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Effect of biofilm-hybrid mixing ratio and incubation time on tomato fruit quality parameter coated with *MLECB*

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Abstract
The purpose of the study was to determine how well moringa oleifera leaf extracts and chitosan edible bio-preservative (MLECB) extended the shelf life of fresh tomato fruits. The existence of imines (C=N) at 1661.8 cm-1, as shown by functional group analysis, confirmed the cross-linking reaction between the -NH2 of chitosan and the carbonyl groups of MLE. Gas chromatography mass spectrometry (GC-MS) hydrocarbon and phytochemical investigation revealed that MLE contains a high concentration of antimicrobial hydrocarbons (Pentadecane = 18.46%; Tridecane = 12.386%; Heptacosane = 12.013 %). In a similar vein, it was discovered that MLECB included notable antioxidants, such as quercetin (10.265%). Following a 20-day period of non-refrigerated storage at 1% chitosan content and a 40:60 MLECB ratio, the tomato fruit retained a 2.1706 g/kg titratable acidity (TA) and a 20.01% decrease in weight loss.

Keywords: Bio-preservation, biofilm, coating, tomato fruit, moringa oleifera leaf extract.

1. Introduction

Consumers are consuming more fresh, naturally grown fruits and vegetables, yet the food industry is facing significant challenges worldwide with regard to how to preserve these crops (Kayode and Afolayan, 2014). Tomatoes (*Solanum Cycopersium*), are one of the most extensively cultivated vegetable crops worldwide (Mahfoudhi et al., 2014). Because tomatoes are available year-round in most tropical regions of the entire globe, about 130 mega tons of tomatoes are produced globally each year. Tomato is rich in vitamins (such as vitamin A, vitamin C), carbohydrate, protein, fibre and potassium (Ruelas-Chacon et al., 2017). It is also rich in lycopene which has several health benefits such as, the avoidance of malignancies, and improvement in the skin's capacity to protect itself from the sun's damaging ultraviolet rays (Tarangini et al., 2022). Tomato is considered a fruit that is "easy to damage" because of its climacteric traits, which include continual ripening after harvest. Depending on the method of preservation, untreated tomato fruit has a 4–8 day shelf life at room temperature (Pinheiro et al., 2015, Nasrin et al., 2008). Nigeria is the 2nd largest Tomato producer in Africa and the 14th largest producer of tomato in the world (Olanrewaju et al., 2019). More than 3.9 million tonnes of tomatoes were produced in

Nigeria in 2019, with losses accounting for 45–60% of this total yearly production (Sibomana et al., 2019). Microbial deterioration and physiological activity during transportation and storage are to blame for these post-harvest losses.Historically, food crops were preserved using a variety of techniques, including drying, freezing, curing, and canning (Islam et al., 2018); but were labor-intensive, energy-intensive, and changed the food's flavor and nutritional content.Additionally, a number of health issues have been linked to the use of chemical preservatives in food goods to increase their shelf life (Teshome et al., 2022). As a result, bio-preservatives are increasingly being used in place of chemical preservatives since they preserve the food products' extended shelf life, nutritional value, and hygienic quality.Animal extracts (prawn, shrimp, crab shell) and plant extracts moringa oleifera leaf (Naik et al., 2023), green tea, clove extracts have been used as bio-preservatives to prevent the growth of pathogenic microorganisms, reduce oxidative reactions in the food chain, and increase the shelf life of fruits and vegetables (Islam et al., 2018). An edible covering made of chitosan is non-toxic, biofunctional, biodegradable, and biocompatible. Chitosan's antifungal and antibacterial properties allow it to prevent fruit deterioration (Ruvubu and Roy, 2023). Fruits and vegetables' respiration rate has been successfully lowered using chitosan-based coverings that prevent carbon dioxide and oxgyen from penetrating (Panda et al., 2023). Additionally, applying chitosan to food products like papaya, guava, strawberries, and bananas has shown promising efficacy (Hong et al., 2012, Islam et al., 2018).

Prior research (Islam et al., 2018, Butt et al., 2023) has shown that utilizing a combination of natural preservative and chitosan coating extends the shelf life of a number of perishable goods.Similarly, it has been claimed that moringa oleifera leaves contain a variety of biological effects, including antidiabetic, hypertensive, and thyroid hormone control (Alsamhary, 2023). The antibacterial and antioxidant effects of the leaf extracts are attributed to their high concentrations of phenolics, flavonoids, alkaloids, tannins, and anthraquinone, as well as their known richness in β-carotene (Butt et al., 2023, da Silva et al., 2022). Edible coating technology is one of the promising postharvest methods used to increase the shelf life of food. When fresh fruits are coated with edible material, the quality and deterioration index are less affected. It is intended for the synthesis of chitosan and moringa oléifera extract to create a biofilm that may be used to cover tomato fruit in an edible manner. To the best of our knowledge, no published report has been made on the use of chitosan and a biomix of moringa oleifera leaf extract for coating tomato fruit.Tomato biopreservation storage conditions must be optimized in order to maintain the fruit's qualities and length of shelf life. Managing fresh tomato fruit involves controlling the physicochemical dynamics that take place when it is being stored (Mai and Pathare, 2021). The parameters of importance will be quantitatively analyzed to achieve all of this. One useful method for anticipating and managing these dynamic shifts in fresh

product quality criteria is kinetic modeling (Liao et al., 2024). Historically, kinetic models have been used to characterize changes in the internal (titratable acidity) and external (weight loss) characteristics of fruits and vegetables (Al-Dairi et al., 2023). To the best of our knowledge, not much has been written about the physicochemical kinetics of tomato fruit bio-preservation.

Nigeria has a great capacity for producing tomatoes, and since there are substantial post-harvest losses, it is preferable to use a benign strategy to control shelf-life to minimize waste. This can be accomplished by using the chitosan complex and MLE's unique bio-preservation potential in the biofilm layer of fresh tomato fruit. Precisely, the current study will consider: (1) synthesis and characterization of *MLE*, chitosan and *MLECB* biofilm; (2) investigation of bio-preservation process variables.

