Effects of Pre-harvest Application of Hexanal Formulation on Losses and Quality of Tomato (*Solanum lycopersicum* Mill)

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Abstract

The study was conducted to determine the effects of field application of enhanced freshness formulation (EFF) on pre-harvest losses and tomato quality. The parameters assessed include pest defects on fruits, marketable and percentage non-marketable fruit, fruit firmness, and fruit weight. Three popular open pollinated varieties grown in Tanzania namely, Mwanga, Rio Grande, and Tanya were used. The experiment was laid out as Completely Randomized Design in a 4 x 4 x 3 factorial arrangement. Three factors, EFF concentrations, time of EFF application prior to the harvest, and tomato variety were evaluated. EFF concentrations of 0.01, 0.02, 0.04% m/v were tested. Untreated plots were included as control. The time of application was 7, 14, 21, and 28 days prior to the harvest. The results showed that pre-harvest application of EFF at 0.01 percent reduced percent non-marketable tomato fruit of Mwanga, Rio Grande, and Tanya cultivars by 28.99, 26.98 and 37.17 percent, respectively compared with the control. Moreover, pest defects were reduced by 29.45, 24.51, and 27.45 percent for Mwanga, Rio Grande, and Tanya, respectively over the control. Furthermore, fruit firmness was increased by 7.69 N/mm^2, 6.33 N/mm^2 and 5.98 N/mm^2 compared with the control for tomato cv. Mwanga, Rio Grande, and Tanya, respectively.

Keyword: Hexanal, Non-marketable, Pest defects, Firmness, Tomato

Introduction

Tomato (*Solanum lycopersicum* Mill.) is one of the most important vegetables grown in Tanzania (Mushobozi, 2010) with a total annual production of more than 962 684 tons from an area of 26 612 ha (MMA, 2017). Productivity of tomato ranges from 2.2 t/ha to 3.3 t/ha for small-scale farmers in Eastern zone of Tanzania (Minja *et al.,* 2011). Tomato production in Tanzania accounts for 51 percent of the total fruit vegetables produced in the country (NBS, 2008). Tomato provides income to growers; and it can be eaten either fresh or processed in different products (Ahmad *et al.,* 2007). Nutritional value of tomato has made it one of the most popular vegetable crops (BCSL, 2009). Tomato is a good source of vitamin A, B, and C as well as iron and phosphorus (Yilmaz, 2001; Sowley and Damba, 2013). However, the productivity of tomato in Tanzania is below the world's average of 37.60 t ha^{-1} (FAO, 2017).The major causes of low productivity include fruit defects due to competition for photo-assimilates and unfavourable abiotic conditions especially drought stress, and declining soil fertility. In addition, high incidences of diseases such as late blight, powdery mildew and anthracnose, and insect pests such as mealy bugs, aphids, whiteflies, and the highly devastating tomato leaf miner *(Tuta absoluta)* contributes to low tomato yields (Roberts *et al.,* 2002; Normand *et al.,* 2009; Roemer, 2011). Tomato yield losses due to insect pests and diseases during the rainy season range from 80 to 100 percent (BCSL, 2009).

High yielding, good firmness, medium fruit size, and pest resistance are the most important characteristics for grower's selection of a tomato variety to use (Barickman *et al.,* 2017). Flower and fruit drops are serious constraints in tomato production especially during hot weather; while, diseases and insect pests are the major constraints affecting yield and quality, especially during the rainy season (Asgedom *et al.,* 2011). The most popular techniques of reducing flower and fruit drops and pre-harvest fruit defects include fungicide application prior to flowering, irrigation and fertilizer application during flowering and fruit development (Mertely *et al.,* 2002; Bulletin, 2009). As reported by Hao and Paradopoulos (2003), application of calcium and magnesium fertilizers increases tomato fruit size, firmness, and marketability of yields. However, the application of fungicides in controlling pre-harvest fruit losses is limited by consumers' desire for the reduced fungicide residues in fruit (Song *et al.,* 2010; Moser *et al.,* 2011; McCluskey, 2015).

