Shelf life and quality of tomato (*Lycopersicon esculentum* Mill.) fruits as affected by neem leaf extract dipping and beeswax coating

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**ABSTRACT**

The effect of Beeswax (BW) coating and Neem leaf extract (NLE) dipping on the shelf life of tomatoes (*Lycopersicon esculentum* Mill.) was investigated over a storage period of 36 days. The experiment was done by a factorial combination of four levels of NLE dipping (0, 15%, 20% and 25%) and BW coating (0, 3%, 6% and 9%) in randomized complete block design with three replications. It was observed that BW9*NLE25 significantly (P < .001) extended the shelf life of tomato fruits by eight days and effectively reduced weight loss by 55–68%, the changes in TA by 20–66.7%, PH by 6.3–14.1%, and TSS by 2.8–6.9%. Toward the end of the storage period (on the 28th day), BW9*NLE25 found to have 39.4% higher marketable fruits while reducing the percent decay by 96.1% compared to untreated tomato fruits. The study result showed that BW9*NLE25 could potentially be utilized to extend the shelf life and maintain the quality of tomato fruits by farmers, retailers, and wholesalers.

**KEYWORDS**

 Marketable; Weight loss; Total soluble solids; Titratable acidity; Ascorbic acid; Decay

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**INTRODUCTION**

Tomato (*Lycopersicon esculentum* Mill.) is one of the most popular and widely grown vegetables in the world. According to Central Statistical Agency (CSA),[1] tomato production in Ethiopia was estimated to be 34,947.259 ton from 6,012.28 hectares. Tomato is rich in vitamins; minerals such as phosphorus, sodium, potassium, calcium, and magnesium, and trace elements such as iron, copper, zinc and dietary fibers.[2] The deep-red coloration of the ripened tomato is due to the high amount of lycopene, a form of carotenoid pigment and a notable antioxidant that is beneficial in reducing the incidence of certain chronic diseases such as prostate cancer, cardiovascular disease, and diabetes.[3] Despite the numerous benefits that can be derived from the crop; postharvest losses make its production in many parts of the world less profitable. Tomato fruits are highly perishable, with a short shelf life when stored at ambient temperature. Postharvest losses in tomatoes can be as high as 25–42% globally.[4] In Ethiopia, the post- harvest loss of horticultural crops accounts 25–50%.[5] According this author, the post- harvest losses of tomatoes fruit accounts 45.32% from the total post- harvest loss of horticultural crops.

In Ethiopia major causes for the deterioration losses of tomatoes are mainly connected to harvesting techniques, transport, and storage facilities.[5,6] The susceptibility of fresh harvested tomato fruit to postharvest deterioration increases during extended storage as a result of physiological, chemical and pathological changes occurring during storage and marketing. Therefore, its preservation and storage are important to the economy of individual homes, farmers and the country considering the vital role it plays in the health of people.[7]
In Sub-Saharan African countries, cold storage facilities often cannot be afforded by small-scale growers and retailers, which may be due to a lack of capital or lack of technical knowledge. Plant extracts, which are cheap and easy to use, are suggested as alternative options to preserve fruits. Plant extracts acts as a semi-permeable membrane to regulate the diffusion of oxygen and carbon dioxide into and out of the fruit which ultimately reduce the rate of metabolism and also prevent water loss. Plant extracts are eco-friendly and can provide an excellent opportunity to reduce the use of harmful chemicals in postharvest treatment of fruits.

Among the plant extracts, Neem (Azadirachta indica), Lantana (Lantana camara), Chinaberry (Melia azadirach), Ashok (Polyalthia longifolia), Cinnamomum (Cinnamomum zeylanicum) have been used for improving the shelf-life and quality of fruits. Azadirachtin is considered as the most active substance in Neem. Neem also contains many active ingredients such as nimbudin, nimbin, and nimbidol with antibacterial and antifungal activity. The ability of Neem leaf powder to decrease the decay level of tomatoes is an indication that Neem leaf extracts can serve as a possible alternative in the prevention of tomato decay by pathogens. On the other hand, edible surface coatings are used as a film packaging to improve fruits’ external appearance by altering gas permeability, reducing water loss and delaying ripening.

Beeswax coating is derived from honeycomb and is produced by honey extraction. A study by Purwoko and Fitrades showed that beeswax coating can reduce weight loss and maintain total soluble solids of papaya. Beeswax coating and Neem leaf extracts dipping are low cost, non-hazardous, and easily accessible postharvest treatments that can help to reduce postharvest loss of tomato. Therefore, this study investigated the efficacy of beeswax (BW) coating in combination with Neem leaf extract (NLE) dipping on shelf life and quality of tomato fruits.

Materials and methods

Sample preparation

Fully mature green tomato, Shanty PM fresh variety, was harvested from Erer Woreda, Eastern Harerge in Ethiopia. Fruits were selected based on uniformity in size, maturity (mature green) and absence of external injuries. The fruits were cleaned properly with distilled water to remove dust, dirt, mud, filth, etc.

Preparation and concentration of Neem leaf extract

Neem (Azadirachta indica) leaves were collected from Dire Dewa (Tonny Farm). Fresh Neem leaves were collected from middle part the tree and then air dried. After drying the plant material was ground with mechanical grinder to prepare a fine powder. Then, 200 g of the powder was soaked in 1000 mL of distilled water for 6 h and then passed through muslin cloth. The filtrate was used as a stock solution (100%) for preparing NLE coating solution of 15%, 20% and 25% by further dilution as described by Gupta & Jain.

Preparation and concentration beeswax emulsion

Beeswax emulsion was prepared following the methods described by Mukdisari et al. Beeswax emulsion concentration of 3%, 6%, and 9% was prepared by heating 30 g, 60 g, and 90 g of beeswax in a 2 L container and melted at 70°C, the contents were heated continuously to attain a temperature of 80 °C to 90 °C. Oleic acid (160 mL) was added to the melted wax with a constant stirring. Then, 840 mL of distilled water (which was pre-heated at the same temperature of 80 °C to 90 °C was added to the mixture slowly with continued stirring for five minutes and air dried.
**Experimental design and treatment of tomato fruits**

A factorial combination of four levels of Neem plant extract (control, 15%, 20% and 25%) and four levels of beeswax coating (control, 3%, 6% and 9%) storage treatments with three replications were applied on fully matured green tomatoes in the study. The treatments were arranged in a randomized complete block design.

A total quantity of 432 kg tomato fruits was used for the study, where each treatment combination had 27 kg fruits which were subdivided into three lots (9 kg of tomato fruits for each replication). For each treatment combination, tomato fruits were dipped in the Neem leaf extract solution for five minutes and surface air dried, then after the fruits were treated with the beeswax coating material. Untreated tomato fruits were used as a control. The fruits were then stored at ambient storage condition for physicochemical analysis and shelf life determination at four days interval during 36 days of storage period. The average storage room air temperature and relative humidity varied from 15.2°C to 20.4°C and 55.53% to 69.46% RH during 36 days of storage period at Haramaya University from February to April 2020.

On each sampling date, a sample of 1 kg tomato was randomly taken from each replication in the treatment for quality analysis. Data were recorded on 4, 8, 12, 16, 20, 24, 28, 32, and 36 days after storage. Data on physiological loss in weight, chemical compositions (total soluble solids, pH, titratable acidity, and ascorbic acid), decay (%), percentage marketability, and shelf life were assessed at an interval of four days during 36 days of storage period under ambient conditions.

**Analytical methods**

Physiological loss in weight: The physiological weight loss was calculated for each interval and converted into a percentage of initial weight. The cumulative weight loss was expressed in percentage for different treatments as described by Gharezi. [28]

**Total soluble solids**

The total soluble solids (TSS) of tomato juice extracted were measured using the method described by Mazumdar with a hand refractometer. The prism of refractometer was washed with distilled water and blotted out with tissue paper before use. This was done for each test interval between samples. The device was standardized against distilled water in which TSS reading is 0 °Brix. The total soluble solids were determined by placing two to three drops of tomato juice on the prism of the hand refractometer that have capacity to read in the range between 0–32 °Brix.

Titratable acidity: The titratable acidity (TA) of tomato fruits was determined in terms of percent anhydrous citric acid as described by Mazumdar. [29] An aliquot of tomato juice was extracted from three tomatoes with a Kenwood juice extractor (6001x model No. 31JE35 6x.00777). The fruit slurry was filtered through cheesecloth and 100 mL was centrifuged for 15 min. The decanted clear juice was used for the analysis. The TA expressed as percentage citric acid, was obtained by titrating 10 mL of tomato juice with 0.01 N NaOH to pH 8.2.

**pH value**

The pH of tomato fruit juice was determined using a digital pH meter (Model: Jenway 3310) according to AOAC. [30] The pH meter (Hannan) was calibrated by using buffer solutions of pH 7 and pH 4 when correction for temperature was also taken into consideration. After calibration, the electrode was washed twice with distilled water and dipped into the tomato juice and the pH was recorded.

