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Quality characteristics of stored tomato fruit treated with two formulations of African black pepper

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Abstract. Tomato *Solanum lycopersicum* L. (Solanales: Solanaceae) is highly perishable and requires postharvest treatment to extend its shelf life. Use of synthetic chemicals to control post-harvest loss in tomato has adverse effect on health and there is need to explore natural alternatives to chemical. Two formulations of African black pepper *Piper guineense* Schumach. (Piperales: Piperaceae), aqueous extract and essential oil, were evaluated as preservatives for Roma type tomato fruit. Tomato fruits were treated with different concentrations of aqueous extracts and undiluted essential oil and stored under refrigeration condition. Samples were taken at 5-days interval and analyzed for weight loss, total soluble solids and ascorbic acid. Total Viable Count (TVC) and total mould count (TMC) were determined. Chemical composition of essential oil was identified using gas chromatography-mass spectrometer. Percent weight loss (PWL) in aqueous extract-treated tomato (0.0%-0.68%) was lower than the control (0.3%-19.97%). The total soluble solid (brix) of samples in untreated fruit was lower than fruit treated with higher of *P. guineense*. Ascorbic acid contents were higher in aqueous extract-treated samples than the control. Essential oil-treated fruit had lower physiological weight loss TVC and TMC than the control. Twelve compounds were identified in *P. guineense* essential oil, the most of which were β-sesquiphellandrene (23.7%). The *P. guineense* aqueous extract or essential oil is recommended as a bio-rational preservative for postharvest storage of tomato.

Keywords: *Piper guineense*; Tomato; Aqueous extract; Essential oil; Post-harvest storage.

Resumo. ***Características de qualidade de frutos de tomate armazenados tratados com duas formulações de pimenta preta africana***. O tomate *Solanum lycopersicum* L. (Solanales: Solanaceae) é altamente perecível e requer tratamento pós-colheita para estender seu prazo de validade. O uso de substâncias químicas sintéticas para controlar a perda pós-colheita em tomate tem efeito

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adverso sobre a saúde e há necessidade de explorar alternativas naturais às substâncias químicas. Duas formulações de pimenta preta africana *Piper guineens* Schumach. (Piperales: Piperaceae), extrato aquoso e óleo essencial, foram avaliadas como conservantes para o fruto de tomate tipo Roma. Frutos de tomateiro foram tratados com diferentes concentrações de extratos aquosos e óleo essencial não diluído e armazenados sob refrigeração. As amostras foram coletadas no intervalo de cinco dias e analisadas para perda de peso, sólidos solúveis totais e ácido ascórbico. Contagem Total Viável (CTV) e contagem total de moldes (TMC) foram determinados. A composição química do óleo essencial foi identificada usando espectrômetro de massa por cromatografia gasosa. A perda percentual de peso (PPP) no tomate tratado com extrato aquoso (0,0%-0,68%) foi menor do que no controle (0,3%-19,97%). O sólido solúvel total (brix) das amostras em frutos não tratados foi menor que os frutos tratados com maior teor de *P. guineense*. O conteúdo de ácido ascórbico foi maior em amostras tratadas com extrato aquoso do que o controle. Os frutos tratados com óleo essencial apresentaram menor perda de peso fisiológico, CTV e TMC do que o controle. Doze compostos foram identificados em óleo essencial de *P. guineense*, sendo a maior parte deles β-sesquipelenareno (23,7%). Recomenda-se o extrato aquoso de *P. guineense* ou óleo essencial como conservante bio-racional para o armazenamento pós-colheita de tomate.

Palavras-chave: *Piper guineense*; Tomate; Extrato aquoso; Óleo essencial; Estocagem pós-colheita.

Introduction

Damage to vegetable quality in storage has been attributed to mechanical, physiological and microbial effects (Idah et al., 2007). Isack and Lyimo (2015) reported that postharvest handling practices affect qualitative and quantitative attributes of tomato. Some losses limit availability of crop to consumers and there is a need to extend shelf-life of crops. Extending shelf-life of climacteric fruit is important in tropical countries where production of vegetable is seasonal (Idah et al., 2012). If production continues and postharvest losses issues are not addressed there could be food and nutrition insecurity and increased poverty among smallholder farmers. Synthetic chemicals had been used over the years to extend shelf-life of fruits and vegetables, however, retention of residues of these chemical on food has become a major concern to consumers (Plotto et al., 2003; Katalinic et al., 2013). Therefore, an alternative to the use of synthetic chemicals such as natural biodegradable compounds is necessary.

