Pre-harvest field application of enhanced freshness formulation reduces yield loss in orange

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Abstract
Background: Orange (Citrus sinensis L.) production in Tanzania is constrained by several pre-harvest factors that include pests. Hexanal, sprayed as Enhanced Freshness Formulation (EFF) is a relatively new technology that has been reported to reduce pre-harvest loss in fruits. However, the effects of hexanal on pre-harvest yield loss of orange are not known. We studied the effects of hexanal as EFF on yield losses of three sweet orange cultivars namely, Early Valencia, Jaffa, and Late Valencia. Factorial experiments tested the effects of EFF concentration, variety, and time of EFF application on number of dropped fruit, percentage of non-marketable fruit and incidence of pest damage.

Results: Results showed significant negative correlation \((p < 0.001)\) between EFF and the percentage of dropped fruit, non-marketable yield, and incidence of pest damage. An increase in hexanal concentration by 1%, is expected to reduce number of dropped fruit by 50, percentage of non-marketable by 35.6, and incidences of pest damage by 36.5% keeping other factors constant. Results also show significant association \((p < 0.001)\) between time of hexanal application and non-marketable yield. Percentage of dropped fruit is expected to increase by 1 for each day away from harvest, keeping other factors constant.

Conclusion: Pre-harvest application of hexanal as EFF can significantly reduce number of dropped fruits, percentage of non-marketable fruit and incidence of pest damage.

Keywords: Early valencia, Jaffa, Late valencia, Non-marketable fruit, Pre-harvest fruit drop, Citrus sinensis

Background
Orange (Citrus sinensis L.) is one of the most important fruit crops due to distinct flavours, therapeutic, and economic values of its fruit [1]. Orange production in Tanzania is 249 641 mt [2] from an area of 42 335 ha with an annual productivity of 4.67 t ha\(^{-1}\) [3]. Orange production is constrained by several factors including pre-harvest fruit drops, incidence of serious diseases such as powdery mildew and anthracnose, and insect pests such as hopper and mealy bugs [4, 5].

Pre-harvest fruit drop is a major cause of low productivity of orange fruit worldwide [6–8]. Orange trees bear many fruits but most of them drop at early stages of development or before attaining the commercial ripening stage [9, 10]. Several techniques have been reported to reduce fruit drop and increase retention of fruit on orange trees [11–16]. For example, an aldehyde (\(n\)-hexanal and (\(E\))-2-hexanal) improves fruit retention on trees and fruit quality such as aroma, skin colour, and firmness [17, 18]. Similarly, auxin alleviates fruit abscission at post-bloom and early development stages of the fruit, which results in the reduction of fruit drop [11]. Moreover, 3, 5, 6-trichloro-2-pyridloxyacetic acid (3, 5, 6-TPA) was investigated to control fruit drop properties, fruit weight, diameter, length, and leaf/fruit

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ratio [19, 20], whereas the application of fungicides and a combination of 2,4-dichlorophenoxyacetic acid (2,4-D) and gibberellic acid (GA3) improves fruit retention on trees by reducing high flower and fruit drop [21]. A combination of urea and GA3 enhances fruiting and fruit quality, fruit set, and fruit retention on trees [16].

Pre-harvest application of hexanal as Enhanced Freshness Formulation (EFF) containing hexanal, ethanol and tween 20, was recently reported to be effective against premature fruit drop, superficial scald and fungal infection (18). EFF has also been reported to increase fruit firmness, quality, freshness, and fruit retention on trees of various fruit species such as apple, cherry, longan, mango, strawberry, guava, and tomato [17, 18, 22]. The effects of such pre-harvest applications on yield loss in orange is not well known. Therefore, the objective of this study was to determine the effects of pre-harvest application of hexanal on three important orange cultivars in Tanzania for the fruit retention and marketability.

Materials and methods

Description of study area

The study carried out at Semngano (254.0 m a.s.l., 05°14'14.8"S and 038°46'33.1"E) and Mamboleo (263.0 m a.s.l., 05°13'59.9"S and 038°42'58.2"E) villages in Muheza District, Tanga Region. These sites have the same agro-climatic conditions. Muheza district experiences bimodal rainfall from 800 to 1400 mm with an average annual minimum and maximum temperatures of 24 °C and 32 °C, respectively [23]. The long rainy season is between March and May, while the short rain season is between October and December. The experiments were carried out in semi-commercial farmers’ orange orchards (10–15 years old trees), which were well established and maintained according to the recommended agricultural practices.