Ascorbic acid: The ascorbic acid (AA) content of tomatoes was determined by the 2, 6-dichlorophenol indophenol method. [30] An aliquot of 10 mL tomato juice extract was diluted to 50 mL with 3% met phosphoric acid in a 50 mL volumetric flask. An aliquot was centrifuged at 10000xg for 15 min and titrated with the standard dye to a pink end-point (persisting for 15 sec). The AA content (mg/100 g) was calculated from the titration value, dye factor, dilution and volume of the sample.
Marketability (%): Marketable quality was evaluated according to the scoring method used by Mohammed et al.\[31\] based on a 1–9 rating scale with 1–2.49 = unusable; 2.5–4.49 = unsalable; 4.5–6.49 = good; 6.5–8.49 = very good; and 8.5–9 = excellent. Descriptive quality attributes of tomatoes were determined subjectively by observing the level of visible mold growth, decay, shriveling, smoothness, and shininess of fruit. The number of fruits receiving a rating of five and above was used to calculate the percentage marketable fruit during storage.

Decay (%): The percentage decay of the stored tomato fruits was determined visually as described by El-Anany et al.\[32\] Percentage decay was calculated as the ratio between the number of fruits infected by microbes or showing rotting and number of sample fruits initially stored.

Shelf life: The shelf life was calculated by counting the days required to attain the last stage of ripening, but up to the stage when fruit remained still acceptable for consumption or marketing as described by Liamngee et al.\[31\] If 40% of tomato fruits showed symptoms of spoilage, the fruits sample was considered to have reached end of the shelf life.\[33\]

Data analyses
The significance of the effects of Neem leaf extract and beeswax coating on measured parameters was assessed by analysis of variance (ANOVA) and the multiple comparisons of the treatments means was made by the Fisher’s least significant difference (LSD), with the goal of pinpointing statistically significant differences (at the 5% level) using XLSTAT 2015 (AddinSoft™ SARL, Paris, FRANCE). Correlation and Principal Component Analysis (PCA) were conducted on the observations or variables table to show a visual interpretation of the interaction effect of Neem leaf extract dipping and beeswax coating on shelf life and quality of tomato fruits.

Results and discussion
Physiological loss in weight
In this study significant (P < .05) difference in physiological loss in weight (PLW) of tomato fruits was observed due to the interaction effect of Neem leaf extract (NLE) dipping and beeswax (BW) coatings during 36 days of storage period under ambient conditions (Table 1). The highest rates of PLW was observed in control fruit samples (1.43% to 17.12%) while the lowest PLW was recorded in 9% BW*25% NLE (0.7% to 6.27%) with increasing in the storage period from day 8 to day 28. Notably PWL of tomato fruits decreased with increase in the concentration of BW coating from 0 to 9% and NLE dipping from 0 to 25%. Tomato fruits treated with the combinations of 9% BW and 25% NLE resulted in the lowest percentage of PWL throughout the storage period, followed by those tomatoes treated with 9% BW coating and 20% NLE coating. Tomato fruits with 0% BW coating combined with 0%, 3% and 6% NLE dipping resulted in a relatively higher percentage of PWL. Tomato fruits not treated with beeswax coating were rotten and discarded after the 32nd day of storage.

A general trend of an increase in PLW of tomato fruits (irrespective of BW or NLE treatments) with the advancement of storage period was noted, which is in agreement with the findings of Getinet et al.; Tefera et al.; Tigist et al.\[34-36\] The variation in the rate of PWL of tomato fruits could be due to the difference in physiological processes and response to the atmospheric conditions among BW or NLE treated or untreated tomato fruits. Pérez et al.\[37\] reported that weight loss of most fruits and vegetables during storage depends on the temperature and relative humidity conditions. Mohammed et al.\[31\] indicated that transpiration is a major cause of deterioration because it results in direct quantitative loss (loss of weight) which is the principal cause of fruit softening and shriveling. Respiration on the other hand utilizes the plant product as its substrate thereby leading to weight loss and shriveling. Ball & Marini and Kissinger et al.\[38,39\] reported that the effect of weight loss in sweet pepper and other vegetables caused damage to fruit appearance and subsequent loss of market value and consumer acceptability. Additionally, the loss of water through transpiration and respiration
### Table 1. The interaction effect of Neem leaf extract dipping (NLE) and beeswax coating (BW) on the mean weight loss (% ±SD) of tomato fruits during 36 days of storage period.

| Treatments | Weight loss (%) | Storage period (days) |
|------------|-----------------|-----------------------|
| Neem leaf extract dipping (%) | Beeswax coating (%) | 4 | 8 | 12 | 16 | 20 | 24 | 28 | 32 |
| 0 | 0 | 1.430 a (0.01) | 2.720 a (0.01) | 3.600 a (0.01) | 5.060 ±g (1.74) | 10.700 j (0.01) | 17.120 m (0.01) | - | - |
| 0 | 3 | 1.230 k (0.01) | 1.630 k (0.01) | 2.563 f (0.06) | 4.530 n (0.01) | 6.610 j (0.01) | 9.460 j (0.01) | 12.610 k (0.01) | 17.117 h (0.02) |
| 0 | 6 | 1.030 n (0.01) | 1.393 h (0.01) | 2.180 g (0.01) | 4.070 de (0.01) | 6.140 g (0.01) | 9.010 g (0.01) | 12.150 h (0.01) | 16.680 h (0.01) |
| 0 | 9 | 0.940 e (0.01) | 1.220 f (0.01) | 1.680 d (0.01) | 3.570 cd (0.01) | 5.360 f (0.01) | 8.010 f (0.01) | 11.140 d (0.01) | 15.660 d (0.01) |
| 15 | 0 | 1.350 h (0.01) | 2.500 h (0.01) | 3.250 m (0.01) | 5.290 h (0.01) | 6.790 k (0.17) | 10.490 i (0.01) | 16.960 m (0.01) | - |
| 15 | 3 | 1.140 d (0.01) | 1.530 i (0.01) | 2.320 h (0.01) | 4.210 def (0.01) | 6.280 g (0.01) | 9.120 i (0.01) | 12.260 j (0.01) | 16.780 j (0.01) |
| 15 | 6 | 0.940 f (0.01) | 1.280 g (0.01) | 2.050 f (0.01) | 3.620 cd (0.01) | 5.410 h (0.01) | 8.060 f (0.01) | 11.160 g (0.01) | 15.670 d (0.01) |
| 15 | 9 | 0.840 h (0.01) | 1.110 d (0.01) | 1.470 c (0.01) | 2.760 ab (0.01) | 4.500 b (0.01) | 7.150 c (0.01) | 10.270 d (0.01) | 14.790 b (0.01) |
| 20 | 0 | 1.300 h (0.01) | 2.310 m (0.01) | 3.170 c (0.01) | 4.850 n (0.01) | 6.580 k (0.01) | 10.070 i (0.01) | 16.870 i (0.01) | - |
| 20 | 3 | 1.090 a (0.01) | 1.480 a (0.01) | 2.470 f (0.01) | 4.360 m (0.01) | 6.430 l (0.01) | 9.270 i (0.01) | 12.400 j (0.01) | 16.920 j (0.01) |
| 20 | 6 | 0.870 d (0.01) | 1.150 e (0.01) | 1.850 g (0.01) | 3.340 b (0.01) | 5.060 c (0.01) | 7.560 e (0.01) | 10.680 g (0.01) | 15.220 f (0.01) |
| 20 | 9 | 0.800 h (0.01) | 0.950 b (0.01) | 1.250 a (0.01) | 2.500 g (0.01) | 4.220 b (0.01) | 6.870 b (0.01) | 9.980 c (0.01) | 14.510 c (0.01) |
| 25 | 0 | 1.250 f (0.01) | 2.110 i (0.01) | 3.010 k (0.01) | 4.690 n (0.01) | 6.110 d (0.01) | 9.110 h (0.01) | 16.810 i (0.01) | - |
| 25 | 3 | 1.010 g (0.01) | 1.120 d (0.01) | 2.140 g (0.01) | 3.510 cd (0.01) | 4.800 a (0.01) | 7.400 d (0.01) | 10.520 e (0.01) | 15.050 e (0.01) |
| 25 | 6 | 0.810 h (0.01) | 1.050 c (0.01) | 1.490 c (0.15) | 2.600 a (0.01) | 4.583 c (0.22) | 7.163 c (0.17) | 10.273 d (0.17) | 14.783 b (0.17) |
| 25 | 9 | 0.700 e (0.01) | 0.900 a (0.01) | 1.130 a (0.01) | 2.360 a (0.01) | 3.770 a (0.01) | 6.270 a (0.01) | 9.370 b (0.01) | 13.890 b (0.01) |

P value: 0.000 0.0001 0.0001 0.0001 0.0001 0.0001 0.0001

a-o Means within a column not sharing a superscript letter are significantly different (p < .05). SD= standard deviation
could lead to the buildup of free water on the surfaces of the fruit. This provides a good growth condition for pathogens and could lead to deterioration of the fruit. Ahmed[40] reported that tissue maceration due to pathogen leads to progressive deterioration.