Spices and herbs have been reported to prolong shelf-life of foods due to their bacteriostatic and anti-oxidative activities (Jałosińska and Wilczak, 2009). Natural compounds are preferred by many consumers because they have reduced, or no mammalian toxicity, less environmental hazards, and wide public acceptability (Hassani et al., 2012). African black pepper *Piper guineense* Schumach. (Piperales: Piperaceae), is a spice that is widely consumed in Nigeria and Ghana due to its nutritional, antimicrobial and medicinal properties (Negbenebor et al., 1999). Okonkwo and Okoye (1996) investigated the efficacy of the crude extracts, essential oils and seed powders of *P. guineense* against the cowpea bruchid *Callosobruchus maculates* (Fabricius, 1775) (Coleoptera: Chrysomelidae). Its

preservative potential in extending shelf life of fruits and vegetables can also be investigated due to its antimicrobial and antioxidant properties.

Tomato *Solanum lycopersicum* L. (Solanales: Solanaceae) is a climacteric fruit that is rich in vitamins and minerals (Nasrin et al., 2008). It is also an excellent source of lycopene (a very powerful antioxidant) that helps to prevent development of some cancers (Upangalawar et al., 2010). Tomato fruits outrank all other vegetables in total contribution to human nutrition because so much is consumed in so many different ways. It may be possible to reduce post-harvest loss in tomato using a biodegradable compound. Ahmed et al. (2017) reported that due to increase in fungicides residues on fruit and vegetables, there is therefore need for postharvest treatments of fruit and vegetables with natural products to reduce losses.

Hence, this study was designed to evaluate the preservative potentials of African black pepper on tomato fruit.

Materials and methods

Freshly harvested tomato fruits, cv. Roma VF, were obtained from a local farm in Iresaadu, Ogbomoso, Nigeria, sorted, cleaned and washed. African black pepper was procured from Bode Market, Ibadan, Nigeria.

Preparation of *P. guineense* aqueous extract

The dried seeds of *P. guineense* were cleaned and pulverized into fine powder using a hammer mill. Aqueous extracts (1%, 2%, 3%, 4% and 5% w/v) of these seeds were prepared. The solutions were kept in the refrigerator for 5 days followed by centrifugation as described by Adegoke et al. (2002).

Extraction of essential oil

Essential oil was extracted from 500 g pulverized *P. guineense*. The pre-weighed powder of *P. guineense* was added to 2 L water in 5 L capacity round bottom flask and essential oil was extracted using hydro-distillation with a Clevenger type apparatus.

Treatment of tomato with aqueous extract and essential oil

Tomato fruits were weighed into batches of 200 g. Each batch was dipped separately into concentrations of 1%, 2%, 3%, 4% and 5% w/v in water of *P. guineense* at the rate of 1000 mL spice extract per 200 g fruit for 20 min. Another batch of 200 g was dipped into distilled water as the control. All samples were drained at ambient temperature (26 ± 2 °C) for 10 min according to Senevirathna and Daundasekera (2010). Extract-treated-fruits were placed in polyethylene packs which were sealed leaving 2.5-5 cm of head space in packs. Treated tomato fruits were stored at 5 °C for 30 days. Samples were removed at a 5-day-interval from each batch for analyses. The experiment was done in three replicates.

Freshly harvested tomato fruits were sorted to remove damaged ones. The fruits were rinsed in distilled water, drained and packaged in low density polyethylene (LDPE) bags. Cotton wool (approximately 0.5 g) was attached to the inner tip of the LDPE bags and 10 µL of the oil dispensed into the wool using a Hamilton® syringe so that the volatile oils could diffuse around tomato fruit without directly contacting them. Untreated samples were packaged using the same method but without essential oil. The LDPE bags were sealed with an electric sealer. Packaged samples were stored at 5 °C for 30 days. Samples were removed every 5 days for analyses.

Physical analyses

Physiological weight loss was determined by periodically weighing fruit and expressed as percent of original weight as described by Nasrin et al. (2008).

Chemical analyses

Total soluble solids was determined using a refractometer (Tech-Jam International Inc., Tokyo, Japan) and was expressed in terms of Brix (Akbudak and Akbudak, 2007). Ascorbic acid content in tomato pulp was determined according to Kirk and Sawyer (1991) using 2, 6, dichlorophenol indophenols visual titration.