Description of orange varieties

Three orange varieties namely Early Valencia, Jaffa, and Late Valencia were selected for the study. Early Valencia is the most popular variety with an extended production from May to September, high yield, and firm fruits that tolerate long distant transportation [24, 25]. Late Valencia is a popular variety, which matures from January to March, produces high yield, retains mature fruit on trees for an extended period, and has fruits that withstand tough transportation and environmental conditions [25]. Jaffa variety matures from May to July and produces high yield; however, its fruit do not withstand harsh transport conditions [24, 25]. Early Valencia and Late Valencia are the most preferred orange varieties in Muheza District, and whose farmers’ preference stands at 45.8 and 31%, respectively, of all the orange varieties grown in the district [26].

Design of experiment

The experiment was laid out in a randomized complete-block design in a 3 × 4 × 4 factorial arrangement. The first factor was orange variety (Early Valencia, Jaffa, and Late Valencia), the second factor was EFF concentration (0.01, 0.02, 0.04% of hexanal and untreated fruit/control), and the third factor was time of EFF application prior to fruit harvest, i.e. days to harvest (7, 21, 42 and 60 days). The experiment was repeated twice, from April 2017 to July 2017, and from August 2017 to December 2017. A single tree of each variety was considered as an experimental unit, which was replicated ten times. EFF was manually sprayed on orange fruits until dripping using a low-pressure knapsack sprayer. Each variety was sprayed according to its phenology. The EFF concentrate was prepared by mixing 100 ml of Ethanol (95%) with 100 ml of Tween 20 in a suitable container while stirring. Hexanal volumes of 5, 10, and 20 ml, were then added to the mixture separately. We diluted the mixture with water to make 50 l solution corresponding to 0.01, 0.02, and 0.04% of hexanal. The volume for the pre-harvest spray ranged from 1 to 3 l depending on the size of the tree and the number of fruits per tree.

Data collection and analysis

Fruits were harvested at maturity, based on commercially acceptable indicators of colour (yellow peel colour), size (minimum 53 mm) and shape (slight or no defect in shape) [24, 27]. Data were collected immediately after the harvest of the fruits on the total number of fruits, the number of non-marketable fruits and incidences of pest damage as described below. The dropped fruits per tree were collected and counted at an interval of 1 week from the 7th day after the application of EFF concentration and stopped just before the first fruits were harvested. The harvested fruits were sorted into marketable and non-marketable fruits per tree. According to OEDC [24], orange fruit with sunburn, stem end rot, anthracnose, bruising, scar, and powdery mildew infections were considered as non-marketable. Data on incidences of pest damage were obtained by sorting and counting fruits with pest defects. The major pest defects were caused by fruit flies, fruit Piercing moth, false codling moth, and anthracnose.

All data were averaged by replications and growing period. Data in percentage were transformed using
Results

Effects of EFF concentration on fruit drop

Our results show that independent variables EFF concentration, variety and time significantly predicted the fruit drop from trees of the three varieties \((F_{(4, 91)} = 9.24, p < .0001)\). EFF concentration has a significant inverse relationship with fruit drop (Table 1). An increase in EFF concentration by 1%, is expected to reduce number of dropped fruit by 326, keeping other factors constant. Furthermore, number of dropped fruit is expected to increase by 0.71 for each day away from harvest, keeping other factors constant. Varieties Jaffa and Late Valencia are expected to have lower numbers of dropped fruit by 1.80 and 2.95, respectively, compared with Early Valencia, although in both cases the correlations were not significant (Table 1).

Effects of EFF concentration on percentage of non-marketable fruit

Our results further show that independent variables EFF concentration, variety and time significantly predicted the percentage of non-marketable fruit \((F_{(4, 91)} = 4.69, p = .002)\). EFF concentration has a significant negative correlation with non-marketable fruit. An increase in EFF concentration by 1%, is likely to reduce percentage of non-marketable fruit by 36, keeping other factors constant. Varieties Jaffa and Late Valencia are expected to have lower percentages of non-marketable fruit by 0.14 and 0.54, respectively, compared with Early Valencia, although in both cases, the correlations were not significant (Table 2).

Effects of EFF concentration on incidence of pest damage

We also found that EFF concentration, variety and time significantly predicted incidence of pest damage on three orange varieties \((F_{(4, 91)} = 4.09, p = .004)\). Incidences of pest damage are likely to be lower by 36.5% for 1% increase in EFF concentration. Compared with Early Valencia, variety Jaffa is likely to have a lower incidence of pest damage by 0.26% while Late Valencia is expected to have a higher incidence by 0.22% (Table 3).