2. Method

2.1 Synthesis of *MLECB* biofilm

Using pure ethanol, *Moringa Oleifera* leave extract was prepared by leaching *MLE* from dried *Moringa Oleifera* leaves. Five percent (5g weight of sample/100ml of solvent) dried moringa leaves were added to pure ethanol while being constantly stirred. The solution was left to agitate at 50 ◦C, for 12 h. To obtain MLE, it was then filtered, and oven dried at 70 ◦C onto a consistent weight. A variety of chitosan solution concentrations, including 0.5%, 1%, 1.5%, 2.0%, and 2.5% (weight of sample/100 ml), were created by dissolving measured volumes of chitosan powder in 1% acetic acid solution while stirring continuously for five hours at 60 ◦C. Using glycerol plasticizer (25 % w/w), the pure (chitosan 100 % or *MLE* 100 %) and blended (*MLE*: chitosan = 20%: chitosan 80% ; 40%:chitosan 60% ; 60%:chitosan 40% ; 80%:chitosan 20%) substrates of MLE and chitosan were combined to create *MLECB*. The mixture was permitted to stir at 40 ◦C and 500 rpm for 2 h using Axiom hotplate magnetic mixer (Model 85-2). After this procedure the sample was permitted to chill to room temperature. Fresh tomatoes were bio-coated with the chilled samples right away, and the rest was labeled *MLECB* and refrigerated until needed again.

2.2 Coating and bio-preservation studies

Half of the tomato samples were divided into two equal groups, one of which received *MLECB* coating and the other was left uncoated, acting as a negative control.Five subgroups were then chosen to represent the process variable levels in each of the two batches. Three tomato fruit samples comprised each level, and the mean value of the three tomato fruit samples was the result at the end of the experiment for each level of the process variable. Each tomato fruit sample was coated by submerging it in the *MLECB* biofilm for 60 seconds, and then letting it dry for two minutes. Every sample was put into a custom-made bio-respirator, as shown in Figs. S1 and S2 (supplementary material). At various sampling days, physicochemical

characteristics such as titratable acidity and weight loss rate were determined and documented.Equations (1) through (3) provide the governing equations for predicting the weight loss, weight loss rate, and TA, respectively.

$$\text{Weight loss } (\%) = \left(\frac{W_1 - W_2}{W_1}\right) \times 100 \quad (1)$$

$$\text{Weight loss rate } \left(\frac{g}{day}\right) = \left(\frac{W_1 - W_2}{t}\right) \quad (2)$$

Where W_1 and W_2 represent the initial and final weight (g) of tomato fruit at sampling time, while t denotes the sampling time (day).

$$\text{TA } \left(\frac{\text{g citric acid}}{\text{kg of tomato}}\right) = \left(\frac{V \times 0.1 \times 1000 \times 0.064}{m}\right) \quad (3)$$

Where 0.1 is the normality of NaOH (N), 0.064 is the conversion factor from sucrose acid to citric acid, V is the volume of NaOH required for the titration (ml), while m is the mass of tomato juice (g), (AOAC, 2000).

2.3 Characterization

In accordance with the AOAC standard, the physicochemical characterization was performed to ascertain the characteristics of *MLE*, chitosan, and *MLECB* biofilm (Ohale et al., 2022, Feldsine et al., 2002). Section S1 of the supplemental material contained the comprehensive methods for determining the phenolic compounds, flavonoids, alkaloids, tannins, styrene, and benzoic acid. Using SHIMADZU GC-MS equipment (model QP2010), the hydrocarbon and phytochemical components of the substrates were measured in accordance with AOAC criteria (Feldsine et al., 2002). Furthermore, a Buck Scientific infrared spectrophotometer (Model 530) was used to determine the chemical functional groups.

3. Results and Discussion

3.1 Characterization

3.1.1 Phytochemical properties

The phytochemical compositions of *MLE* and *MLECB* are illustrated in **Table 1.** The findings indicate that both *MLE* and *MLECB* have moderate levels (++) of tannins and alkaloids together with large amounts (+++) of flavonoids and phenolic chemicals. While the aromatic compounds remained undetectable following *MLECB* production, trace quantities of benzoic acid were found in *MLE* and *MLECB*. The feasibility of *MLECB* to support antioxidant activities during tomato fruit preservation is suitably validated by these results.

Table 1. Phytochemical result of MLE and MLECB

Constituent	Qualitative mount	
	MLE	MLECB
Alkaloids	++	++
Tannins	++	++
Flavonoids	+++	+++
Polyphenols and phenolic acids	+++	+++
Hydroxycinnamic acid and styrene	+	-
Benzoic acid	+	+

3.1.2 FTIR

3.1.2.1 FTIR of *MLE* and chitosan

The Fourier transform infra-red spectra (FTIR) of *Moringa Oleifera* leaf extract (*MLE*) and chitosan film were depicted in Figs. 1 and 2, respectively. The infrared spectrum of MLE produced nine prominent peaks at 3482.97, 2947.17, 2004.61, 1688.03, 1641.42, 1557.34, 1081.72, 1026.22 and 528.86 cm^{-1}. The O-H group is responsible for the broad wavelength at 3482.97 cm^{-1}, which emphasizes the presence of the phenol functional group in *MLE* (Khalid et al., 2023). Given that the MLE substrate is loaded with flavonoids that have an O-H functional group, this is not out of place. The peak at 2004.61 cm^{-1} indicates a strong C–C triple bond alkyne group, whereas 2947.17 cm^{-1} vibration revealed both symmetric and asymmetric stretching of the aliphatic C–H group of alkanes (Naik et al., 2023). These findings support those of previous researchers (de Silva et al., 2022, Khalid et al., 2023, Marrufo et al., 2013) who found a noteworthy presence of alkane chemicals in Moringa oleifera leaf extracts. Furthermore, these compounds' antibacterial properties have been demonstrated (Wei et al., 2023, Carev et al., 2023a). Antioxidants are present in *MLE* when there are infrared peaks between 1700 and 1800 cm^{-1} (Johnson et al., 2020). Cusioli et al. (2023) reported that distinct vibrations at 1688.03 and 1641.42 cm^{-1} confirm the existence of C=O carbonyl groups, which are found in carboxylic acids and ketones, and NH stretching of amide groups, respectively. The signal at 1557.34 cm^{-1} is indicative of the bending vibrations and C-N elongation present in the primary and secondary amides of *MLE* proteins. According to Aisida et al. (2021) alcohols' C–H and C–O elongation vibrations are linked to the sharp bands at 1081.72 and 1026.22 cm^{-1}. This is expected, since ethanol was used in the extraction of MLE substrates. The spectral peak at 528.86 cm^{-1} is