Total viable count

The Petri dishes were in triplicate per dilution to be tested. The dishes were labeled appropriately. A portion (0.2 mL) of each dilution was pipette into the centre of appropriate dishes containing sterilized nutrient agar. The dishes were rotated in a clockwise and anticlockwise direction. The media were allowed to set, inverted and incubated at 27 °C for 48 h. Colonies were counted using an electronic counter (FISON Model CNW - 330-010X) (Collins et al., 1989).

Total mould count

A portion (0.2 mL) of each dilution was dispensed into the centre of the appropriate plate containing potato dextrose agar. The plate was allowed to set, inverted and incubated at room temperature for 48 h. All counts were carried out at regular intervals during fermentation period. Colonies were counted using the electronic counter (Collins et al., 1989).

Identification of chemical compounds of essential oil

The *P. guineense e*ssential oil was resolved in methanol. The resolved oil was injected into a GC-MS machine (GCMS-QP2010 Plus®, a product of Shimadzu, Kyoto, Japan), using an AOC-20i auto sampler. The column used was Rtx-5MS (A product of Restek, USA) (30 m x 0.25 mm internal diameter x 0.25 µm film thickness) coated with 5% diphenyl 95% dimethylpolysiloxane packing materials. The carrier gas was helium and a purge flow rate of 3 mL·min^{-1} and column flow rate 0.99 mL·min^{-1}. The GC was operated with the following conditions: oven temperature 60 °C, for 2 min, ramp of 10 °C/min up to 180 °C held for 3 min, subsequent increase to 280 °C with a 15 °C·min^{-1} heating ramp maintained at 280 °C for 3 min. Injection temperature and volume were 250 °C and 1.0 µL, respectively. The MS operating conditions were ionization with an ion trap detector in full scan mode under electron impact ionization (EI) at 70 eV, ion source temperature 200 °C; interface temperature 250 °C, scan range, 40-700 m·z^{-1}. Identifications of components was based on comparison of the relative Kovets Retention Index (RI) using a hydrocarbon homologous. The relative concentrations of components (% composition) were obtained by peak areas (Rtx-5MS column). They were estimated by dividing the area of each component by the total area of all components isolated under MS (Maggi et al., 2009).

Statistical analysis

Data of extract bioassays were analyzed by analysis of variance (ANOVA) using SPSS (2006). The Duncan multiple range test was used to separate differences among means. For essential oil bioassay, data were subjected to studentized t-test.

Results and discussion

Aqueous treated samples

Physical Properties. Treatment with extracts of *P. guineense* reduced percent weight loss compared to the control (Table 1). Percent weight loss increased as storage time progressed. There was no weight loss for 5% treated samples at day 5 but significant weight loss was observed at day 20; significantly higher losses occurred in the control. Higher concentrations of 4%-5% *P. guineense* extract had the lowest percent weight loss at the end of storage. Weight loss can be attributed to respiration rate because a carbon atom is lost from the

fruit each time a carbon-dioxide molecule is produced (Bhowmilk and Pan, 1992). Sammi and Masud (2007) treated tomato with calcium chloride, boric acid and potassium permanganate and reported significant reduction in weight of treated fruit. Reduction in weight loss could be attributed to some constituents of *P. guineense* extract leading to reduced respiration rate. Sudha et al. (2007) postulated that reduction of weight loss of tomato treated with gibberellic acid could be due to its anti-senescent action.

Table 1 Percent weight loss of *P. guineense* aqueous treated tomato fruit during refrigerated storage.

***Piper guineense* extract (%, w/v)**	**Storage period (days)**					
	5	**10**	**15**	**20**	**25**	**30**
1	0.02±0.01[a]	0.07±0.01[b]	0.18±0.01[b]	0.30±0.00[b]	0.41±0.01[b]	0.68±0.04[b]
2	0.01±0.00[a]	0.02±0.00[b]	0.13±0.01[b]	0.35±0.00[b]	0.44±0.01[b]	0.50±0.01[c]
3	0.02±0.01[a]	0.02±0.00[b]	0.12±0.01[b]	0.33±0.03[b]	0.42±0.02[b]	0.49±0.01[c]
4	0.03±0.01[a]	0.04±0.00[b]	0.14±0.03[b]	0.31±0.01[b]	0.37±0.01[b]	0.49±0.01[c]
5	0.00±0.00[a]	0.02±0.00[b]	0.13±0.03[b]	0.29±0.01[b]	0.34±0.01[b]	0.43±0.01[c]
Control	0.09±0.01[a]	0.27±0.05[a]	0.55±0.11[a]	3.45±0.12[a]	18.23±1.87[a]	19.97±1.37[a]

[a]Values in columns followed by the same letter are not significantly different (P > 0.05).