About 10% physiological loss in weight is considered as an index of termination of shelf life (threshold level) of commodities. Acedo[42] also stated that 10% weight loss makes most fruits wilt or shrivel and lose the appearance of being “fresh.” Accordingly, the mean value of physiological weight loss of NLE0*BW0, NLE20*BW0, and NLE15*BW0 treated tomato fruits reached the threshold level between 20 and 24 days. Whereas tomato fruits treated with BW coating (irrespective of the concentrations of NLE dipping) reached the threshold levels on the 28th day of the storage period. Notably, tomato fruits treated with both BW coating and NLE dipping reached the threshold levels on the 32nd day of the storage period.

The rate of weight losses in fruit could be linked to the thickness of the peel of the fruits. Wills et al.[43] indicated that fruit transpiration rate in which water moved out and resulted in wilted rind and a shriveled appearance is directly related the thickness of the pericarp or the peel. In this regard modification of the micro atmospheric condition surrounding the fruit, e.g., edible coating, could minimize the rate of transpiration and respiration of the fruits. In this study, PWL of tomato fruits decreased with increase in the concentration of BW coating from 0 to 9% and NLE dipping from 0 to 25%. The combined application of BW coating and NLE dipping might have been an effective treatment to reduce weight loss by covering the stomata and lenticels of the cell wall of the fruits which reduces the rate of transpiration and respiration. Gupta & Jain[26] reported that slow rate of physiological loss in weight in mango fruits treated with 20% NLE dipping which could be due to its ability to retard moisture loss from the fruits. Rathore et al. and Siddiqua et al.[44,45] also indicated that 20% Neem extract effectively reduced the rate of weight loss in bananas which supports the findings of the current study.

Interestingly, the combined application of 9% BW coating and 25% NLE dipping of tomato fruits reduced the rate of PLW throughout the storage period. This could be related to the modification of the atmospheric conditions surrounding the fruits which probably lowered the rate of respiration and transpiration. Banks et al. and Dhall[23,24] reported that BW coating of fruits could create a modified atmospheric condition by altering gas permeability, reducing water loss and delaying ripening. The result of this study is in agreement with the findings of Mukdisari et al. The authors observed a reduced weight loss in papaya fruit treated with combination of 0.75% Chitosan and 6% Beeswax coating.

**Total soluble solids (TSS)**

The study revealed that there was a significant (P ≤ .001) interaction between NLE dipping and BW coatings on TSS content of tomato fruits during 36 days of storage period under ambient conditions (Table 2). According to the study, the application BW coating with NLE dipping and sole BW coating showed a gradual increase in TSS value of the fruits up to 32nd day followed by a decrease. Whereas sole application of NLE dipping resulted in an increasing trend in TSS content of the treated fruits up to 28th day of storage followed by a drop. The other finding was that TSS content of untreated tomato fruits (control sample, NLE0*BW0) showed an increasing trend up to 24th day of storage then onwards a decrease.

The TSS values of tomato fruits in this study ranged from 4.5 °Brix to 5 °Brix which is in agreement with the findings of Tigist et al.,[36] where the TSS values of different varieties of tomato fruit range from 4 to 6 °Brix. The faster TSS increment in the untreated fruits might be due to faster metabolic activities through respiration and transpiration than fruits treated with combined application of BW coating and NLE dipping. Jholgiker & Reddy[46] indicated that a gradual increment in TSS of coated fruits is the twin role of the coating material, acting as a physical barrier for transpiration losses and creating a modified atmosphere resulting in building of internal CO2 and depletion of O2.
Table 2. The interaction effect of Neem leaf extract dipping (NLE) and beeswax coating (BW) on the mean total soluble solids (TSS) content (°Brix, ±SD) of tomato fruits during 36 days of storage period.

| Treatments | Storage period (days) | Total soluble solids (°Brix) |
|------------|-----------------------|-----------------------------|
|            | 4    | 8    | 12   | 16   | 20   | 24   | 28   | 32   | 36   |
| Neem leaf extract dipping (%) | Beeswax coating (%) | 4.650±(0.01) | 4.690±(0.01) | 4.740±(0.01) | 4.800±(0.01) | 4.900±(0.01) | 4.993±(0.01) | 4.780±(0.01) | -    | -    |
| 0          | 3    | 4.610±(0.01) | 4.650±(0.01) | 4.690±(0.01) | 4.740±(0.01) | 4.810±(0.01) | 4.897±(0.01) | 4.904±(0.01) | 4.940±(0.01) | 4.900±(0.00) |
| 0          | 6    | 4.580±(0.01) | 4.610±(0.02) | 4.640±(0.01) | 4.700±(0.01) | 4.770±(0.01) | 4.853±(0.01) | 4.903±(0.01) | 4.953±(0.01) | 4.933±(0.01) |
| 0          | 9    | 4.560±(0.01) | 4.590±(cd) | 4.620±(cd) | 4.660±(cd) | 4.720±(cd) | 4.783±(cd) | 4.887±(de) | 4.973±(b) | 4.943±(b) |
| 15         | 0    | 4.640±(f) | 4.680±(a) | 4.730±(a) | 4.790±(a) | 4.863±(a) | 4.943±(a) | 4.997±(a) | 4.810±(a) | -    |
| 15         | 3    | 4.600±(e) | 4.620±(e) | 4.650±(e) | 4.710±(e) | 4.750±(e) | 4.833±(e) | 4.910±(e) | 4.943±(e) | 4.913±(e) |
| 15         | 6    | 4.570±(d) | 4.597±(de) | 4.630±(d) | 4.690±(d) | 4.760±(d) | 4.820±(d) | 4.897±(d) | 4.973±(d) | 4.933±(d) |
| 15         | 9    | 4.540±(b) | 4.560±(b) | 4.593±(b) | 4.630±(b) | 4.690±(b) | 4.773±(b) | 4.890±(b) | 4.993±(b) | 4.953±(b) |
| 20         | 0    | 4.630±(a) | 4.670±(a) | 4.720±(a) | 4.780±(a) | 4.850±(a) | 4.940±(a) | 4.997±(a) | 4.823±(a) | -    |
| 20         | 3    | 4.590±(a) | 4.610±(a) | 4.640±(a) | 4.680±(a) | 4.740±(a) | 4.813±(a) | 4.907±(a) | 4.953±(a) | 4.923±(a) |
| 20         | 6    | 4.550±(c) | 4.580±(c) | 4.610±(c) | 4.650±(c) | 4.700±(c) | 4.783±(c) | 4.900±(c) | 4.983±(c) | 4.933±(c) |
| 20         | 9    | 4.530±(b) | 4.550±(b) | 4.580±(b) | 4.620±(b) | 4.670±(b) | 4.747±(b) | 4.867±(b) | 5.000±(b) | 4.973±(b) |
| 25         | 0    | 4.620±(a) | 4.650±(a) | 4.690±(a) | 4.740±(a) | 4.800±(a) | 4.953±(a) | 4.993±(a) | 4.843±(a) | -    |
| 25         | 3    | 4.580±(b) | 4.600±(b) | 4.630±(b) | 4.670±(b) | 4.720±(b) | 4.803±(b) | 4.903±(b) | 4.963±(b) | 4.943±(b) |
| 25         | 6    | 4.540±(c) | 4.560±(c) | 4.590±(c) | 4.630±(c) | 4.690±(c) | 4.763±(c) | 4.873±(c) | 5.000±(c) | 4.963±(c) |
| 25         | 9    | 4.520±(d) | 4.540±(d) | 4.570±(d) | 4.610±(d) | 4.680±(d) | 4.730±(d) | 4.853±(d) | 5.000±(d) | 4.983±(d) |

P value 0.948 0.631 0.144 0.009 0.0001 0.0001 0.0001 0.0001 0.0001

a-m Means within a column not sharing a superscript letter are significantly different (p < .05). SD = standard deviation.
The low TSS values of tomato fruits that received the combination of BW coating with NLE dipping probably be due to the slow rate of respiration linked to the modified atmosphere created by the coating materials. Rohani et al.,[47] noted that low respiration rate slows down the synthesis and use of metabolites resulting in lower TSS content. Islam et al.,[48] indicated that mango fruits coated with 20% Neem leaf extract retained the maximum TSS compared to others.

Notably, the combination application of BW coating with NLE dipping showed better retention and a gradual increase in the TSS value of tomato fruits up to the 32nd day of storage. Ahmed[40] reported a gradual increase in the TSS level of Neem leaf extract treated fruits. The finding of the current study is also supported by Rathore et al.,[45] who reported banana fruits treated with Neem extract (20%) resulted in slower rate of sugar increase in comparison to control. Eshetu et al.,[22] also stated that mango fruits coated with both beeswax and chitosan (2%) significantly delayed the rate increase in TSS content of the fruits.

**Titratable acidity**

The study revealed that the interaction effect of NLE dipping and BW coatings were significant (P ≤ .001) on titratable acidity (TA) of tomato fruits during 36 days of storage period under ambient conditions (Table 3). According to the study, TA of tomato fruits showed a general decreasing trend during the first 12 days of storage followed by an increase and the fruits attained maximum value of TA on 16th day and then a decline afterwards. The maximum TA was recorded in fruits treated with 9%BW and 25%NLE (0. 90%) on the 16th day of storage, followed by fruits treated with 9% BW and 20% NLE (0.89%). The lowest value of titratable acidity was recorded in untreated tomato fruits (0.72%) on the 26th day of storage. Tomato fruits that received BW coating contained higher amount of titratable acidity compared to NLE dipped fruits.