associated with the existence of halo chemicals.Likewise, the chitosan infrared spectra showed the same number of significant peaks ranging from 3324.8 to 659.15 cm-1 (see Fig. 2). The presence of N–H bonds inherent in the amino groups of chitosan molecules is explained by the strongest absorption band at 3324.4 cm-1. The presence of O-H stretching vibration brought on by intramolecular hydrogen bonding is also indicated by this peak (Ruvubu and Roy, 2023). The amine groups of chitosan are linked to the N–H bending vibration found in the absorption band at 1649.8 cm-1 (Tan et al., 2018). The presence of the ϛNH2 vibration of the main amine, a highly reactive functional group of the chitosan biopolymer, is shown by the peak at 1593.11 cm-1 (Bhat et al., 2023). According to Smolarkiewicz-Wyczachowski et al. (2023) the wave bands at 1420.60 and 1350.60 cm-1 correspond to the symmetric deformation of CH2 groups and the vibration of the amide III group, which was caused by the NH group deformation.The presence of infrared absorption bands between 1250 and 800 cm-1 is generally linked to glycosidic rings. Particularly, the pressure of the glucosamine ring's C–O stretching bonds is indicated by distinctive vibrations at 1061.04 and 993.03 cm-1 in the chitosan sample (Hadidi et al., 2020). Furthermore, the O–C–O vibration of acetic acid is visible near the peak of 659.14 cm-1. This observation is related to the fact that the chitosan solution was formed using acetic acid as the dissolution solvent. A common finding from the surface chemistry of chitosan and *MLE* indicates that whereas chitosan primarily exhibits antibacterial activities, the *MLE* substrate has favorable characteristics to support both antioxidant and antimicrobial properties during tomato fruit bio-preservation.

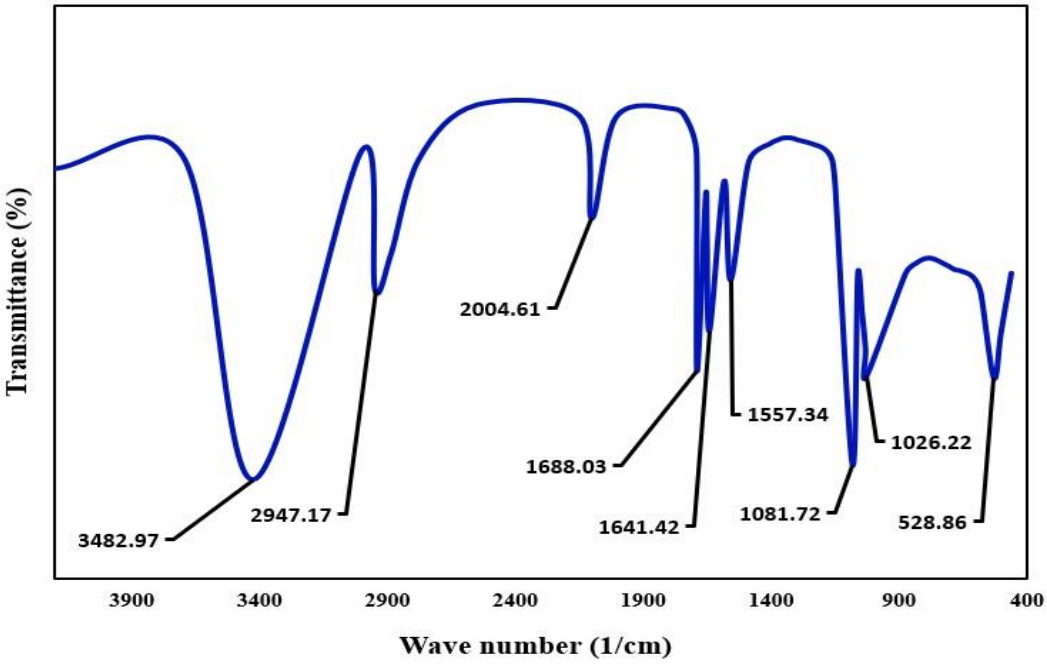


Figure 1. FTIR Spectrum of *Moringa Oleifera* leaf extract (*MLE*)