Chemical Properties. Total soluble solid (TSS) of fresh fruit decreased (Table 2) as storage progressed. Initial reduction in TSS of samples treated with *P. guineense* was observed in the first 25 days. The increase in TSS could be due to transformation of pectin substances and other polysaccharides into soluble sugar (Mounika et al., 2017). At the end of the storage, samples treated with 4% and 5% *P. guineense* had the highest brix values.

Table 2 Influence of *P. guineense* aqueous extracts on total soluble solids (brix) of tomato during refrigerated storage.

***Piper guineense* extract (%, w/v)**	**Storage period (days)**						
	0	**5**	**10**	**15**	**20**	**25**	**30**
1	3.37±0.06[a]	2.03±0.06[c]	1.73±0.06[c]	1.20±0.06[c]	1.00±0.00[c]	1.00±0.00[b]	1.27±0.06[b]
2	3.37±0.06[a]	2.33±0.06[b]	1.83±0.06[b]	1.37±0.06[b]	1.07±0.06[b]	1.03±0.06[b]	1.40±0.10[a]
3	3.37±0.06[a]	2.47±0.06[b]	1.93±0.01[b]	1.43±0.06[a]	1.10±0.06[b]	1.07±0.06[b]	1.53±0.06[a]
4	3.37±0.06[a]	2.63±0.06[a]	2.13±0.06[a]	1.47±0.06[a]	1.17±0.06[b]	1.07±0.06[b]	1.67±0.06[a]
5	3.37±0.06[a]	2.93±0.06[a]	2.47±0.06[a]	1.47±0.04[a]	1.23±0.06[a]	1.17±0.06[a]	1.80±0.00[a]
Control	3.37±0.06[a]	2.30±0.01[b]	1.70±0.10[c]	1.30±0.00[b]	1.17±0.06[b]	1.07±0.06[b]	1.00±0.00[b]

Means with similar letters along the column are not significantly (P > 0.05) different.

Ascorbic acid was lower in untreated samples on day 5 (Table 3). Ascorbic acid was further reduced on day 15 of storage and the control samples had the lowest values. Ascorbic acid was best retained in samples treated with

4%-5% *P. guineense*. At the end of storage, samples treated with 4% *P. guineense* had the highest values. Babarinde and Adegoke (2015) reported antioxidative effect of *Xylopia aethiopica* (Dunal) A. Rich. (Magnoliales: Annonaceae) when tomato was treated with aqueous extract of *X. aethiopica*. Better retention of ascorbic acid in *P. guineense* treated samples confirmed the antioxidative properties of the material (Adegoke and GopalaKrishna, 1998; Besong et al., 2016). The ascorbic acid content of tomato as reported by Akbudak and Akbudak (2007) was 28.1 mg/100 g which is higher than the value reported in this study. The reason for the variation could be due to varietal differences and maturity stage (Leonardi et al., 2000). Renhua et al. (2008) reported that exogenous pre-treatment of vegetables could change the antioxidant system and maintain their nutritional value. This treatment could be responsible for maximum ascorbic acid in *P. guineense* treated samples.

Table 3 Ascorbic acid contents (mg/100 g) of tomato treated with *P. guineense* aqueous extract during refrigerated storage.

***P. guineense* extract (%, w/v)**	**Storage period (days)**						
	0	**5**	**10**	**15**	**20**	**25**	**30**
1	22.00±0.00^{a}	19.27±0.23^{b}	16.13±0.12^{c}	13.63±0.12^{b}	13.63±0.12^{b}	10.70±0.10^{c}	11.30±0.10^{b}
2	22.00±0.00^{a}	20.10±0.23^{b}	17.10±0.12^{b}	14.43±0.12^{b}	14.43±0.12^{b}	12.03±0.06^{b}	11.96±0.06^{a}
3	22.00±0.00^{a}	20.77±0.17^{a}	17.77±0.06^{a}	15.20±0.00^{a}	15.20±0.00^{a}	12.67±0.06^{a}	12.67±0.06^{a}
4	22.00±0.00^{a}	21.10±0.10^{a}	18.53±0.10^{a}	15.37±0.06^{a}	15.37±0.06^{a}	13.17±0.06^{a}	13.03±0.06^{a}
5	22.00±0.00^{a}	21.80±0.06^{a}	18.53±0.06^{a}	15.53±0.06^{a}	15.53±0.06^{a}	13.63±0.06^{a}	12.23±0.06^{a}
Control	22.00±0.00^{a}	18.17±0.29^{c}	17.27±0.06^{b}	12.10±0.10^{c}	11.20±0.17^{c}	10.40±0.10^{c}	10.07±0.06^{c}

Means with similar letters along the column are not significantly ($P > 0.05$) different.