Hexanal (molecular formula $C_6H_{12}O$) is an alkyl aldehyde and a strong inhibitor of activity of phospholipase-D enzyme, which slows down ethylene-stimulated ripening processes (Subramanian *et al.,* 2014; Karthika *et al.,* 2015). The application of EFF is a relatively new technology, which has shown to be effective in reducing pest defects, extending shelf life, and increasing fruit quality (Cheema *et al.,* 2014). Hexanal, an inhibitor of phospholipase D, has been used for pre-harvest treatment of fruit and vegetables. Phospholipase D is a key enzyme involved in membrane deterioration that occurs during fruit ripening and senescence (Cheema *et al.,* 2014). Field application of EFF has been reported to be among the most effective in increasing fruit firmness, freshness and fruit retention on trees in several fruit species including apple, cherry, longan, guava, and mango (Subramanian *et al.,* 2014; Karthika *et al.,* 2015). Moreover, field application of EFF was earlier reported to reduce premature fruit drop, insect pest, and disease infections in mangoes, strawberry, apple, cherry, guava, and tomato (Subramanian *et al.,* 2014; Karthika *et al.,* 2015). However, there is limited information on the effect of field application of EFF on the pre-harvest defects on fruit, percentage of non-marketable fruit, firmness, and weight of tomato.

Materials and Methods

Description of study area and tomato varieties

Studies were carried out at Horticulture Unit (6°50'41.478"S and 37°39'43.476"E, 523.40 m a.s.l.) of Sokoine University of Agriculture (SUA) in Morogoro Region from October to December 2017. The site has two rainy seasons with short rainy season from October to January and long rainy season from March to May. The annual precipitation ranges from 700 to 2 300 mm and temperatures range from 18 to 30°C (URT, 2016).

Three tomato varieties were selected for the study namely, 'Mwanga', 'Rio Grande', and 'Tanya'. Rio Grande is an early maturing variety (75 – 85 days), which produces high yield with good fruit retention. The variety is known to withstand poor transport conditions (Ahmad *et al.,* 2007; Jonathan, 2017). Tanya variety is also characterized by early maturity, resistance to brown rot and verticillium wilt diseases, and tolerant to bumpy road transportation (NTIF, 2018). Mwanga variety is early maturing, high yielding but susceptible to early blight disease and insect pests (Testen *et al.,* 2016).

Experimental design

The experiments were laid out as a Completely Randomized Design in a 4 x 4 x 3 factorial arrangement. It comprised three main factors: EFF concentrations (0.01, 0.02, 0.04% m/v and control - untreated fruit), time of EFF application prior to fruit harvest (7, 14, 21 and 28 days), and three tomato varieties. A plot with 11 tomato plants was taken as a treatment for EFF and its time of its application prior to fruit harvest. The experiment was replicated three times. The space between plots was 1 m while replications were separated by 1.5 m. The plot size was 2.1 m x 2.8 m with 11 plants. Seedlings were raised in the seedling trays and transplanted three weeks after sowing. Standard management practices of gap filling, application of fertilizers (DAP at planting and UREA four weeks after planting with the dosage of 5 mg per plant), and weeding were followed. EFF was sprayed using a knapsack sprayer on tomato fruit until the solution dripped off.

Composition for pre-harvest sprays

The solution compositions known as enhanced freshness formulation (EFF) contain ethanol 95percent, Tween® 20 (P1379, Sigma-Aldrich) and hexanal (115606, Sigma-Aldrich) (Paliyath *et al.,* 2003). The volumes for ethanol 95 percent and Tween® 20 (used as a surfactant) were constant (100 ml). The volumes of hexanal used were 5 mls, 10 mls, and 20 mls. The obtained EFF mixtures were diluted with 50 l of distilled water to make EFF concentrations of 0.01, 0.02, and 0.04 percent, respectively. Ethanol was pure without preservatives. Ethanol, which is an emulsifier, was mixed with Tween® 20 in a suitable container by stirring. Hexanal was then added to this mixture and stirred. The mixture was stored in a glass bottle in the dark.

Data collection and analysis

Tomato fruit were all harvested at a ripening stage. Data were collected immediately after the fruit harvest (with 12 ripening weeks after transplanting) based on incidences of pest defects on fruit peel, the number of non-marketable fruit yield, fruit firmness, and fruit weight.

Marketable and non-marketable fruit from each plot were sorted and counted. The percentage of non marketable yield was expressed as the proportion of harvested fruit with pest defects and other disorders (such as bruises and scars) per plot. Incidences of pest defects (OECD, 2010) was expressed as the percentage of harvested fruit with pest defect per plot. The major causes of defects were diseases (early and late bright and stem cancer) and insect pests (*T. absoluta* and *aphids Aphis* spp.). All ripened tomatoes from the treatment were harvested and put together at the cool place, and then fifteen tomato fruit from each treatment were randomly picked by considering the size (5 = small size, 5 = medium size and 5 = larger size) and used to determine fruit firmness and fruit weight. Fruit firmness was measured using a hand penetrometer with a probe diameter of 8 mm (Wagner instruments-Greenwich CT). Fruit weight was measured using a digital vernier balance (Kenwood Weighing Scales DS400) and fruit diameter was measured using digital caliper (New Type LCD Reading Long Jaw Internal Diameter Digital Vernier Callipers). Three way ANOVA was used to analyse data using R statistical package version 3:3:2 2016 (R CoreTeam, 2013); and post- hoc separated of treatment means was based on Tukey test at 5% probability.

