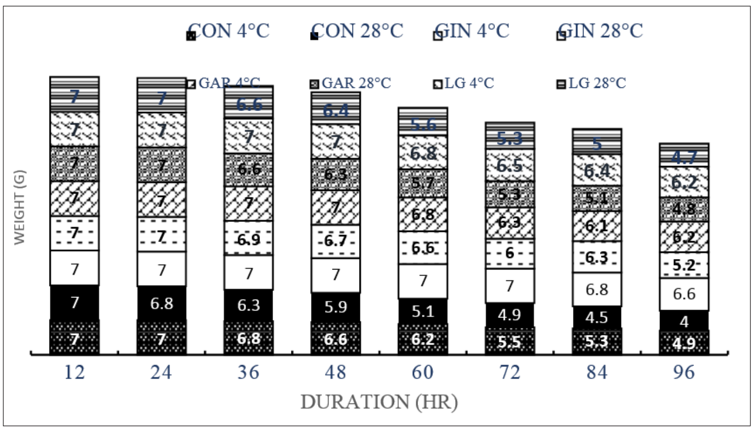

Table 2: Chart of Weight and Duration of Fresh-Cut Tomatoes

Microbiological Analysis

Total Fungal Count

Table 3 shows the fungal count of the assayed tomatoes at all temperatures. It was observed that the control tomatoes at 28⁰C had the highest (60 x 10^6) fungal count, while the ginger oil assisted tomatoes at 4°C had the lowest (6 x 10^5). Fungi such as Nirosporo spp, Aspergillus flavus, Absidia spp, Aspergillus fumigatus, Mucor spp were observed in the tomatoes at 6 hours after the application of the bio-products. Table 4.3 showed the fungal count of the experimental tomatoes after 120 hours of the bio-products application. It was observed that the control at 28⁰C had the highest (9 x 10^6) fungal count meanwhile, the tomatoes assisted with ginger oil at 28⁰C was observed to show lowest (10 x 10^5) fungal count. Furthermore, suspected fungal that were present in the tomatoes were Aspergillus tamari, Nigrospora spp. and Mucor spp. among others.

Table 3: Morphological Properties of Fungal Isolates at 6 Hours after Application of Bio-Products

Isolates	Macroscopy	Possible Identity	Fungi count (cfu/g)
GIN in 28°C	Woolly colonies. Gray-green with fluggy colonies	*Nigrospora* spp. *Aspergillus fumigatus*	9×10^5
GIN in 4°C	Light green and powdery Flat, woolly to cottory brown Creamy colonies	*Aspergillus flavus Absidia* spp. *Fusarium* spp.	6×10^5
Control in 28°C	Light green and powdery Flat, woolly to cottory brown	*Aspergillus flavus* *Absidia* spp.	60×10^6
Control in 4°C	Light green and powdery Flat, woolly to cottory brown Creamy colonies	*Aspergillus flavus Absidia* spp. *Fusarium spp.*	16×10^5
LG in 28°C	Gray-green with fluggy colonies Creamy colonies	*Aspergillus fumigatus Fusarium* spp.	50×10^5
LG in 4°C	Whitish/light cotton like Light green and powdery Flat, woolly to cottony brown Creamy colonies	*Mucor spp. Aspergillus flavus* *Absidia* spp. *Fusarium* spp.	9×10^6
GR in 28°C	Gray-green with fuggy colonies	*Aspergillus fumigatus*	15×10^6
GR in 4°C	Gray-green with fluggy colonies	*Aspergillus fumigatus*	7×10^6

GR=Ginger Oil, GAR=Garlic Oil, LG=Lemongrass Oil

Table 4: Morphological Properties of Fungal Isolates at 120 Hours after Application of Bio- Products

Isolates	Macroscopy	Possible Identity	Fungi count(cfu/g)
GIN in 28°C	Brown and cottony- like Woolly colonies. Olive brown colonies Gray-green with fluggy colonies	*Penicillium* spp. *Nigrospora* spp. *Aspergillus tamarii Aspergillus fumigatus*	15×10^5
GIN in 4ᴼC	White and woolly colony olive brown colour	*Nigrospora* sp. *Aspergillus tamarii*	10×10^5
Control in 4°C	Light green and powdery	*Aspergillus flavus*	9×10^6
Control in 28°C	Light green and powdery Woolly colonies White mucelia with brown spores	*Aspergillus tamarii* Nigrospora spp. *Mucor* spp.	25×10^5
LG in 4°C	Light green and powdery Brown and cotton- like Creamy colonies	Aspergillus tamarii Penicillum spp. Fusarium spp.	6×10^6
LG in 28°C	White mycelia with green spore Gray-green with fluggy colonies Whitish and light cotton-like colonies	*Aspergillus* spp. *Aspergillus fumigatus* Mucor spp.	27×10^5
GR in 28°C	White mycelia with green spore White woolly colony Flat woolly to cotton white	*Aspergillus spp.* *Trichoderma spp. Absidia spp.*	15×10^5