Titratable acidity of tomato fruits was significantly (P ≤ .001) different among the BW coating and NLE dipping treatments and varied from 0.98% (on the 4th day) to 0.22% (on the 36th day) of the storage period. This result is in agreement with the findings of Davies & Hobson; Getinet et al.; Salunkhe,[35,49,50] where TA content of tomato fruits ranged from 0.889% at harvest to as low as 0.25% at the end of the storage period. Titratable acidity of tomato fruits in this study showed that a general trend of a decrease during the first 12 days of storage followed by an increase and the fruits attained maximum value of TA on 16th day and then a decline afterwards. A decreasing trend of TA with the advancement of the storage period could be due to utilization of the organic acids of the fruits by respiration and other metabolic processes. The results are in line with the findings reported by Sonkar et al. and Mohammed et al.,[31,51] The authors reported similar relationship in the changes of titratable acidity of tomatoes during ripening and storage, where overall acidity slightly increased soon after harvest and then tended to decrease throughout the storage period. The higher loss of titratable acidity during the storage time could be related to higher respiration rate as ripening advances where organic acids are used as substrate in respiration process.[52]

Bhattarai & Gautam[53] stated that during storage fruit utilizes the acids as respiration substrate and, thus, the acid in tomato fruits decreases with the advance of storage time. This view has been further substantiated by Ramana et al.[54] change in TA during storage was mainly due to the metabolic activities of living tissues which deplete organic acids (e.g. malic and citric acid) of the fruits. Further, Ramana et al.[54] also reported a decrease in TA linked with an increase in total sugars and TSS content of mandarin fruits during storage at room temperature. In general, fruit acidity tends to decrease with maturation and a concomitant increase in sugar content of cherry tomato fruit.[55]

Tomato fruits treated with both BW coating and NLE dipping maintained better titratable acidity over sole BW or NLE treated fruits. Combination treatment of BW coating and NLE dipping could slow down the dissociation of organic acids by lowering the rate of respiration, ethylene production, and other metabolic activity of the tomato fruits compared to sole application of BW coating or NLE dipping. Studies reported that the application of beeswax (12%) coatings[56] and Neem leaf extract (20%)[57] showed higher retention of titratable acidity in sweet orange and
### Table 3. The interaction effect of Neem leaf extract dipping (NLE) and beeswax coating (BW) on the mean titratable acidity (% citric acid, ±SD) of tomato fruits during 36 days of storage period.

| Neem leaf extract dipping (%) | Beeswax coating (%) | Storage period (days) |
|------------------------------|---------------------|-----------------------|
|                              | 4                   | 8                     | 12                    | 16                    | 20                    | 24                    | 28                    | 32                    | 36                    |
| 0                            | 0.74±(0.01)         | 0.580±(0.01)          | 0.600±(0.01)          | 0.720±(0.01)          | 0.560±(0.01)          | 0.400±(0.01)          | 0.240±(0.01)          | -                     | -                     |
| 0                            | 3                   | 0.840±(0.01)          | 0.720±(0.01)          | 0.580±(0.01)          | 0.790±(0.01)          | 0.670±(0.01)          | 0.550±(0.01)          | 0.500±(0.01)          | 0.377±(0.01)          | 0.217±(0.01)          | 0.273±(0.03)      |
| 0                            | 6                   | 0.873±(0.01)          | 0.767±(0.02)          | 0.673±(0.04)          | 0.840±(0.01)          | 0.733±(0.01)          | 0.627±(0.02)          | 0.527±(0.02)          | 0.417±(0.02)          | 0.273±(0.03)          | 0.307±(0.03) |
| 0                            | 9                   | 0.910±(0.01)          | 0.810±(0.01)          | 0.700±(0.01)          | 0.850±(0.01)          | 0.750±(0.01)          | 0.650±(0.01)          | 0.550±(0.01)          | 0.440±(0.01)          | 0.307±(0.03)          | 0.307±(0.03) |
| 15                           | 0                   | 0.760±(0.01)          | 0.610±(0.01)          | 0.577±(0.12)          | 0.740±(0.01)          | 0.590±(0.01)          | 0.440±(0.01)          | 0.320±(0.01)          | 0.187±(0.02)          | -                     | -                     |
| 15                           | 3                   | 0.860±(0.01)          | 0.750±(0.01)          | 0.617±(0.08)          | 0.810±(0.01)          | 0.720±(0.01)          | 0.610±(0.01)          | 0.500±(0.01)          | 0.410±(0.01)          | 0.247±(0.03)          | 0.247±(0.03) |
| 15                           | 6                   | 0.890±(0.01)          | 0.783±(0.01)          | 0.703±(0.02)          | 0.860±(0.01)          | 0.770±(0.01)          | 0.670±(0.01)          | 0.570±(0.01)          | 0.450±(0.01)          | 0.320±(0.01)          | 0.320±(0.02) |
| 15                           | 9                   | 0.930±(0.01)          | 0.830±(0.01)          | 0.750±(0.02)          | 0.870±(0.01)          | 0.790±(0.01)          | 0.700±(0.01)          | 0.610±(0.01)          | 0.493±(0.02)          | 0.373±(0.02)          | 0.373±(0.02) |
| 20                           | 0                   | 0.800±(0.01)          | 0.660±(0.01)          | 0.627±(0.01)          | 0.750±(0.01)          | 0.630±(0.01)          | 0.490±(0.01)          | 0.440±(0.01)          | 0.303±(0.01)          | -                     | -                     |
| 20                           | 3                   | 0.877±(0.01)          | 0.777±(0.01)          | 0.647±(0.07)          | 0.820±(0.01)          | 0.737±(0.01)          | 0.633±(0.02)          | 0.533±(0.02)          | 0.423±(0.02)          | 0.287±(0.02)          | 0.287±(0.02) |
| 20                           | 6                   | 0.920±(0.01)          | 0.820±(0.01)          | 0.740±(0.03)          | 0.833±(0.02)          | 0.760±(0.01)          | 0.680±(0.01)          | 0.590±(0.01)          | 0.483±(0.02)          | 0.363±(0.02)          | 0.363±(0.02) |
| 20                           | 9                   | 0.950±(0.01)          | 0.860±(0.01)          | 0.793±(0.02)          | 0.890±(0.01)          | 0.820±(0.01)          | 0.750±(0.01)          | 0.670±(0.01)          | 0.577±(0.02)          | 0.467±(0.02)          | 0.467±(0.02) |
| 25                           | 0                   | 0.820±(0.01)          | 0.690±(0.01)          | 0.663±(0.14)          | 0.760±(0.01)          | 0.650±(0.01)          | 0.520±(0.01)          | 0.470±(0.01)          | 0.323±(0.02)          | -                     | -                     |
| 25                           | 3                   | 0.900±(0.01)          | 0.810±(0.01)          | 0.697±(0.08)          | 0.810±(0.01)          | 0.720±(0.01)          | 0.660±(0.01)          | 0.580±(0.01)          | 0.470±(0.03)          | 0.347±(0.03)          | 0.347±(0.03) |
| 25                           | 6                   | 0.940±(0.01)          | 0.860±(0.01)          | 0.787±(0.03)          | 0.850±(0.01)          | 0.750±(0.01)          | 0.690±(0.01)          | 0.620±(0.01)          | 0.527±(0.02)          | 0.417±(0.02)          | 0.417±(0.02) |
| 25                           | 9                   | 0.980±(0.01)          | 0.910±(0.01)          | 0.837±(0.03)          | 0.900±(0.01)          | 0.840±(0.01)          | 0.780±(0.01)          | 0.720±(0.01)          | 0.637±(0.02)          | 0.537±(0.02)          | 0.537±(0.02) |

P value: 0.530 0.238 1.000 0.002 <0.0001 0.000 <0.0001 <0.0001 <0.0001 <0.0001

*a-o* Means within a column not sharing a superscript letter are significantly different (p < .05). SD = standard deviation.
mandarins, respectively. These findings support the current study and higher TA of both BW and NLE treated tomato fruits could explain their lower incidence of fungal infection compared with untreated tomato fruits.

**pH**

In this study significant (P ≤ .001) difference in pH of tomato fruits was observed due to the interaction effect of NLE dipping and BW coatings during 36 days of storage under ambient conditions (Table 4). Beeswax coating alone or in combination with NLE dipping resulted in significantly lower pH value compared to sole NLE dipped or the control sample (BW or NLE untreated tomato fruits).

The pH value of the tomato fruits showed an increment from 3.99 to 4.69 for the control fruits, which is in agreement with the findings of Tefera et al. and Tigist et al. The authors reported an increase in the pH value of processing and fresh market tomatoes from 3.37 and 4.92 during 32 days of storage under ambient conditions. Getinet al. and Islam et al. also reported an increase in pH of tomato fruits with maturity and mango fruits during storage, respectively. An increasing trend of pH during storage could probably be due to the oxidation of organic acids. The tendency of increasing pH value and reduced acidity level observed with the advancement of the storage time.