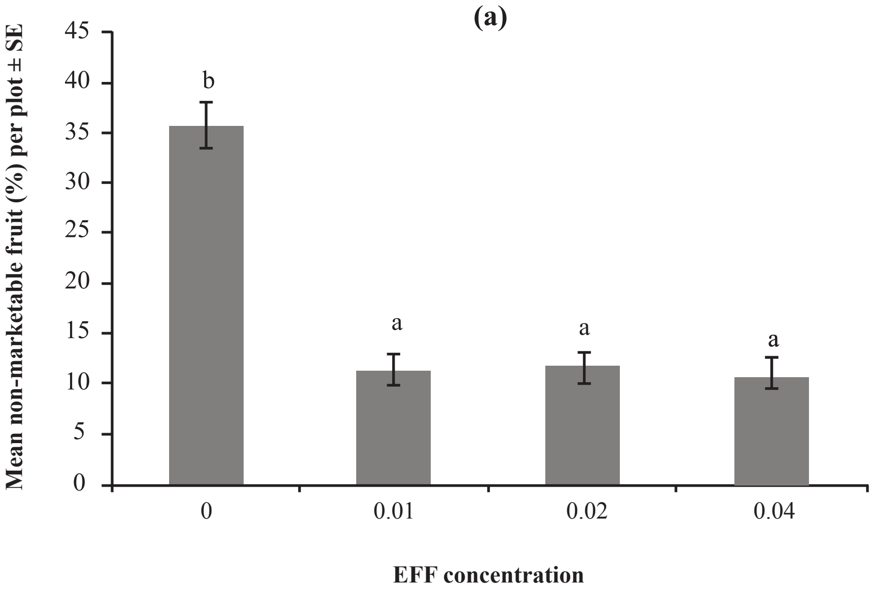
Results

The effects of EFF, time of application and variety on percentage of non- marketable fruit of tomato

The results showed that EFF and variety ($p<0.001$) reduced significantly the percentage of non-marketable fruit of Mwanga, Rio Grande, and Tanya tomato varieties. However, the results showed that the interactions between EFF x time ($p<0.842$), EFF × variety ($p<0.594$), time × variety ($p<0.455$) as well as EFF × time × variety ($p<0.781$) had no significant effects on percentage of non-marketable fruit. These results imply that, the efficacy of EFF concerning percentage of non-marketable fruit was not affected by variety or time of application. The examination of the main significant effects showed that, generally, EFF concentrations lowered percentage of non-marketable fruit up to 10.78 percent as compared to the control with 35.74 percent (Fig. 1a). However, there were no significant differences among the three EFF concentrations. Further analysis of the main effects also showed that, Tanya variety had higher non-marketable fruit than had Mwanga and Rio Grande varieties (Figure 1b). Generally, the number of non-marketable fruit of Mwanga, Rio Grande, and Tanya varieties was reduced by 28.99, 26.98, and 37.17 percent respectively compared to the control. The percentage of non-marketable fruit ranged from 20.77 percent in Tanya variety to 14.93 percent in Rio Grande variety (Fig. 1b).

Effects of EFF, time of application and variety on incidences of pest defects on tomato fruit

The results show that EFF reduced significantly ($p<0.001$) incidences of pest defects on fruit of the three varieties compared to the control. There were no significant effects on time of application ($p<0.586$) and the interactions of EFF × time ($p<0.889$), EFF × variety ($p<0.962$), time × variety ($p<0.677$),