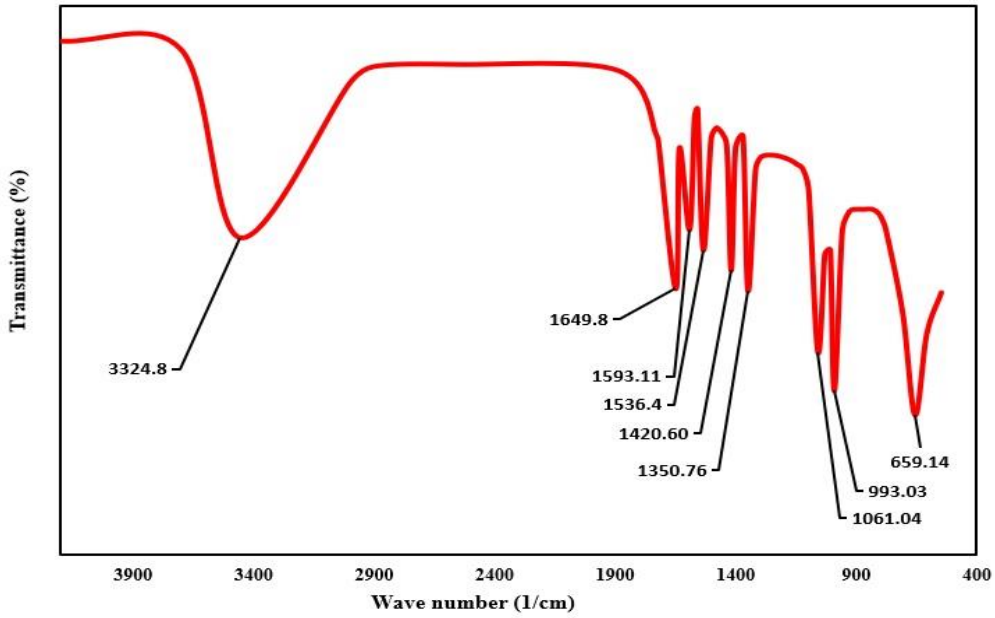


Figure 2. FTIR Spectra of Chitosan

3.1.2.2 FTIR of *MLECB*

The physicochemical properties of the *MLECB* biofilms were influenced by the chemical interactions between chitosan and MLE, which could be investigated using FTIR spectroscopy. Fig 3 showed the infrared spectra of pure substrates (*MLE* and chitosan) and their derivatives with varying content. The *MLE* and chitosan functional groups' interactions may be the cause of the novel peaks that are formed in the *MLECB* species' spectra. A small shift from the OH functional group of 3482.97 cm^{-1} found in *MLE* is highlighted by the vibrational peak at 3454.61 cm^{-1}. However, the NH amino group of chitosan (3324.8 cm^{-1}) and the OH functional group of *MLE* may have interacted to generate this band. This reactive interaction may indicate that the flavonoids and chitosan in the *MLE* substrate are cross-linking. Furthermore, the absorption peak at 2948.11 cm^{-1} identified the alkane group, but the alkyne group changed from 2004.61 cm^{-1} in MLE to 1980.83 cm^{-1} in the *MLECB* sample. The development of the C=N stretching mode of imines is highlighted by the new, sharp absorption peak at 1661.8 cm^{-1}. This could be the consequence of amine groups (N–H, $–NH_2$) of chitosan interacting with carbonyl groups (C=O, ketones, carboxylic acid) of *MLE* in a Schiff-base reaction (Božič et al., 2012, Tan et al., 2018). Amides II and III were detected at 1560.25 and 1357 cm^{-1}, respectively, in the *MLECB* biofilm. Notably, in *MLE* and chitosan, these amide peaks moved from their initial bands, indicating effective interactions in *MLECB* synthesis. The presence of glucosamine and alcohol traces in the *MLECB* biofilm is shown by the wave bands at 1071.7 cm^{-1} and 936.3 cm^{-1}, which are marginally different from 1081.72 cm^{-1} (the *MLE* substrate) and 993.3 cm^{-1} (the chitosan substrate). It is significant to notice that in every mixed species containing chitosan sample, the

presence of acetic acid at 659.84 cm-1 remained unchanged. This suggests that MLE and the acetic acid residues in the chitosan solution did not interact.

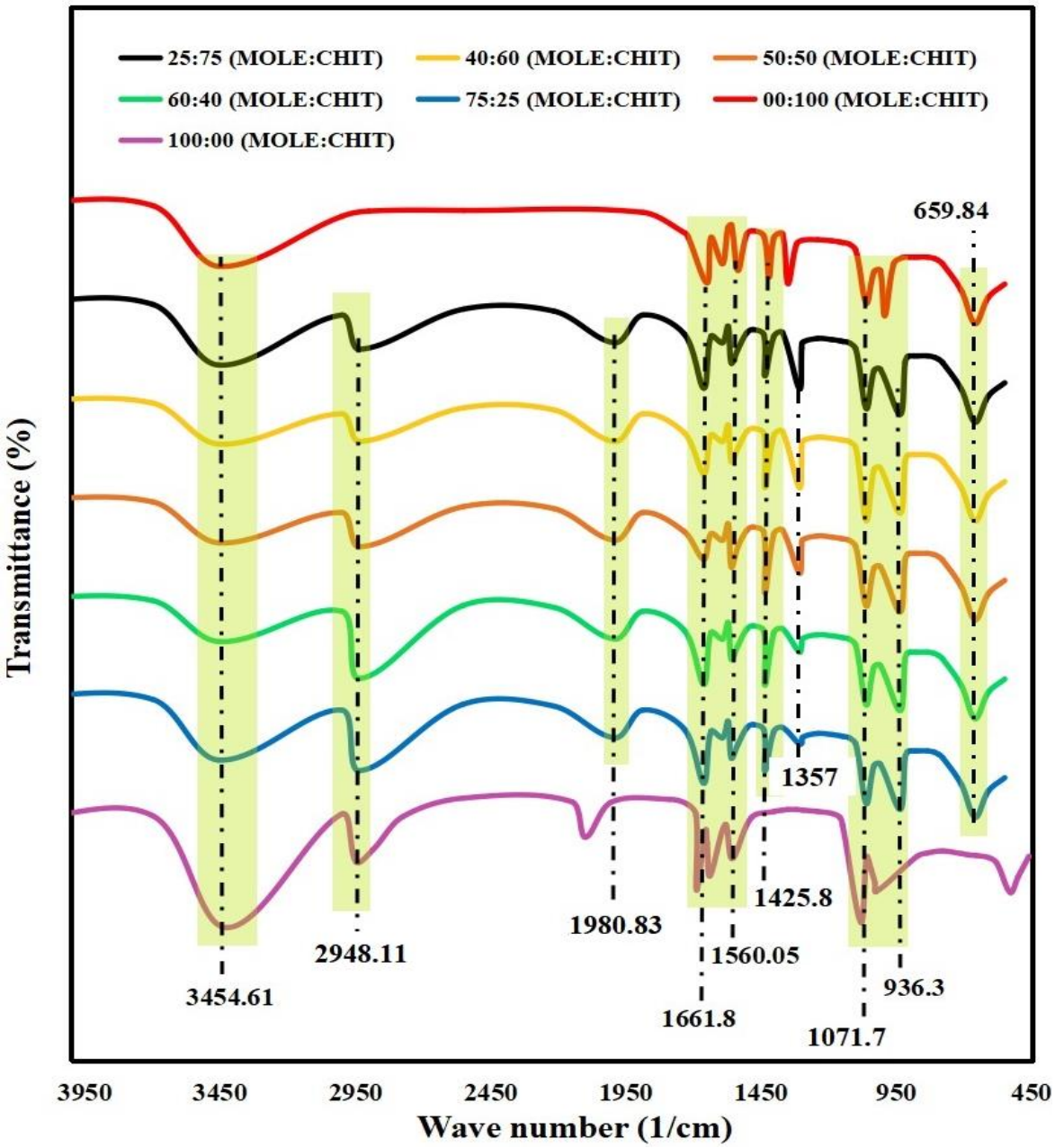


Figure 3. FTIR Spectra for *MLECB* at different mixing ratios

3.1.3 Gas chromatography–mass spectrometry

The hydrocarbon and phytochemical constituents of *MLE* and *MLECB* were given in Figs. 4 - 5, respectively. Table S1 (Supplementary material) presents the hydrocarbons and phytocomponents of *MLE* and their associated retention time (RT), molecular weight (MW), peak area, and concentrations (%).