According to this study, beeswax coating alone or in combination with NLE dipping resulted in a significantly lower pH value compared to sole NLE dipped or the control sample (BW or NLE untreated tomato fruits). This probably is attributed to the alteration or slow rate of respiration and metabolic activity of the tomato fruit due to BW or NLE treatments. Which is in agreement with the findings of Eshetu al. and Shrestha et al. where minimum pH was recorded in beeswax (2%) and Neem leaf extract (20%) treated mango fruits, respectively.

In the present study extreme significant negative correlations (r² = −0.963, p < .05) observed between TA and pH of tomato fruits (Table 9). This is probably be due to the fact that high titratable acidity fruits have high free organic acids linked with less hydrogen ion concentration. Saliba-Colombani et al. and Georgelis et al. reported that TA was negatively correlated to pH.

**Ascorbic acid content**

The study revealed that there was a significant (P < .001) interaction between NLE dipping and BW coating on the ascorbic acid contents of tomato fruits during 36 days of storage period under ambient conditions (Table 5). According to the study, tomato fruits treated with a combination of BW coating and NLE dipping resulted in significantly higher ascorbic acid content compared to sole applications of either NLE or BW and the control samples. The highest AA content of tomato fruits was recorded from the combined application of 9% BW and 25% NLE on storage day 28th (13.52 mg/100 g), 32nd (14.55 mg/100 g), and 36th (12.7 mg/100 g) compared to the remaining treatment combinations on the respective storage days. The AA content of the tomato fruits showed first slight decline from the 4th to 12th day of storage followed by an increasing trend until they reached their peaks on day 20th and followed by a decline for sole application of BW or NLE and the control samples. Whereas the combined application of BW coating with NLE dipping resulted in an initial decline up to 12th day of storage followed by a gradual increasing trend to 32nd day of storage and then a decline in AA content of the treated tomato fruits.

The ascorbic acid content of the tomato fruits ranged from 5.01 mg 100 g⁻¹ to 14.55 mg 100 g⁻¹ and showed first slight decline followed by an increasing trend until they reached their peaks and followed by a decline. This trend was in agreement with the previous data that AA content increased with ripeness. For example, an increase in ascorbic acid content in guava fruit is thought to be an indication that the guava fruit is still in the ripening stage (fully ripe guava fruits had high ascorbic acid
(136.4–247.9 mg 100 g⁻¹), while a decrease indicates a senescent fruit. Accordingly, sole BW coated or NLE dipped and untreated tomato fruits attained fully ripe stage or ascorbic acid peaks on day 20th and followed by a decline. Whereas tomato fruits treated with both BW coating and NLE dipping attained fully ripe stage or ascorbic acid peaks on day 32nd and then a decline. The higher AA content of tomato fruits treated with both BW coating and NLE dipping near the end of the storage period could be due to a slower rate of metabolic activities. This could have a better implication toward the maintenance of higher AA content in tomato fruits treated with both BW coating and NLE dipping. The general trend in the ascorbic acid content of BW and NLE treated tomato fruits is in harmony with the findings of Ali et al. and Fufa et al who reported similar trend when tomato fruits are coated with gum Arabic and plant extract with coating material, respectively.

The retention of ascorbic acid was high in tomato fruits treated with both BW coating and NLE dipping and sole BW or NLE treated tomato fruits compared to the control samples. This could probably be due to BW coating and NLE dipping of tomato fruits slow down or retard the oxidation of ascorbic acid into dehydro-ascorbic acid in presence of the ascorbate peroxidase enzyme. Mapson reported that GA3 at 500 ppm and 10% (NLE) treatment of tomato fruits resulted in maximum retention of ascorbic acid than control. Singh et al. also reported that Neem leaf extract dipping resulted in retardation of ripening and oxidation of ascorbic acid of mango fruits.

**Percentage marketability**

In this study significant (P ≤ .001) difference in the percentage of marketable tomato fruits was observed due to the interaction effect of NLE dipping and BW coatings during 36 days of storage under ambient conditions (Table 6). The combined application of BW coating with NLE dipping resulted in the highest percent marketable fruits throughout the storage periods. Interestingly, tomato fruits treated with 9% BW*25% NLE and 9% BW*20% NLE kept 100% marketability up to 20th day of storage. On the other hand, sole application of BW coating or NLE dipping and the control samples showed a lower percent marketable fruit compared to the other treatments. Beeswax coating or NLE dipping untreated tomato fruits were discarded after 28th day of storage, whereas tomato fruits treated with the combination of 9% BW*25% NLE and 9% BW*20% had the highest percentage of marketable fruits (82.85%) on the same date. On the 36th day of storage, all treatment combinations consisting of 0% BW were discarded (irrespective of the NLE dipping treatments).

The percentage marketability of tomato fruits showed a general trend of decrease during the 36 days of storage period. A similar trend was observed in PWL of tomato fruits under the different treatment conditions, this could probably be because water loss from fruits equates to a loss of salable weights and volumes in marketing. Mutari and Debbie indicated that postharvest treatments that minimize weight loss, excessive shrinkage, and spoilage usually enhance the marketability of fruits. According to this study, a strong negative correlation was observed between percentage marketable fruits and physiological weight loss ($r^2 = -0.949$, p < .05). The combined application of BW coating with NLE dipping and sole BW or NLE treatments decreased the PWL while maintaining the percentage of marketable fruits throughout the storage periods.

The highest percentage of unmarketable fruits in the control samples was due to visible mold growth, blossom end rot, and blotchy ripening. Shriveling of fruits was also a serious problem encountered in tomatoes under the various treatment conditions that contributed to a reduction of the number of marketable fruits as the storage period advanced which is in agreement with the findings of Getinet et al. and Tigist et al. The present study clearly indicates that Neem leaf extract dipping and BW coatings had significantly maintained the marketability of fruits, particularly, treatment with 9% BW + 25% NLE consistently kept the highest percentage of marketable fruits up to the last date of storage. This result is similar to the findings of Verghese who studied the efficacy of botanical extracts of Neem, Mahua and Mentha leaves on pomegranate fruit and reported that these extracts were effective in retaining marketable quality beyond 22 days of storage.
Table 4. The interaction effect of Neem leaf extract (NLE) dipping and beeswax (BW) coating on the mean pH value (±SD) of tomato fruits during storage 36 days of storage period.

| Treatments | pH value | Storage period (days) |
|------------|----------|-----------------------|
|             |          | 4  | 8  | 12 | 16 | 20 | 24 | 28 | 32 | 36 |
| Neem leaf extract dipping (%) | Beeswax coating (%) |  |  |  |  |  |  |  |  |  |
| 0          | 0        | 3.900 (0.01) | 4.080 (0.01) | 4.150 (0.01) | 4.220 (0.01) | 4.400 (0.01) | 4.550 (0.01) | 4.690 (0.01) | -  | -  |
| 0          | 3        | 3.840 (0.01) | 3.890 (0.01) | 3.950 (0.01) | 3.980 (0.01) | 4.120 (0.01) | 4.230 (0.01) | 4.340 (0.01) | 4.550 (0.01) | 4.680 (0.01) |
| 0          | 6        | 3.810 (0.01) | 3.860 (0.01) | 3.910 (0.01) | 3.930 (0.01) | 4.060 (0.01) | 4.160 (0.01) | 4.250 (0.01) | 4.460 (0.01) | 4.660 (0.05) |
| 0          | 9        | 3.790 (0.01) | 3.823 (0.02) | 3.860 (0.01) | 3.890 (0.01) | 3.980 (0.01) | 4.070 (0.01) | 4.160 (0.01) | 4.350 (0.01) | 4.550 (0.01) |
| 15         | 0        | 3.870 (0.01) | 3.950 (0.01) | 4.020 (0.01) | 4.080 (0.01) | 4.250 (0.01) | 4.390 (0.01) | 4.530 (0.01) | 4.880 (0.16) | -  |
| 15         | 3        | 3.830 (0.01) | 3.880 (0.01) | 3.940 (0.01) | 3.970 (0.01) | 4.110 (0.01) | 4.220 (0.01) | 4.330 (0.01) | 4.540 (0.01) | 4.660 (0.01) |
| 15         | 6        | 3.800 (0.01) | 3.840 (0.01) | 3.890 (0.01) | 3.910 (0.01) | 4.040 (0.01) | 4.140 (0.01) | 4.230 (0.01) | 4.440 (0.01) | 4.590 (0.01) |
| 15         | 9        | 3.760 (0.01) | 3.797 (0.01) | 3.833 (0.02) | 3.850 (0.01) | 3.963 (0.01) | 4.050 (0.02) | 4.140 (0.03) | 4.327 (0.01) | 4.540 (0.01) |
| 20         | 0        | 3.860 (0.01) | 3.930 (0.01) | 4.000 (0.01) | 4.060 (0.01) | 4.210 (0.01) | 4.340 (0.01) | 4.470 (0.01) | 4.630 (0.01) | -  |
| 20         | 3        | 3.820 (0.01) | 3.870 (0.01) | 3.920 (0.01) | 3.950 (0.01) | 4.070 (0.01) | 4.170 (0.01) | 4.270 (0.01) | 4.470 (0.01) | 4.640 (0.01) |
| 20         | 6        | 3.780 (0.01) | 3.823 (0.02) | 3.857 (0.01) | 3.860 (0.01) | 4.010 (0.01) | 4.100 (0.01) | 4.190 (0.01) | 4.380 (0.01) | 4.570 (0.01) |
| 20         | 9        | 3.750 (0.01) | 3.780 (0.01) | 3.810 (0.01) | 3.820 (0.01) | 3.930 (0.01) | 4.010 (0.01) | 4.090 (0.01) | 4.270 (0.01) | 4.527 (0.02) |
| 25         | 0        | 3.850 (0.01) | 3.910 (0.01) | 3.980 (0.01) | 4.020 (0.01) | 4.170 (0.01) | 4.290 (0.01) | 4.410 (0.01) | 4.610 (0.01) | -  |
| 25         | 3        | 3.810 (0.01) | 3.850 (0.01) | 3.900 (0.01) | 3.923 (0.01) | 4.050 (0.01) | 4.150 (0.01) | 4.250 (0.01) | 4.450 (0.01) | 4.600 (0.01) |
| 25         | 6        | 3.770 (0.01) | 3.793 (0.01) | 3.827 (0.01) | 3.850 (0.01) | 3.957 (0.01) | 4.040 (0.02) | 4.123 (0.03) | 4.313 (0.03) | 4.530 (0.01) |
| 25         | 9        | 3.740 (0.01) | 3.750 (0.01) | 3.770 (0.01) | 3.780 (0.01) | 3.890 (0.01) | 3.950 (0.01) | 4.030 (0.01) | 4.210 (0.01) | 4.500 (0.01) |
| P value    | <0.0001  | <0.0001  | <0.0001  | <0.0001  | <0.0001  | <0.0001  | <0.0001  | <0.0001  | <0.0001  | <0.0001  |