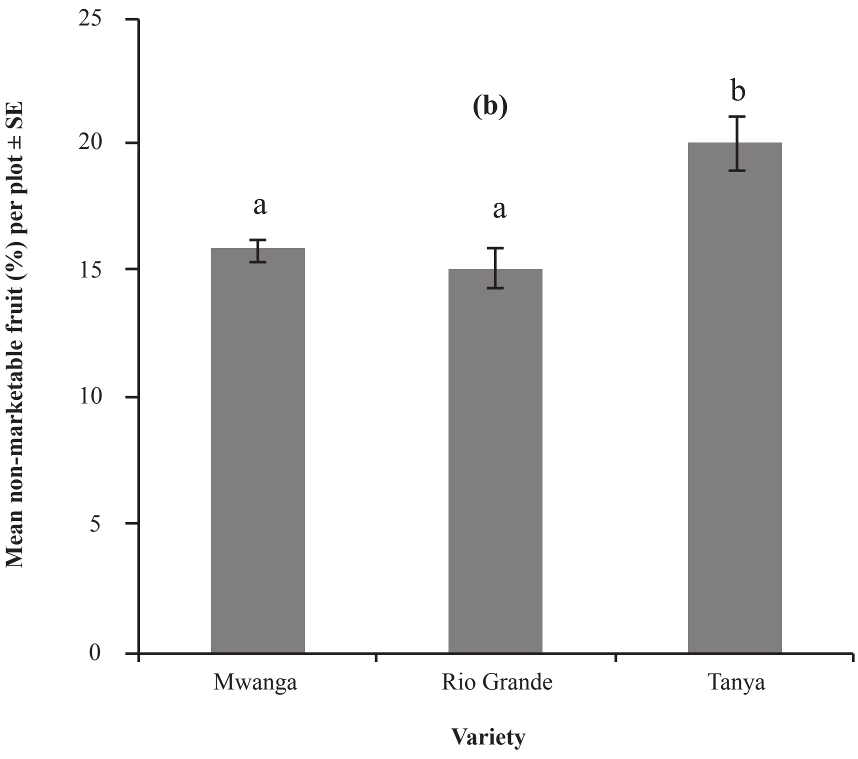


Figure 1: Effects of (a) EFF (b) variety, on percentage of non-marketable fruits of tomato fruit Post Hoc Tukey test = HSD $_{(p=0.05)}$. Error bars represents ± 5 % standard error around sample means.

and EFF × time × variety (p<0.817) on the incidences of pests on three tested varieties. Time of application and variety did no influence efficacy of EFF concentration in reducing pest defects. The analysis of the main effects showed that EFF lowered incidences of pest defects by up to 8.33 percent compared to the control with 30.74 percent. Incidences of pest defects did not differ significantly among fruit treated with the three EFF. Incidences of pest-inflicted defects ranged from 30.74 percent for untreated fruit to 8.33 percent when fruit were exposed to 0.04 percent of EFF (Fig. 2).

Effects of EFF, time of application and variety on tomato fruit firmness

The effects of EFF on fruit firmness was significantly ($p<0.001$) higher compared to that of the controls. However, effects of time of EFF application ($p<0.678$) and EFF × time ($p<0.710$), EFF × variety ($p<0.142$), time × variety ($p<0.549$), and EFF × time × variety ($p<0.521$) were observed to have no significant effects. This implies that the efficacy of EFF on improving firmness did not change with variety or time of application. The examination of the main means revealed that fruit treated with EFF had firmness of up to 9.61 N/mm^2 compared

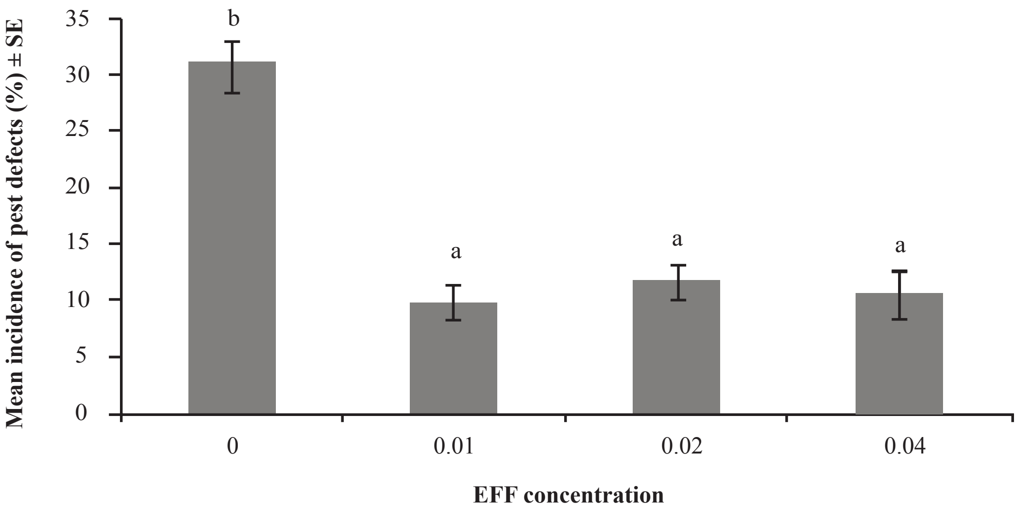


Figure 2: Effects of EFF on incidence of pest defects on tomato fruit. Post Hoc Tukey test = HSD $_{(p=0.05)}$. Error bars represents ± 5 % standard error around sample means.