Essential oil treated samples

Percentage weight loss of samples treated with essential oil of *P. guineense* was lower than the control (Figure 1). Loss in weight progressively increased with storage period. When tomato fruit were refrigerated for 30 days, oil-treated samples had less weight loss than the untreated control. Hassani et al. (2012) reported the potential of essential oils in weight loss reduction could be attributed to the oily characteristics which covers the fruit surface and reduces respiration.

Effect of essential oil of *P. guineense* on microbial load of tomato fruits during storage

The results obtained for total viable count and total mould count are shown in Figures 2 and 3. The essential oil of *P. guineense* demonstrated antimicrobial property. The dosage of 10 µL of essential oil of *P. guineense* per 200 g tomato fruits significantly ($P < 0.05$) inhibited the growth of microorganism. Lowest counts were recorded in day 25. The role of monoterpenes in disturbing the membrane functions of yeasts has been reported by Adegoke et al. (2000) and synergistic effects of essential oil components were established (Parveen et al., 2004).

Abd Alla et al. (2008) reported that cinnamaldehyde vapour used in the packaging of apricot reduced fungal rot on inoculated fruits. Their observation supports the results obtained in this study as the colony forming ability of mould was reduced during storage of tomato fruits. Krisch et al. (2011) observed that essential oils can be used to prolong shelf life of fruits in vapour phase in active packaging or as coating on the surfaces of fruits.

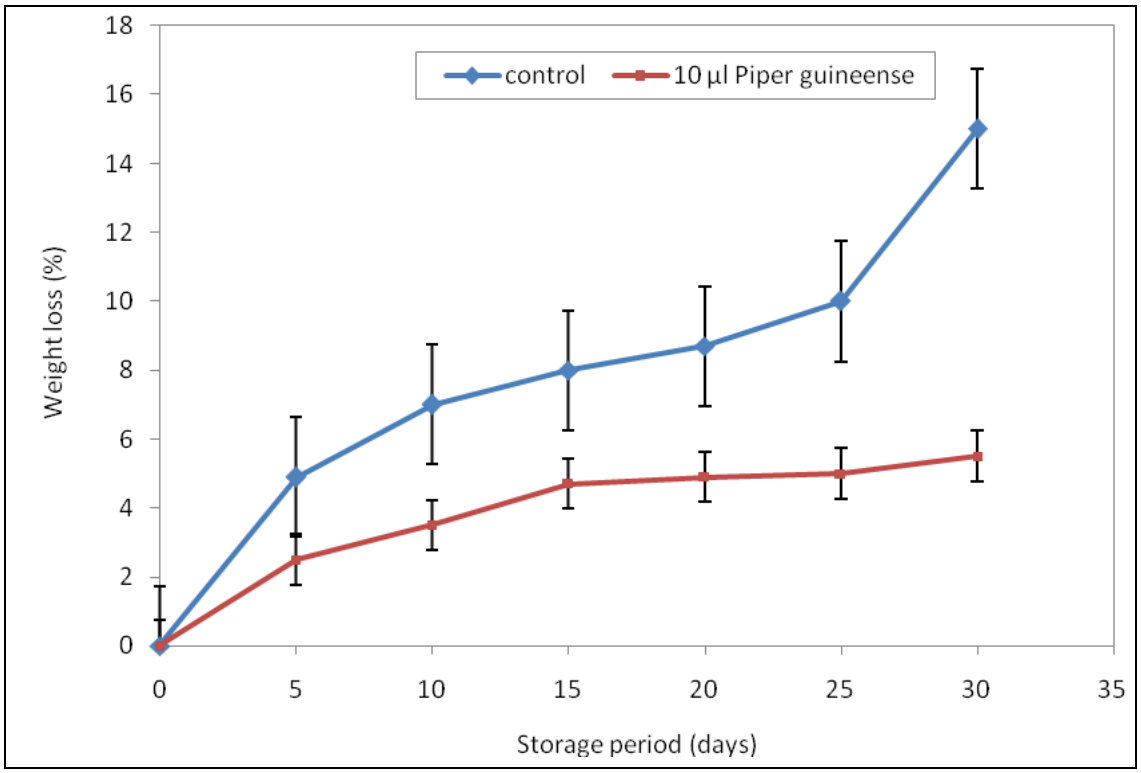

Figure 1. Weight loss of tomato fruits treated with essential oil of *P. guineense* stored at refrigeration condition.