a-m Means within a column not sharing a superscript letter are significantly different (p < .05). SD= standard deviation.
Table 5. The interaction effect of Neem leaf extract (NLE) dipping and beeswax (BW) coating on the mean ascorbic acid (AA) content (mg/100 g, ±SD) of tomato fruits during storage 36 days of storage period.

| Treatments | Ascorbic Acid (mg 100 g⁻¹) |
|------------|----------------------------|
|            | Storage period (days)      |
|            | 4  | 8  | 12 | 16 | 20 | 24 | 28 | 32 | 36 |
| Neem leaf extract dipping (%) | Beeswax coating (%) |  |  |  |  |  |  |  |  |  |
| 0          | 0  | 8.297 b (0.03) | 7.563 b (0.02) | 6.017 b (0.02) | 10.863 b (0.02) | 12.820 b (0.02) | 10.083 b (0.02) | 9.253 b (0.01) |  | - |
| 0          | 3  | 5.717 b (0.02) | 5.740 b (0.02) | 6.117 b (0.02) | 8.320 b (0.02) | 9.573 b (0.02) | 12.500 b (0.02) | 12.090 b (0.01) | 12.983 b (0.02) | 7.877 b (0.09) |
| 0          | 6  | 5.878 b (0.02) | 5.570 b (0.02) | 5.937 b (0.02) | 7.930 b (0.01) | 9.163 b (0.02) | 12.010 b (0.02) | 12.600 b (0.01) | 13.440 b (0.01) | 8.817 b (0.02) |
| 0          | 9  | 5.463 b (0.02) | 5.300 b (0.02) | 5.627 b (0.02) | 7.590 b (0.01) | 8.773 b (0.02) | 11.520 b (0.02) | 12.890 b (0.01) | 13.870 b (0.01) | 11.067 b (0.02) |
| 15         | 0  | 6.693 b (0.02) | 6.140 b (0.02) | 6.147 b (0.02) | 9.230 b (0.01) | 11.543 b (0.02) | 12.540 b (0.02) | 10.130 b (0.01) | 7.860 b (0.01) |  |
| 15         | 3  | 5.833 b (0.02) | 5.640 b (0.02) | 6.017 b (0.02) | 8.120 b (0.01) | 9.363 b (0.02) | 12.570 b (0.02) | 12.280 b (0.01) | 13.260 b (0.01) | 8.270 b (0.02) |
| 15         | 6  | 5.713 b (0.02) | 5.530 b (0.02) | 5.630 b (0.02) | 7.870 b (0.01) | 9.093 b (0.02) | 11.890 b (0.02) | 12.550 b (0.01) | 13.753 b (0.01) | 10.213 b (0.02) |
| 15         | 9  | 5.367 b (0.02) | 5.210 b (0.02) | 5.527 b (0.02) | 7.470 b (0.01) | 8.643 b (0.02) | 11.320 cde (0.02) | 13.060 b (0.01) | 14.500 b (0.01) | 11.903 b (0.02) |
| 20         | 0  | 6.253 b (0.02) | 5.890 b (0.02) | 5.917 b (0.02) | 8.770 b (0.01) | 11.143 b (0.02) | 13.030 b (0.02) | 11.310 c (0.01) | 8.760 c (0.01) |  |
| 20         | 3  | 5.803 b (0.02) | 5.580 b (0.02) | 5.957 b (0.02) | 7.950 b (0.01) | 9.193 b (0.02) | 12.040 b (0.02) | 12.500 b (0.01) | 13.350 b (0.01) | 8.553 d (0.02) |
| 20         | 6  | 5.413 cd (0.02) | 5.240 d (0.02) | 5.567 cd (0.02) | 7.530 c (0.01) | 8.703 c (0.02) | 11.410 d (0.02) | 13.130 m (0.01) | 13.920 i (0.01) | 11.543 c (0.02) |
| 20         | 9  | 5.217 b (0.02) | 5.090 b (0.02) | 5.417 b (0.02) | 7.360 b (0.01) | 8.510 b (0.02) | 11.100 c (0.02) | 13.440 c (0.01) | 14.340 c (0.01) | 12.427 c (0.03) |
| 25         | 0  | 6.093 b (0.02) | 5.820 b (0.02) | 5.867 b (0.02) | 8.420 b (0.01) | 10.113 b (0.02) | 12.643 b (0.05) | 12.340 c (0.01) | 9.740 d (0.01) |  |
| 25         | 3  | 5.617 b (0.02) | 5.410 f (0.02) | 5.777 e (0.02) | 7.760 g (0.01) | 8.917 e (0.06) | 11.780 f (0.02) | 12.750 g (0.01) | 13.540 e (0.01) | 9.390 f (0.02) |
| 25         | 6  | 5.263 b (0.02) | 5.130 c (0.02) | 5.397 b (0.02) | 7.430 c (0.01) | 8.587 c (0.01) | 11.220 cd (0.02) | 13.310 h (0.01) | 14.150 m (0.01) | 12.050 f (0.02) |
| 25         | 9  | 5.013 b (0.02) | 4.983 d (0.02) | 5.237 (0.02) | 7.170 c (0.01) | 8.303 c (0.02) | 10.850 b (0.02) | 13.520 p (0.01) | 14.550 p (0.01) | 12.703 m (0.02) |

P value 0.0001 0.0001 0.0001 0.0001 0.0001 0.0001 0.0001 0.0001 0.0001

a-p Means within a column not sharing a superscript letter are significantly different (p < .05). SD= standard deviation.
Table 6. The interaction effect of Neem leaf extract dipping (NLE) and beeswax coating (BW) on the mean percentage marketability (%), ±SD of tomato fruits during storage 36 days of storage period.