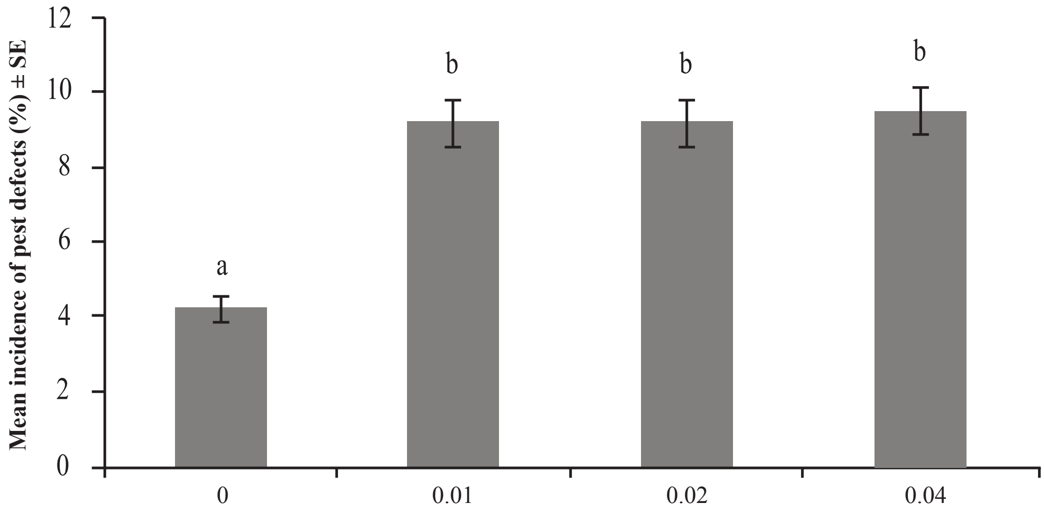


Figure 3: Effect of EFF on firmness of tomato fruit. Post Hoc Tukey test = HSD $_{(p=0.05)}$. Error bars represents ± 5 % standard error around sample means.

to the control fruit with 4.27 N/mm^2 (Fig. 3). However, there were no significant differences among the three EFF concentrations. Generally, EFF concentration of 0.01 percent increased fruit firmness significantly on Mwanga, Rio Grande, and Tanya varieties by 7.69 N/mm^2, 6.33 N/mm^2 and 5.98 N/mm^2 respectively compared to the untreated controls. Firmness ranged from 0.36 N/mm^2 for untreated fruit to 9.61 N/mm^2 for fruit exposed to 0.04 percent EFF concentration (Fig. 3).

Effects of EFF, time of application and variety on fruit weight of tomato varieties

The results showed that EFF, time of its application prior to fruit harvest, variety, and

Table 1: Mean weight of tomato fruit as affected by EFF, time of applications, and variety

Application time (Days) (%)	Hexanal concentration	Mwanga (g)	Rio Grande (g)	Tanya (g)
28	0.00	81.68a	68.81a	72.81a
	0.01	77.79a	81.10a	69.97a
	0.02	82.87a	79.81a	61.39a
	0.04	75.01a	76.13a	59.91a
21	0.00	77.25a	79.35a	74.12a
	0.01	70.58a	72.99a	85.13a
	0.02	79.75a	81.62a	84.12a
	0.04	81.44a	82.14a	73.21a
14	0.00	85.48a	81.82a	71.38a
	0.01	81.28a	73.15a	71.36a
	0.02	91.91a	78.88a	81.23a
	0.04	82.61a	85.30a	80.78a
7	0.00	79.37a	83.13a	68.17a
	0.01	86.11a	75.17a	69.29a
	0.02	62.05a	82.98a	81.42a
	0.04	82.72a	84.10a	85.65a
Application time (Main effects)				
28		79.34a	76.47a	66.02a
21		77.25a	79.03a	79.15a
14		85.32a	79.79a	76.19a
7		77.56a	81.34a	76.13a
	Hexanal conc. (Main effects)			
	0.00	80.94a	78.28a	71.62a
	0.01	78.94a	75.60a	73.94a
	0.02	79.14a	80.82a	77.04a
	0.04	80.45a	81.92a	74.89a

Note: Means in the same column bearing the same letter(s) are not significant different (Tukey HSD). C = Concentration, T = Time, V = Variety.

all interactions had no significant (p>0.05) effects on fruit weight of the tested varieties. Fruit weight ranged from 62.05 to 91.91 g for Mwanga, 68.81 to 85.30 g for Rio Grande, and 59.91 to 85.65 g for Tanya (Table 1).