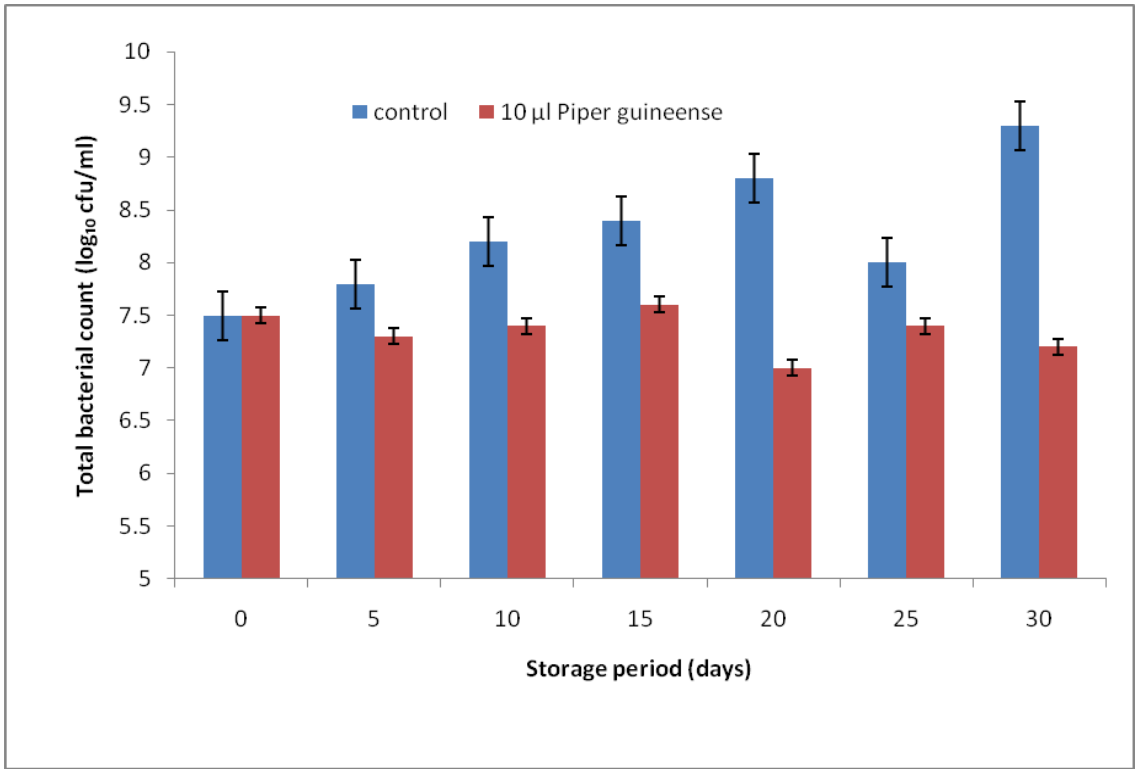

Figure 2. Total bacterial counts of tomato fruits treated with essential oil of *Piper guineense* stored at refrigeration condition.