| Treatments                          | Marketability of tomatoes (%) | Storage period (days) |
|-------------------------------------|-------------------------------|-----------------------|
| Neem leaf extract dipping (%)       | 8                             | 12                    | 16                    | 20                  | 24                  | 28                  | 32                  | 36                  |
| Beeswax coating (%)                 |                               |                       |                       |                     |                     |                     |                     |                     |
| 0                                   |                               |                       |                       |                     |                     |                     |                     |                     |
| 0                                   | 94.187 a (3.00)               | 91.423 a (2.86)       | 85.710 a (2.86)       | 79.997 a (2.86)     | 71.423 a (2.86)     | 57.140 a (2.86)     |                     |                     |
| 3                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 91.423 bc (2.86)      | 85.710 b (2.86)     | 82.853 bc (2.86)    | 74.187 bc (3.00)    | 65.710 c (2.86)     | 52.373 h (1.65)    |
| 6                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 94.280 cd (2.86)      | 91.423 cd (2.86)    | 85.710 cd (2.86)    | 79.997 de (2.86)    | 71.423 de (2.86)    | 57.140 cd (2.86)   |
| 9                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 100.000 e (0.00)      | 95.233 def (3.30)   | 88.567 def (2.86)   | 84.760 def (4.36)   | 77.140 fg (2.86)    | 62.853 ef (2.86)   |
| 15                                  | 0                             | 100.000 b (0.00)      | 94.280 b (2.86)       | 88.567 bcd (2.86)   | 85.710 b (2.86)     | 79.997 b (2.86)     | 71.423 b (2.86)     | 57.140 b (2.86)    |
|                                     |                               |                       |                       |                     |                     |                     |                     |                     |
| 3                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 94.280 cd (2.86)      | 88.567 bc (2.86)    | 84.757 bcdef (3.30) | 77.140 cd (2.86)    | 68.567 cd (2.86)    | 54.280 bc (2.86)   |
| 6                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 97.140 cde (2.86)     | 94.280 cde (2.86)   | 88.567 cde (2.86)   | 82.853 cde (2.86)   | 74.187 cde (3.00)   | 59.999 cde (2.86)  |
| 9                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 97.140 cde (2.86)     | 94.280 cde (2.86)   | 91.423 cde (2.86)   | 79.997 cde (2.86)   | 65.710 cde (2.86)   |                     |
| 15                                  | 0                             | 100.000 b (0.00)      | 97.140 cde (2.86)     | 91.423 cde (2.86)   | 82.853 cde (2.86)   | 74.187 cde (3.00)   | 59.997 cde (2.86)   |                     |
|                                     |                               | 0                     |                          | 0                   | 0                   | 0                   | 0                   |                     |
| 3                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 95.233 def (3.30)     | 91.377 cd (2.79)    | 85.710 cd (2.86)    | 79.247 de (2.86)    | 71.423 de (2.86)    | 57.140 cd (2.86)   |
|                                     |                               | 95.233 def (3.30)     | 91.377 cd (2.79)      | 85.710 cd (2.86)    | 79.247 de (2.86)    | 71.423 de (2.86)    | 57.140 cd (2.86)    |                     |
| 6                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 100.000 e (0.00)      | 97.140 cde (2.86)   | 91.423 cde (2.86)   | 85.710 cde (2.86)   | 77.140 cde (2.86)   | 62.853 cde (2.86)  |
| 9                                   | 100.000 b (0.00)              | 100.000 d (0.00)      | 100.000 e (0.00)      | 100.000 g (0.00)    | 94.280 cde (2.86)   | 91.423 cde (2.86)   | 82.853 cde (2.86)   | 68.473 gh (2.72)   |
| 20                                  | 0                             | 100.000 b (0.00)      | 98.093 cde (1.65)     | 91.423 bcd (2.86)   | 88.567 bcd (2.86)   | 85.710 cde (2.86)   | 74.187 bc (3.00)    | 60.663 b (3.08)    |
|                                     |                               | 98.093 cde (1.65)     | 91.423 bcd (2.86)     | 88.567 bcd (2.86)   | 85.710 cde (2.86)   | 74.187 bc (3.00)    | 60.663 b (3.08)    |                     |
| 25                                  | 3                             | 100.000 b (0.00)      | 97.140 cde (2.86)     | 94.280 cde (2.86)   | 88.567 cde (2.86)   | 82.853 cde (2.86)   | 71.423 de (2.86)    | 59.999 cde (2.86)  |
|                                     |                               | 97.140 cde (2.86)     | 94.280 cde (2.86)     | 88.567 cde (2.86)   | 82.853 cde (2.86)   | 71.423 de (2.86)    | 59.999 cde (2.86)   |                     |
| 25                                  | 6                             | 100.000 b (0.00)      | 100.000 d (0.00)      | 100.000 e (0.00)    | 99.047 fg (1.65)    | 91.423 cde (2.86)   | 88.567 gh (2.86)    | 77.233 fg (2.86)   |
|                                     |                               | 100.000 e (0.00)      | 100.000 e (0.00)      | 99.047 fg (1.65)    | 91.423 cde (2.86)   | 88.567 gh (2.86)    | 77.233 fg (2.86)    | 65.710 fg (2.86)   |
| 25                                  | 9                             | 100.000 b (0.00)      | 100.000 d (0.00)      | 100.000 e (0.00)    | 100.000 g (0.00)    | 94.280 cde (2.86)   | 82.853 cde (2.86)   | 71.423 fg (2.86)   |
|                                     |                               | 100.000 e (0.00)      | 100.000 e (0.00)      | 100.000 g (0.00)    | 94.280 cde (2.86)   | 82.853 cde (2.86)   | 71.423 fg (2.86)    |                     |

P value: 0.0001 0.0002 0.041 0.306 0.262 0.040 0.0001 0.133

a-i Means within a column not sharing a superscript letter are significantly different (p < .05). SD = standard deviation
Table 7. The interaction effect of Neem leaf extract dipping (NLE) and beeswax coating (BW) on the mean percentage decay (%) ±SD of tomato fruits during storage 36 days of storage period.

| Treatments | Decay (%) | Storage period (days) | 12 | 16 | 20 | 24 | 28 | 32 | 36 |
|------------|-----------|-----------------------|----|----|----|----|----|----|----|
| Neem leaf extract dipping (%) | Beeswax coating (%) | 12 | 16 | 20 | 24 | 28 | 32 | 36 |
| 0 | 0 | 5.710 b (2.86) | 14.280 a (2.86) | 22.853 b (2.86) | 34.280 a (2.86) | 72.373 b (1.65) | - | - |
| 0 | 3 | 0.000 a (0.00) | 3.803 a (1.65) | 11.423 ab (2.86) | 19.997 cd (2.86) | 25.710 a (2.86) | 32.973 b (1.65) | 71.423 a (2.86) |
| 0 | 6 | 0.000 a (0.00) | 0.000 a (0.00) | 5.710 bc (2.86) | 14.280 cd (2.86) | 17.140 ef (2.86) | 25.710 ab (2.86) | 62.570 a (2.90) |
| 0 | 9 | 0.000 a (0.00) | 0.000 a (0.00) | 2.853 ab (2.86) | 11.423 ab (2.86) | 14.280 de (2.86) | 19.997 de (2.86) | 54.280 cd (2.86) |
| 15 | 0 | 0.000 a (0.00) | 11.423 bc (2.86) | 19.997 ab (2.86) | 28.567 bc (2.86) | 34.280 b (2.86) | 68.567 b (2.86) | - |
| 15 | 3 | 0.000 a (0.00) | 0.000 a (0.00) | 8.567 cd (2.86) | 17.140 cd (2.86) | 22.853 ef (2.86) | 31.423 f (2.86) | 68.567 b (2.86) |
| 15 | 6 | 0.000 a (0.00) | 0.000 a (0.00) | 2.853 ab (2.86) | 11.423 ab (2.86) | 14.280 de (2.86) | 22.853 ef (2.86) | 57.140 d (2.86) |
| 15 | 9 | 0.000 a (0.00) | 0.000 a (0.00) | 0.000 a (0.00) | 5.710 bc (2.86) | 8.567 cd (2.86) | 17.140 cd (2.86) | 48.567 ab (2.86) |
| 20 | 0 | 0.000 a (0.00) | 8.567 cd (2.86) | 17.140 cd (2.86) | 25.710 ab (2.86) | 31.423 f (2.86) | 65.427 f (3.29) | - |
| 20 | 3 | 0.000 a (0.00) | 0.000 a (0.00) | 8.567 cd (2.86) | 17.140 cd (2.86) | 19.997 de (2.86) | 28.567 bc (2.86) | 65.427 f (3.29) |
| 20 | 6 | 0.000 a (0.00) | 0.000 a (0.00) | 0.000 a (0.00) | 0.000 a (0.00) | 5.710 bc (2.86) | 14.280 bc (2.86) | 42.853 cd (2.86) |
| 20 | 9 | 0.000 a (0.00) | 0.000 a (0.00) | 0.000 a (0.00) | 0.000 a (0.00) | 5.710 bc (2.86) | 14.280 bc (2.86) | 42.853 cd (2.86) |
| 25 | 0 | 0.000 a (0.00) | 5.710 bc (2.86) | 14.280 d (2.86) | 22.853 bc (2.86) | 28.567 bc (2.86) | 62.570 b (2.90) | - |
| 25 | 3 | 0.000 a (0.00) | 0.000 a (0.00) | 5.710 bc (2.86) | 14.280 d (2.86) | 17.140 ef (2.86) | 25.710 ab (2.86) | 59.713 ab (2.44) |
| 25 | 6 | 0.000 a (0.00) | 0.000 a (0.00) | 0.000 a (0.00) | 2.853 ab (2.86) | 5.710 bc (2.86) | 14.280 bc (2.86) | 45.710 c (2.86) |
| 25 | 9 | 0.000 a (0.00) | 0.000 a (0.00) | 0.000 a (0.00) | 0.000 a (0.00) | 2.853 ab (2.86) | 11.423 b (2.86) | 39.997 bd (2.86) |

P value: 0.0001, 0.001, 0.578, 0.218, 0.0001, 0.0001, 0.000

a-l Means within a column not sharing a superscript letter are significantly different (p < .05). SD = standard deviation.
Decay percentage

The study revealed that there was a significant \((P < .001)\) interaction between NLE dipping and BW coatings on percentage decay of tomato fruits during 36 days of storage period (Table 7). Tomato fruits treated with the combination of BW coating with NLE dipping resulted in significantly lower percent decay compared to sole application of BW or NLE. According to the study, untreated tomato fruits started to decay on the 8th day of storage whereas tomato fruits treated with NLE dipping or BW coating alone started to decay on the 12th day of the storage period. Combination treatment of BW coating and NLE dipping delayed the onset of rotting or decay in tomato fruits during the storage period. For example, the combined application of 3% BW with all levels of NLE dipping extended the onset of tomato fruits decaying up to the 20th day of the storage. The highest percentage tomato fruits decay was recorded in the control fruits throughout the storage period. In general percentage decay of tomato fruits showed a reducing trend with increasing the concentration of BW coating and NLE dipping.