According to the hydrocarbon study, the *MLE* substrate included four main hydrocarbon compounds at quantities higher than 10%. These hydrocarbons include Tetradecane (41.24 %), Pentadecane (18.46 %), Tridecane (12.386 %) and Heptacosane (12.013 %). It has been determined that tetradecane is a potent anti-microbial agent with numerous uses in food packing and preservation (Karim et al., 2022, Bains et al., 2023). Also, the antimicrobial and anti-oxidant activities of pentadecane (Faridha Begum et al., 2016, Girija et al., 2014) and tridecane (More et al., 2022) have been recognized. These results support the findings of Marrufo et al. (2013), who noted that one of the main ingredients in Moringa Oleifera leaf extract was heptacosane.It is noteworthy to mention that the liquid extracts of plant leaves include heptacosane, a very potent anti-microbial component (Carev et al., 2023b). These findings support the findings of the FTIR study, which indicated that the leaf extract from Moringa oleifera contained alkanes (MLE). In Fig. 4, the phyto-components of MLE were discovered. The findings indicate that *MLE* has notable levels of the flavonoids quercetin (>25%), kaempferol (>20%), and artemetin (>10%).Other substances in the 5–10% concentration range are ferrulic acid acid, robinolic, and daizin. Numerous studies have emphasized the antioxidant qualities of kaempferol (Tian et al., 2021) and quercetin (Marrufo et al., 2013). Furthermore, it has been noted that quercetin possesses natural cross-linking properties, which is a highly desired property when creating biochemical mixes with other substrates and plant extracts (Wiggers et al., 2022, Hong et al., 2022). The findings of Kashyap et al. (2022) and Lin et al. (2018), who revealed the phytochemical contents of Moringa Oleifera leaf extracts, are equivalent to the concentration hierarchy of these flavonoids. These findings highlight *MLE*'s capacity to protect fruits and vegetables coated with *MLE* biofilm from oxidation and microbiological attack.

A decrease in the quantities of certain main constituents found in *MLE* was seen in the phyto-components of *MLECB* biofilm. While kaempferol was not found, the concentration of quercetin dropped dramatically from 26.5035% in *MLE* to 10.2656% in *MLECB*. This decrease in concentration is explained by quercetin's active involvement in the cross-linking process with the chitosan biopolymer. It is implied that kaempferol was used in the creation of a new *MLECB* bioconjugate by its disappearance from the *MLECB* biofilm.A broad comparison of the phytochemical components of *MLE* and *MLECB* indicates that phenolic and flavonoid chemicals played a major role in the biofilm production of *MLECB*.

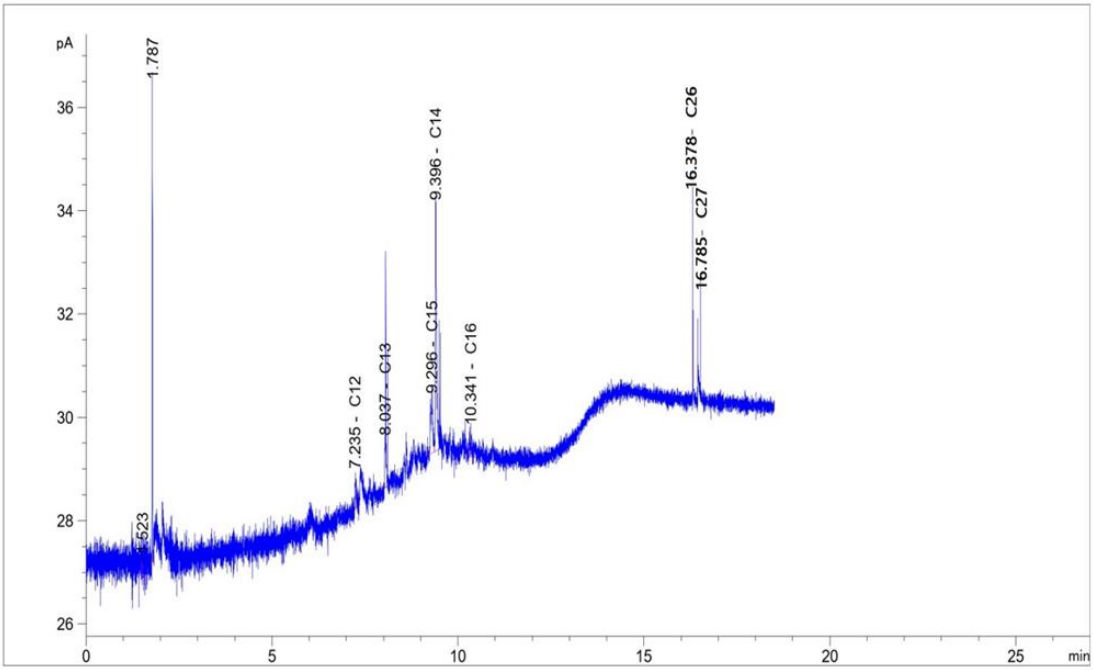
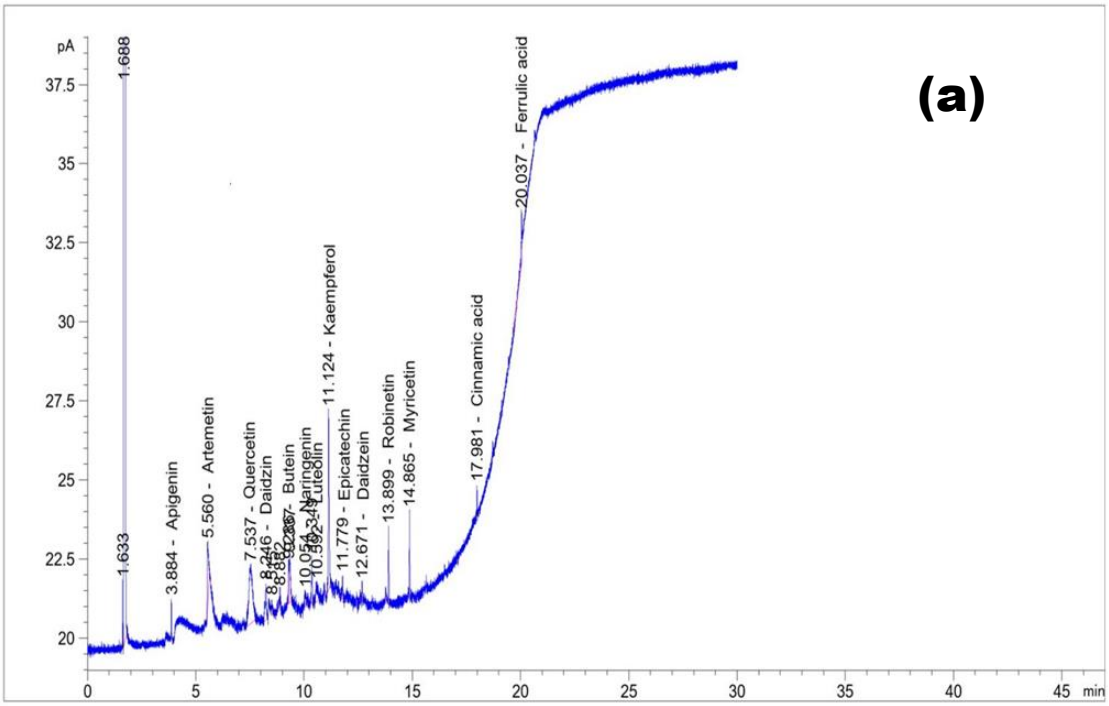