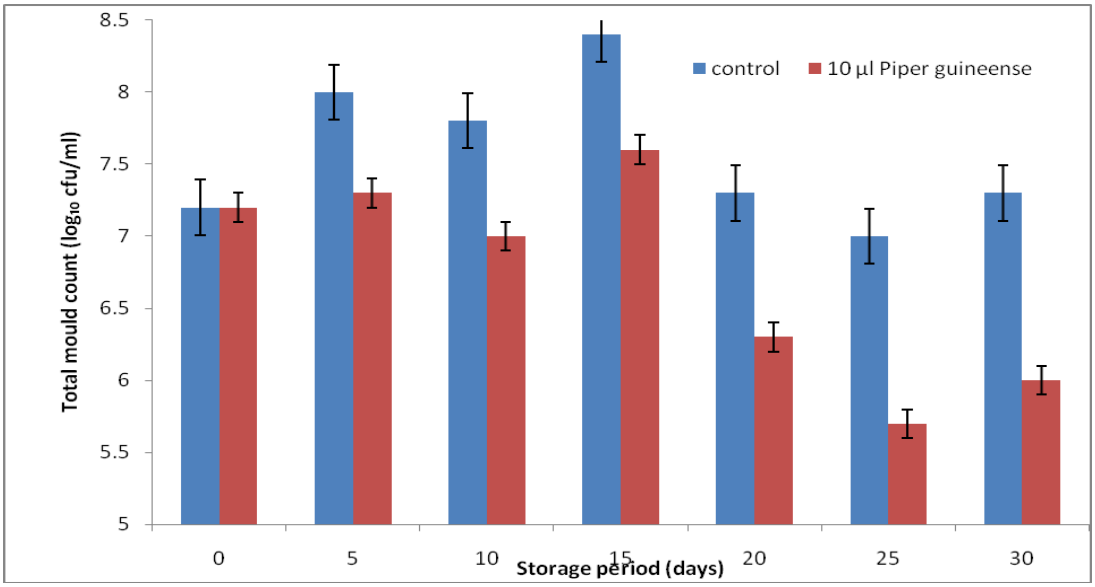

Figure 3. Total mould count of tomato fruits treated with essential oil of *Piper guineense* stored at refrigeration condition.

Chemical composition of the essential oil of *P. guineense*

Twelve chemical components were identified in *P. guineense* essential oil (Table 4). Sesquiterpene hydrocarbons were predominant, followed by oxygenated monoterpenes and monoterpene hydrocarbons. The most abundant constituents of *P. guineense* were β-sesquiphellandrene, 1,5-heptadiene, β-bisabolene, 6-methyl-2-(4-methyl-3-cyclohexene-1-yl), β-caryophyllene, linalool, zingiberene, β-farnesene and Guaia-1(5), 11-diene. Minor constituents were β-pinene, limonene, 2, 4 diisopropenyl-1-methyl-1-vinylcyclohexane, α-cubebene and α-caryophyllene. Martins et al. (1998) analyzed the essential oil of *P. guineense* and reported that the oil was characterized with 20.5% monoterpenes, 5.5% oxygenated monoterpenes, 11.6% sesquiterpenes and 2.1% oxygenated sesquiterpene.

Table 4. Chemical composition of the essential oil of *P. guineense*.

	Component[a]	RI[b]	% composition[c]
1	Beta-pinene	943	1.52
2	Limonene	1018	1.73
3	Linalool	1082	10.70
4	Alpha-cubebene	1344	1.75
5	2, 4 diisopropenyl-1-methyl-1-vinylcyclohexane	1398	3.36
6	Beta caryophyllene	1494	12.63
7	Beta farnesene	1440	8.13
8	Alpha caryophyllene	1579	3.18
9	Guaia-1(5), 11-diene	1490	6.91
10	Zingiberene	1451	9.98
11	1,5-heptadiene, 6-methyl-2-(4-methyl-3-cyclohexene-1-yl)	1500	16.37
12	Beta sesquiphellandrene	1446	23.74

[a]Components identified by comparing MS, with NIST (2005) library spectra software.
[b]Kovats retention indices relative to n-alkanes (C8-C22) on fused silica capillary column Rtx 5MS.
[c]% peak area relative to total peak area obtained from TIC total peak report.

Phenyl propanoid derivatives were the most important group of components in the oil of *P. guineense* identified with dillapiole (44.8%) being the main constituent, followed by myristicin (9.8%). Jirovetz et al. (2002) reported that the essential oil of *P. guineense* contained (black)-β-caryophyllene (57.59%), β-elemene (5.10%), bicyclogermacrene (5.05%) and α-humulene (4.86%); and *P. guineense* (white)-β-caryophyllene (51.75%), *cis*-b-ocimene (6.61%), limonene (5.88%), β-pinene (4.56%), linalool (3.97%) and α-humulene (3.29%).

Conclusion

Postharvest treatment of tomato fruits with *P. guineense* extracts reduced weight loss, inhibited growth of micro organism and retained ascorbic acid better than the untreated sample. The essential oil of *P. guineense* possesses preservative properties and can be used in post-harvest storage of tomato.

Conflict of Interest

The authors declare no conflict of interest.

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