The combination treatment of BW coating and NLE dipping of tomato fruit could probably reduce the growth of microbes on top of its role as a moisture inhibitor and create modified atmospheric conditions. For example, Azadirachtin is considered the most active substance in Neem which has growth-regulating, fungicidal, and insecticidal properties. The ability of Neem leaf powder to decrease the decay level of tomatoes is an indication that Neem leaf extracts can serve as a possible alternative in the prevention of tomato decay by pathogens. Tripathi and Dubey reported that wax coatings (taxol 8% and capton 0.2%) minimized the growth of fungi in mandarin.

The onset of decaying was on day 8th in untreated tomato fruits, on day 12th in tomato fruits treated with NLE dipping or BW coating alone, and on day 20th in both BW coating and NLE dipping treated tomatoes. Singh et al. and Bibi & Baloch reported that 10% Neem leaf extract and 2% beeswax coating treatments reduced microbial growth in mango fruit. Studies reported that Neem (Azadirachta Indica) extract has a good capacity in controlling various postharvest diseases of fruits, which supports the findings of the current study. Eissa also reported that the wax coating of fresh-cut mushrooms extends the shelf-life by limiting the growth of fungi and decreasing spoilage.

Shelf life

In this study significant \((p ≤ .001)\) difference in the shelf life of tomato fruits was observed due to the interaction effect of NLE dipping and BW coatings (Table 8). The study revealed that the combined application of BW coating with NLE dipping of tomato fruits significantly extended the shelf life of the fruits compared to sole application of BW or NLE and control samples. A general trend of improvement in storage life of tomato fruits observed with increasing concentration of BW coating irrespective of the levels of NLE dipping. Untreated tomato fruit maintained their shelf life for 28 days, whereas the combination treatment of BW coating and NLE dipping extended the shelf life of the tomato fruits from six to eight compared to the untreated fruits.

The combined application of BW coating and NLE dipping significantly \((p < .0001)\) extended the shelf life of tomato fruits compared to sole BW or NLE treatment. This is because the BW coating could help to create a thin layer of film that acts as a moisture barrier whereas NLE dipping could reduce or inhibit the growth of microbes. A general trend of improvement in the shelf life of tomato fruits were observed with increasing the concentration of BW coating (irrespective of the levels of NLE dipping). Notably, the BW coating could create modified atmospheric conditions (i.e., decreases \(O_2\) and increasing \(CO_2\) concentration) thereby reducing the rate of respiration and ethylene production of the treated tomato fruits. This could probably delay the ripening processes of the tomato fruits and enhance the shelf life of the fruits. Shahid & Abbasi reported that 5% beeswax coating with 0.5
Table 8. The interaction effect of Neem leaf extract dipping (NLE) and beeswax coating (BW) on the mean shelf life (days, +SD) of tomato fruits during storage 36 days of storage period.

| Neem leaf extract dipping (%) | Beeswax coating (%) | shelf-life (days) |
|------------------------------|---------------------|-------------------|
| 0                            | 0                   | 28 ± (0.00)       |
| 0                            | 3                   | 34 ± (0.58)       |
| 0                            | 6                   | 34 ± (0.00)       |
| 0                            | 9                   | 35 ± (0.00)       |
| 15                           | 0                   | 32 ± (0.58)       |
| 15                           | 3                   | 34 ± (0.00)       |
| 15                           | 6                   | 35 ± (0.00)       |
| 15                           | 9                   | 36 ± (0.00)       |
| 20                           | 0                   | 33 ± (0.58)       |
| 20                           | 3                   | 34 ± (0.00)       |
| 20                           | 6                   | 36 ± (0.58)       |
| 20                           | 9                   | 36 ± (0.00)       |
| 25                           | 0                   | 33 ± (0.00)       |
| 25                           | 3                   | 35 ± (0.58)       |
| 25                           | 6                   | 36 ± (0.00)       |
| 25                           | 9                   | 36 ± (0.00)       |
| P value                      |                     | 0.0001            |

a-f Means within a column not sharing a superscript letter are significantly different (p < .05). SD= standard deviation

banlate improved the overall quality and extended the shelf life of sweet orange fruits at room temperature. Bierhals et al.\textsuperscript{75} also reported that cassava starch coating on fresh-cut pineapple delayed the change in color, maintained quality and prolonged the storage life of the pineapples.

Microbial growth was found to be lowest in tomato fruits treated with NLE dipping which is in agreement with the findings of Siddiqua et al. and Singh et al.\textsuperscript{[44,67]} In this study, a strong negative correlation was observed between percentage decay and shelf life of tomato fruits ($r^2 = -0.939$, $p < .05$). Singh et al.\textsuperscript{[67]} reported that 10% Neem leaf extract dipping reduced the growth of microbes in mango fruits. The application of edible coatings extended the shelf life of plum fruits\textsuperscript{[14]} and strawberries.\textsuperscript{[56]} Soares et al.\textsuperscript{[77]} indicated that the use of edible coating combined with 1% and 1.5% chitson (natural antimicrobials) increased the shelf life of guava fruits.

Correlation analysis demonstrated significant positive and negative correlations of the interaction effects of Neem leaf extract dipping and beeswax coating on shelf life and quality of tomato fruits. For example, titratable acidity of Neem leaf extract dipping in combination with beeswax coating of tomato fruits showed extreme positive correlations with marketability ($r^2 = 0.929$, $p < .05$) and shelf-life ($r^2 = 0.913$, $p < .05$) (Table 9). The first three principal components (F1 and F3) explained 84.93% of the total variability of the interaction effects of Neem leaf extract dipping and beeswax coating on shelf life and quality of tomato fruits (Figure 1). Shelf life was found to be positively correlated with marketability and TA as evident from acute cosines of the angles formed between the vectors of the parameters. These observations also agreed with those found in simple matrix correlations presented in Table 9.

Notably, agglomerative hierarchical clustering (AHC) analysis of Ward’s method showed the interaction effects of Neem leaf extract dipping and beeswax coating on shelf life and quality of tomato fruits as two distinguished groups were depicted in the PCA chart (Figure 2). The first group (square symbol) was composed of Neem leaf extract dipping (0%, 15%, 20%, 25% NLE) in combination with beeswax coating (3%, 6%, 9% BW). The second group (triangle symbol) contained Neem leaf extract dipping (0%, 15%, 20%, 25% NLE) in combination with beeswax coating (0% BW). It can be depicted that among the combination treatments of beeswax coating and Neem leaf extract dipping,
Table 9. Correlation coefficient (Pearson (n)) of the interaction effect of Neem leaf extract dipping and beeswax coating on shelf life and quality of tomato fruits at the 20th day of storage period.

| Variables | TA   | WL   | AA   | pH   | TSS  | Marketability | Decay | Shelf-life |
|-----------|------|------|------|------|------|---------------|-------|------------|
| TA        |      |      |      |      |      | 0.929         |       | 0.913      |
| WL        | -0.909 | 0.831 | 0.911 | 0.925 |      |               |       |            |
| AA        | -0.934 |      | 0.967 | 0.947 |      |               |       |            |
| pH        | -0.963 |      |      | 0.959 |      |               |       |            |
| TSS       | -0.946 |      |      |      | 0.962 |               |       |            |
| Marketability | -0.949 | -0.894 | -0.956 | -0.957 |      |               | 0.998 | 0.919      |
| Decay     | -0.959 |      |      |      |      |               | -0.951 |            |
| Shelf-life | -0.841 | -0.964 | -0.968 | -0.927 |      |               | -0.939 |            |

Values in bold are different from 0 with a significance level alpha=0.05

Figure 1. Principal component analysis (PCA) of the interaction effect of Neem leaf extract dipping (NLE) and beeswax coating (BW) on shelf life and quality of tomato fruits.

9% BW coating in combination with 25% NLE dipping provided the maximum enhancement of the shelf life (extended the shelf life by 8 days compared to the control sample) while maintaining the quality of tomato fruits during the 36 days of storage period.
**Conclusion**

The application of BW coating in combination with NLE dipping significantly increased the shelf life indicating a decrease in the percent decay and changes in the quality of tomato fruits during the 36 days of storage period. The change in the quality parameters was largely dependent on the concentrations of NLE dipping and BW coating. It was observed that the application of 9% BW coating in combination with 25% NLE dipping provided the maximum enhancement in shelf life and effectively maintained the percentage marketable fruits, TSS, AA, and TA content of tomato fruits during the 36 days of storage period. The study result showed that 9% BW coating in combination with 25% NLE dipping could potentially be utilized to extend the shelf life and maintain the quality of tomato fruits by farmers, retailers and wholesalers.

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**Authors’ contributions**

BirukZewde: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Writing (first draft). KebedeWoldetsadik and Tigrst T. Shone: Conceived and designed the experiments; Supervision; Writing (second draft).
**Data availability of materials statement**

All the data used to support this study have been deposited in the Dryad repository (Available at: https://doi.org/10.5061/dryad.5mkkwh772).

**Disclosure statement**

No potential conflict of interest was reported by the author(s).

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