Total Bacterial Count

Table 6 showed the bacterial count of the assayed tomatoes at all temperatures. It was observed that the control tomatoes at 28°C had the highest (9 x 10^6) bacterial count, while the ginger oil assisted tomatoes at 4°C had the lowest (2 x 10^6). Table 4.4 showed the bacterial count of the experimental tomatoes after 120 hours of the bio-products application. It was observed that the control at 28°C had the highest (9 x 10^6) bacterial count meanwhile, the tomatoes assisted with ginger oil at 28°C was observed to show lowest (2 x 10^6) bacterial count.

Table 6: Morphological Properties of Bacterial Isolates at 6 and 120 Hours after Application of Bio-roducts

Isolates	Ambient temperature (°C)	Bacterial count (cfu/g) at 6 hours after application of bio- products	Bacterial count (cfu/g) at 120 hours after application of bio- products
GR tomatoes	4	5×10^6	2×10^6
	28	3×10^5	8×10^6
LG tomatoes	4	2×10^6	2×10^6
	28	5×10^5	4×10^6
GIN tomatoes	4	1×10^5	2×10^6
	28	2×10^5	3×10^6
Control tomatoes	4	3×10^5	6×10^6
	28	9×10^5	9×10^6

Discussion

Tomato is an important food crop, characterized by high consumption numbers worldwide, and numerous uses and health benefits [12]. As a fresh produce commodity, tomato has a relatively short shelf life and its quality is affected by many pre and postharvest factors. During postharvest handling many parameters can influence tomato's quality attributes resulting in rapid deterioration. In this study, natural bio-products such as garlic oil, ginger oil and lemon grass oil were observed to increase the postharvest duration of tomato compared to the control. This result is consistent with the work of who reported increased postharvest duration when tomatoes was dipped in 0.5% sage groundnut oil compared to 0.1% groundnut oil and control fruits after seven and 14 days of storage at 11°C [13].

The first postharvest deterioration that was observed in the control tomato sample at 24 hours under 28 degree may be due to the lack of bio-product in the tomatoes. Permeability of fruit skin is the major factor that causes deterioration during during storage [12]. The application of oil containing substances improve permeability and therefore increase postharvest duration. This result is consistent with the work of where they observed fast deterioration of tomato placenta without any oil amendment [12].

The 10% increase in fungal count in the control tomatoes under 28 degree at every assayed date in comparison with the bio-products oil assisted tomatoes may also indicate fast postharvest spoilage. Microbes are known to invade tomato samples and bring about spoilage. also indicate signs of spoilage. These results showed that the three bio-products (garlic oil, ginger oil and lemongrass oil) used in this research has been able to suppress microbial growth under all the temperatures [14]. According to, ginger oil has been documented to inhibit microbial growth in vegetables and increase postharvest spoilage duration. Fungi such as Nirosporo spp, Aspergillus flavus, Absidia spp, Aspergillus fumigatus, Mucor spp were observed. These fungi are observed to be similar species identified by during postharvest preservation of spinach [15,16].

Conclusion

Results from this research indicated the effectiveness of bio-products (ginger oil, garlic oil and lemon grass oil) in reducing the spoilage rate of freshly cut tomatoes under 40C and 280C. The significant reduction in tomatoes weight in the control showed how deterioration in tomatoes with bio-products application can be easily achieved. Furthermore, increase microbial counts in the control showed how easy microorganisms can colonize freshly cut tomatoes without the application of bio-products. This research would help in reducing the global spoilage of fruits and improve national exports.

Recommendations

Even though the ginger oil treated tomatoes in this study performed better than other bio- products, it is recommended that future studies should be done in order to check the effectiveness of other bio-products. Also, biochemical as well as DNA based identification of microorganisms should be carried out to ascertain specific strains of bacteria that are associated with tomatoes spoilage.

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