Figure 4. GC-MS result of hydrocarbon constituents of *MLE*



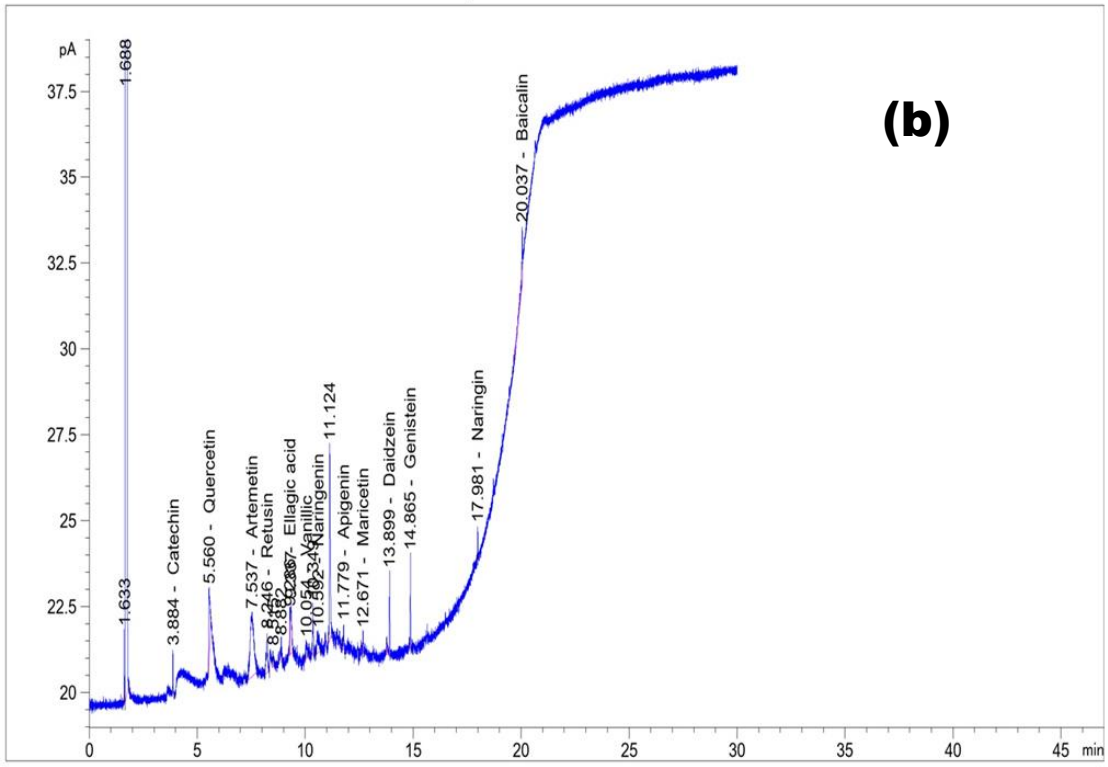


Figure 5. Phytochemical constituents of (a) *MLE* and (b) *MLECB*

3.2 Effect of bio-preservation variables

3.2.1 Effect of chitosan concentration

Five different chitosan concentrations (0.5%, 1.0%, 1.5%, 2.0%, and 2.5%) were tested for their impact on the titratable acidity (TA) of fresh tomato fruits. At days 5, 10, 15, and 20, samples of the results were taken using a 50:50 *MLECB* mixing ratio (MLE: chitosan, v/v). According to Fig. 6's results, at chitosan concentrations of 0.5% and 1.0%, respectively, the value of TA increased from 3.607 g/kg to 3.738 g/kg on the fifth day of incubation. The measured TA value continued to differ very little at 1.0 % conc. until it peaked at 2.5 % chitosan concentration. The complementary sealing action of chitosan in *MLECB* biofilm for coating tomato fruit is responsible for the increase in TA value at higher chitosan concentration. According to Fig. 6, this effect was considerably improved when the chitosan concentration rose from 0.5% to 1.0%, but it only slightly improved outside of this range. Accordingly, it was determined that the optimal concentration of chitosan for later research was 1.0% conc. Other researchers (Chien et al., 2007, Dong et al., 2004) that looked into the impact of chitosan concentration on fruit and vegetable shelf extension obtained similar results.