Discussion

In the present study, the application of hexanal significantly reduced the number of dropped fruits in all the three varieties of the orange tested, namely, Early Valencia, Jaffa, and Late Valencia. This clearly shows that EFF can help in fruit retention. Retaining fruits would be valuable to farmers as it can help them to extend the season, thereby stabilizing the price. Earlier studies have demonstrated that hexanal reduced fruit drop in mango, strawberry, raspberry, and nectarines [29–32]. It is hypothesized that the increased retention of fruits on the orange trees due to hexanal is associated with the delay in abscission [29]. It is not clear how hexanal prevents abscission in mature fruits. El Kayal et al. [31] reported that hexanal reduced the activities of phospholipase D (PLD) and abscisic acid (ABA) regulating genes in strawberry. Although we have not analysed these genes in oranges, it is quite possible that fruit retention in orange is also facilitated by the reduced levels of PLD and ABA, as reported previously [29, 32]. It has been shown further that hexanal also alters the expression of calcium regulating genes in raspberry [30]. Phospholipase D acts on phospholipids generating phosphatidic acid that undergo sequential catabolic breakdown downstream. Therefore,
once Phospholipase D is inhibited, the whole cycle is slowed down and this results in increased fruit retention on trees [17, 18]. Together these data suggest that a similar mechanism of delaying PLD, as well as other ripening related genes could have also resulted in the enhanced retention of fruits in orange.

Application of hexanal can also indirectly benefit orange growers, in addition to fruit retention. Reduction in fruit drop will be more beneficial in early maturing orange varieties, which are naturally prone to high fruit drops compared to the later maturing varieties. In addition to dropped and damaged fruits during harvest, orange encounters a number of other issues such as sooty mold and other disorders like cracking and blossom end rot, which makes them unmarketable [33]. Our study indicates that the application of hexanal reduced to incidences of green mold, sooty mold, and physiological disorders such as blossom end rot, which are all secondary benefits of applying hexanal. Earlier studies have also observed that hexanal reduced insect pest and disease damage on mango, apple, and pear [29, 34]. According to Sholberg and Randall [34], hexanal can exhibit antifungal properties by altering the lipoxygenase pathway. Lipoxygenases are key enzymes that play an important role in the response of plants to wounding and pathogen attack [35].

Conclusions

Results of this study show that hexanal can significantly reduce yield loss in three orange varieties. Hexanal consistently reduced number of dropped fruit, percentage of non-marketable fruit and incidence of pest damage. Although a detailed cost–benefit analysis is yet to be done for East African conditions, based on other studies in Asia and the Caribbean, it is expected to be very affordable to fruit farmers, considering the overall benefits. Nevertheless, application of hexanal can reduce unwanted loss in orange and thereby increase the returns to growers. We, therefore, recommend that a detailed economic and marketing analysis of hexanal on its cost–benefit ratio should be done in the near future.

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Authors’ contributions

JS did the research, collected data and wrote the first draft of the manuscript. TD interpreted the data of fruit drop and non-marketable fruits. AB had contribution in writing the background. HDM interpreted the data of incidences of pest defects. AN Contributed in writing the results. JAS contributed in writing the abstract and editing the manuscript. JS was the PI of the project and major contributor in writing the discussion. MWM was the major contributor in setting the layout and data analysis. All authors read and approved the final manuscript.

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Availability of data and materials

There are no linked research data sets for this submission. The following reason is given: the data used forms part of the thesis work of Jaspa Samwel–submitted to the Sokoine University of Agriculture, Tanzania.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Abbreviations

EFF: Enhanced freshness formulation; P: Probability; PO RALG: President’s Office Regional Administration and Local Government; 3, 5, 6-TPA: 3, 5, 6-trichloro-2-pyridyloxyacetic acid; 2, 4-D: 2, 4-Dichlorophenoxyacetic acid; GA3: Gibberellin; m.a.s.l: Meters above sea level; mm: Millimetre; °C: Degree centigrade; %: Percent; M: Millilitres; L: Litres; PLD: Phospholipase D; ABA: Abscisic acid; CIFSRF: Canadian International Food Security Research Fund; IDRC: International Development Research Centre; GAC: Global Affairs Canada; MMA: Match Maker Associates; NBS: National Bureau of Statistics; TRCO: Tanga Regional Commissioners Office; OECD: Organisation for Economic Co-operation and Development; DTH: Days to harvest.

Table 3 Effect of predictors on incidence of pest damage

| Term     | Coefficient | SE coefficient | T value | P value | VIF |
|----------|-------------|----------------|---------|---------|-----|
| Constant | 2.432       | 0.638          | 3.81    | 0.000   |     |
| DTH      | −0.162      | 0.175          | −0.93   | 0.356   | 1.00|
| Concentration | −36.56   | 9.91           | −3.69   | 0.000   | 1.00|
| Variety* | −0.260      | 0.352          | −0.74   | 0.462   | 1.33|
| Jaffa     | −0.221      | 0.352          | 0.63    | 0.532   | 1.33|
| Late Valencia | 0.221     | 0.352          |         |         |     |

DTH: days to harvest. VIF: Variance Inflation factor.

* Reference variety: Early Valencia

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