Discussion

The present study found that application of various concentrations of EFF reduced significantly pest caused defects on tomato fruit. However, time of EFF application had no significant effects on pest-inflicted defects. The application of EFF was found to reduce incidences of early blight *(Alternaria solani)*, late blight *(Phytophthora infestans)*, stem canker diseases *(Clavibacter michiganensis)*, tomato leafminer *(Tuta absoluta)*, and aphids (*Aphis* spp) (IRAC, 2011). Physiological disorders that included blossom end rot and cracking were similarly reduced. These results are supported by the results of previous studies where pre-harvest EFF application decreased the incidences of pest inflicted defects on mango, apple, and pear fruit (Sholberg and Randall, 2007; Karthika *et al.,* 2015; Anusuya *et al.,* 2016; Bojan *et al.* (2016). Studies by Anusuya *et al.* (2016) and Bojan *et al.,* 2016 reported that EFF reduced the incidences of anthracnose stem end rot, mould, and bacteria in mango varieties. EFF was also found to reduce blue and gray molds in peach and fruit lesion in apple (Sholberg and Randall, 2007).

EFF improves the activity of defence related enzymes such as peroxide, polyphenol oxidase, phenylalanine ammonia-lyase, superoxide dismutase, and catalase in fruit against pathogens (Seethapathy *et al.,* 2016). According to Sholberg and Randall (2007), EFF can exhibit antifungal properties by altering the lipoxygenase pathway. Lipoxygenase are key enzymes that play an important role in the response of plants to wounding and pathogen attack (Gobel *et al.,* 2001). Aldehydes including hexanal derived from the lipoxygenase pathway induce a subset of defence related genes (Kuo and Gardner, 2005). In addition, Cheema *et al.* (2014) reported that EFF strengthens the fruit cell wall structures of tomato, thus drastically reduces the pathogen penetration and infections (Ahemad and Kibret, 2014; Wang *et al.,* 2014). These results also show that pre-harvest application of EFF at 0.01 percent increased firmness of treated tomato fruit compared to the control while time of EFF application had insignificant effects. Sharma *et al.* (2010) reported increased fruit firmness and shelf life of cherry fruit after pre-harvest EFF application. Similarly, EFF increased firmness of apple and pears (Sholberg and Randall, 2007), peach (Shen *et al.,* 2014), and mango fruit (Anusuya *et al.,* 2016). EFF application increases fruit firmness by strengthening cell wall structures of the fruit (Ahemad and Kibret, 2014; Wang *et al.,* 2014). EFF inhibits phospholipase D (PLD) enzyme which is responsible for breaking of the cell membranes during fruit ripening process (Karthika *et al.* 2015; Kumar *et al.* (2018) thus ensuring membrane integrity and enhancing fruit firmness (El Kayal *et al.,* 2017). Fruit firmness enhances fruit textural quality, organoleptic taste, and longevity after harvest (En-Tai *et al.,* 2014).

EFF had no significant effects on fruit weight of the tomato varieties. Similar findings were reported by Anusuya *et al.,* (2016) and Shen *et al.* (2014) who revealed that, fruit weight of mango and peach at harvest from sprayed and unsprayed fruit trees did not differ at harvesting stage. Fruit naturally increase in weight, volume, and length from fertilization to maturity. Furthermore, the interdependence between development and fruit growth shows up in the final stage as the fruit becomes mature prior to ripening. EFF did not influence these physiological processes that affected size, weight, and volume.

Conclusions and Recommendations

The objective of this study was to determine the effects of application of EFF on pre-harvest tomato losses and fruit quality. Field application of EFF at 0.01 percent remarkably improved fruit firmness and pre-harvest tomato marketable yield by reducing pest caused defects and total non-marketable yield for the three tested tomato varieties. Thus, application of the chemical at the concentration of 0.01 percent prior to fruit harvest is recommended to farmers as a technique for reducing pre-harvest losses and increasing harvested tomato fruit firmness.

These preliminary results need further support from trials across locations and seasons.

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