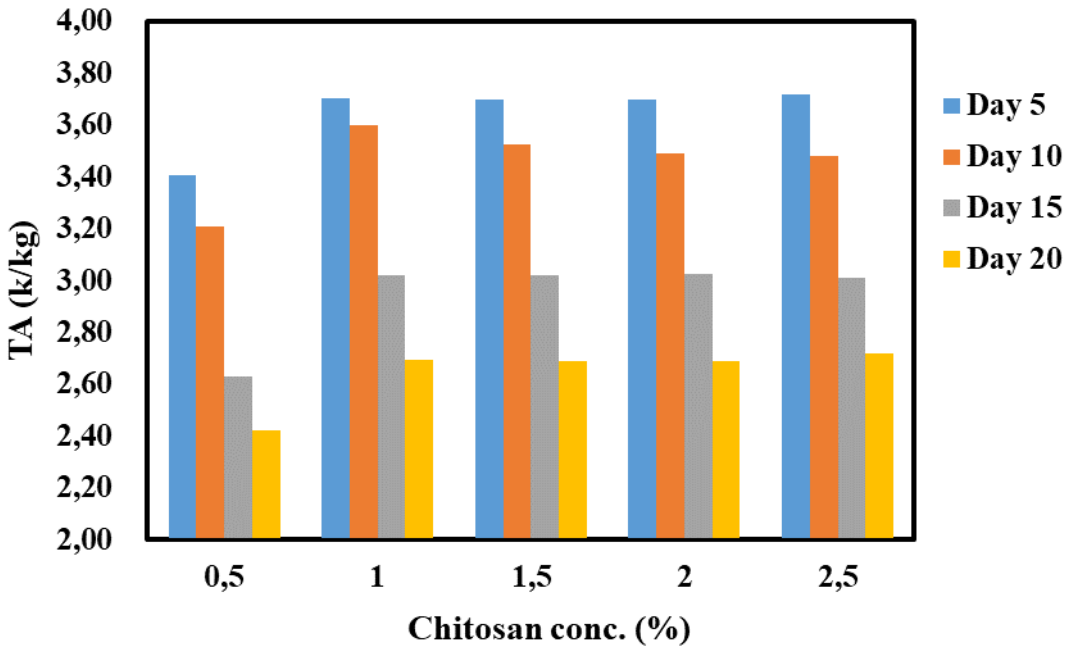


Figure 6. Effect of chitosan concentration on TA

3.2.1 Effect of *MLECB* mixing ratio

The effect of *MLECB* biofilm ratio on weight loss rate and TA of coated tomato species at different sampling time were portrayed in Figs. 7 (a) and (b). Fig. 7 (a) established that the weight loss rate in g/day stayed approximately the same as the *MLE* composition upsurge from 0 to 40 %. The rate of weight loss increased above 40% MLE composition and reached a final *MLECB* mixing ratio of 100:0 (MLE:Chitosan, v/v).For the titratable acidity measurement, a similar finding was made, with the maximum value being found at a *MLECB* ratio of 40:60 (MLE:Chitosan, v/v). For fruits and vegetables to have the best possible shelf life preservation, Chen et al. (Chen et al., 2019) state that a slower rate of weight loss is preferable. A slower rate of weight loss indicates that the organic acid content of tomato fruit is being gradually lost. The rise in weight loss rate (Fig. 7 (a)) and decrease in titratable acidity (Fig. 7 (b)) were occasioned by the decreasing composition of chitosan in the mixing ratio of *MLECB* biofilm. The modest rise in titratable acidity that was induced by an increase in the *MLECB* mixing ratio may have resulted from the complementary *MLE* enhancement in chitosan's bioactivity.

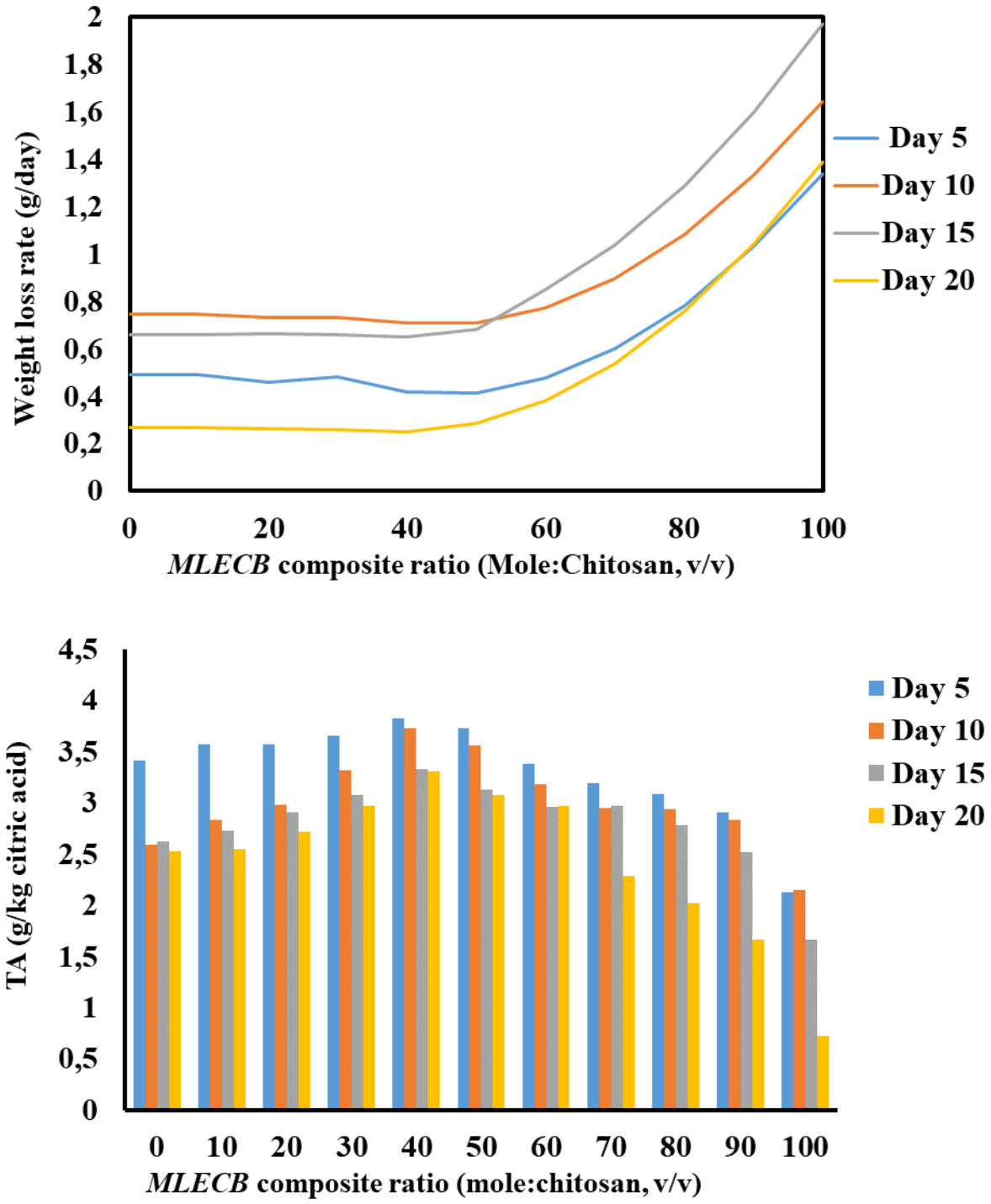


Figure 7. Effect of mixing ratio on (a) TA, (b) weight loss rate

3.2.3 Effect of storage time

Figs. S3(a) and S3(b) show the effect of incubation period on the titratable acidity of coated and uncoated tomato fruits at the optimal chitosan concentration and MLECB mixing ratio, respectively. The weight loss rate measurement of both coated

and uncoated species were portrayed in Fig. S4 (a) (for best mixing ratio of 40:60), and S4 (b) (for best chitosan conc of 1%). The graphical results show that the values of TA decreased proportionately with an increase in storage duration for both coated and uncoated species. The dynamic reduction of citric acid that occurs throughout the storage respiration process is responsible for the decrease in titratable acidity. Gol et al. (Gol et al., 2013) state that because there is a strong correlation between titratable acidity and organic acid content, a drop in acidity may be expected throughout incubation as a result of respiration-induced metabolic processes that consume the organic acids.The titratable acidity of the uncoated samples was much lower than that of the *MLECB*-coated samples for all species. This demonstrates how well *MLECB* biofilm coats tomato fruits and regulates the breakdown of citric acid during fruit respiration.Additionally, the quality of the coated tomato fruit remained good during the incubation period, however on the fifteenth day of sampling, the quality of the uncoated sample dropped below the 0.30 g/kg threshold (TA = 0.30 g/kg) (Kayode and Afolayan, 2014). It's also crucial to remember that the tomato fruit loses moisture during respiration, which results in the cumulative weight loss (%) seen in Fig. S5. The coated tomato fruit's weight loss rate (Figs. S4(a) and S4(a)) showed a quadratic relationship with storage time. After 2 to 9 days of storage, the first area of an accelerated weight loss rate was seen (Fig. S4 (a)).The rate of weight loss increased from 0.068 g/day to 0.6762 g/day over this time. The initial properties of the *MLECB*-coated tomato fruit surface (soft and permeable) may have contributed to the fast rate of weight loss because they largely did not hinder the permeability of water across the membrane.The *MLECB* biofilm toughened over the course of the storage period, reaching the equilibrium stage (Days 10–14: 0.7103–0.7146 g/day) and slowing down the rate of weight loss (Days 15–20: 0.3246 g/day).At the optimal chitosan concentration, a somewhat prolonged time of enhanced weight loss rate was seen (Fig. S4 (b)). During the storage period, the weight loss rate of the uncoated tomato fruit was approximately 2.6 times higher than that of the *MLECB* coated species due to its exposed surface. This level of protection emphasizes even more how crucial *MLECB* coating is for improving the membrane's ability to withstand moisture loss.The percentage weight loss is another significant graphical depiction of the impact of storage time (refer to Figs. S5 (a) and (b)).The percentage weight loss illustrates the gradual loss of moisture that impacts the overall weight during storage, whereas the weight loss rate precisely reflects how phase variations in *MLECB* coating regulate the weight loss throughout storage.The natural process of fruit and vegetable incubation following harvest is the percentage weight loss. All species showed progressive weight loss over the incubation period, as shown in Figs. S5 (a) and (b). The uncoated species ruptured and the covered species shriveled as a direct result of moisture loss. This led to a steady loss of weight.Following a 20-day incubation period, the coated samples

(with the optimal MLECB mixing ratio) lost 12.87 weight percent, while the uncoated samples lost 32.88 percent of their starting weight.Researchers Mahfoudhi et al. (2014), Tarangini et al. (2022), and Ruelas-Chacon et al. (2017) found that the weight loss for uncoated tomato species in this investigation was marginally greater than the value reported by other researchers who did tomato incubation at ambient settings.Following a 20-day incubation period, Mahfoudhi et al. (2012) observed a 23.65% weight loss of the original weight, whereas Tarangini et al. (2022) obtained a 27.5% weight loss. The discrepancy between these outcomes and our discoveries may be ascribed to variances in physicochemical settings, like shifts in humidity and temperature.It is noteworthy, although, that MLECB biofilm outperformed sericin complex (Tarangini and al., 2022) and almond gum (Mahfoudhi et al., 2014) in controlling tomato fruit weight loss. The formation of white rot on the fruit's surface, was observed on the 10th, 15th, and 20th days of sampling, is another obvious result of unprotected exposure.Based on the obtained graphic result, the rot size increased more intensely. The diameter of the first detection on day ten was 6.5 mm; on days fifteen and twenty, it grew to 13.6 mm and 18.53 mm, respectively.It's interesting to notice that during the storage period, this rot was not found in the coated species.

4. Conclusion

In this work, *MLE* and chitosan were dynamically homogenized to successfully generate *MLECB* biofilm. After the biofilm was characterized, it was discovered to include important antibacterial and antioxidant components that could be used to spread fruit and vegetable biopreservation. The impact of the bio-preservation variables showed that the weight loss and titratable acidity of the coated tomato fruits were considerably decreased with an increase in chitosan content. Additionally, the impact of the *MLECB* mixing ratio showed that, given the related improved quality characteristics at this value, the 40:60 *MLECB* ratio was the ideal ratio. Overall, the study's findings show that applying *MLECB* biofilm to tomato fruit can greatly control how quickly its quality deteriorates. Additionally, even after 20 days of preservation, the customers can be assured of receiving harmless and eatable tomatoes by applying *MLECB* coating on fresh tomato fruit.

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