

Effect of Hexanal as a post-harvest treatment to extend the shelf-life of banana fruits

(*Musa acuminata* var. sweet banana) in Kenya

ABSTRACT

The short shelf-life of fruits in the tropics continues to be a pressing problem for farmers and other value chain actors. Hexanal is a naturally occurring compound that has received attention as a novel postharvest compound preservative. This study was conducted to determine the effect of hexanal on enhancing the postharvest shelf-life and quality of 'sweet banana' fruits. Two hexanal concentrations (2% and 3%) were applied as either a pre-harvest spray or a post-harvest dip. Fruits were obtained from two different agro ecological zones of Kenya (AEZs II and IV). The treated fruits were kept under ambient room conditions of $25 \pm 1^{\circ}\text{C}$ and $\text{RH } 60 \pm 5\%$ to ripen. Hexanal treatment maintained the fruits quality and prolonged the shelf-life by 6 days in the dipped fruits, 6 and 3 days in the sprayed fruits from the drier AEZ IV and colder AEZ II respectively compared to the untreated controls. Hexanal treatments significantly ($P = .05$) delayed or reduced the rate of most of the physicochemical parameters analysed irrespective of the concentration and mode of application used. Fruit firmness was significantly ($P = .05$) maintained up to day 6 and 9 of storage in the treated fruits compared to the controls which softened drastically as from day 3 and 6 in the sprayed and dipped fruits respectively. Hexanal treatment delayed ethylene and respiratory peaks by 3 days in both modes of application and significantly delayed progression of other ripening related changes such as $^{\circ}\text{Brix}$, titratable acidity, simple sugars and vitamin C. Sensory evaluation showed no significant differences in the various quality attributes analysed between the hexanal treated and control fruits. The results of this study indicate that, use of hexanal is a potential technology that could be adopted by banana farmers to enhance post-harvest shelf-life without compromising on quality.

Keywords: Hexanal, fruit quality, shelf life, postharvest loss, 'sweet banana'

1. INTRODUCTION

Kenya is endowed with good climatic conditions which favours production of different types of horticultural crops among them fruits, vegetables and cut flowers. Fruits are a key component of horticultural subsector in Kenya and come third in terms of income contribution after flowers and vegetables [11]. However, the full commercial potential of fruits such as banana has not been realized due to various challenges along the value chain among them high postharvest losses estimated at 40% [8]. The huge postharvest losses are mostly attributed to the highly perishable nature of the produce and further aggravated by failure to use appropriate post-harvest technologies.

In Kenya, banana is the most popular fruit crop often consumed as a dessert while the cooking varieties serve as a staple food in different regions of the country [11]. However, most of it is consumed locally with only a small percentage of approximately 7.2% being exported [11]. Production of banana is mostly dominated by small scale farmers though few medium and large scale growers are found in the major banana growing areas [8]. Banana is a security crop at the household level and the surplus is sold to provide the much-needed income for farmers. Nutritionally, banana contains high levels of calorie, a wide

39 range of vitamins, minerals, anti-oxidants and it is naturally low in fats [21]. However, once ripe the fruits
40 have a short shelf life of approximately 3-4 days and this limits their utilization, postharvest handling and
41 marketing [1].

42 Banana is a climacteric fruit which is often harvested at the physiological maturity stage and then ripened
43 before marketing. During ripening, the fruit undergoes different biochemical and physiological changes that
44 transforms the fruit to edible state. Some of these changes include fruit softening, changes in peel color,
45 degradation of starch to sugars, changes in concentration of aroma volatiles and acids. According to
46 Maduwanthi and Marapana [19], sugar levels increases from of an initial of 2% in green banana to
47 approximately 15% -20% in the ripe fruit making it sweeter. However, once the fruit is fully ripe, it becomes
48 very delicate and if not properly handled high postharvest losses can be incurred. In order to increase
49 storage life of the fruits, appropriate post-harvest technologies aimed at reducing the deterioration rate
50 have been developed over the years. These technologies are used to slow down fruits metabolic processes
51 to deliver enhanced shelf-life and optimal quality without compromising on the consumer safety. Recently,
52 efforts have been made to develop new and biological post-harvest technologies for extension of banana
53 shelf-life while retaining quality [33-35]. Use of hexanal and its formulations is one of the new innovations
54 which have been proved effective in enhancing the post-harvest shelf life of banana fruits [33, 34]. Hexanal,
55 is an aldehyde compound produced naturally by plants as a defence response to different biotic stresses
56 and has an odour similar to that of freshly cut grass or cucumber [18]. The United States Food and Drug
57 Administration Agency has approved the use of hexanal as a GRAS compound [22]. Hexanal use offers a
58 human-safe post-harvest preservation product that is environmentally friend and economically viable.
59 Hexanal is oxidized to
60 hexanoic acid in the body after consumption and further oxidized to carbon dioxide and water during respir
61 ation through the tricarboxylic acid cycle [18]. It has also been noted that hexanal, has antimicrobial
62 properties against several post-harvest pathogens such as *Alternaria alternate* and *Botrytis Cinerea* [29]. A
63 biochemical formulation of an artificially synthesized version of hexanal (Enhanced Freshness Formulation)
64 has been developed which delays fruit ripening [26]. This formulation can be applied in different ways such
65 post-harvest dip, pre-harvest spray or as a vapor. Being a relative new technology, there is need to test its
66 suitability to enhance banana shelf-life while preserving post-harvest quality in Kenya. A previous study in
67 'Grand naine' variety [34], showed promising results of hexanal extending fruits shelf life by nine days
68 without compromising on quality. However, since hexanal's effect is physiological, it is possible that its
69 efficacy might vary between varieties. The objective of this study was therefore, to determine the effect of
70 hexanal treatment on the post-harvest shelf-life and quality of 'sweet banana' fruits, a very popular variety
71 in Kenya.

72

73 2. MATERIALS AND METHODS

74 2.1. Study area

75 The experiment was conducted on 'sweet banana' fruits from two contrasting agro ecological zones (AEZs)
76 in Kenya. Meru County is a high potential AEZ II that lies at an elevation of 1980–2700 m above sea level
77 and receives an annual average rainfall of 1500 mm. Machakos County is a semi-arid AEZ IV that lies at an
78 elevation of 1000-1600 m above sea level with an annual average rainfall of 600 mm.

79 **2.2. Experimental setup**

80 For the pre-harvest spray mode of application, 15 banana trees at flowering stage in each study site were
81 randomly selected and tagged in the farmer's field. Two concentrations of hexanal (2% and 3%) and a
82 control (clean, plain water) were sprayed twice at 30 and 15 days before harvest. The dosing range used
83 was informed by a previous study done on 'Grand naine' variety [34]. Since hexanal is immiscible with
84 water, Tween 20 and ethanol were added to increase its solubility [22]. **Tween 20, ethanol and hexanal**
85 **were added in the ratio of 10:10:1.** The stock solutions were mixed with water and diluted accordingly to
86 provide the required hexanal concentrations. Using a knapsack sprayer, the fruits were sprayed to the point
87 of dripping with the solution. Spray contamination was avoided by using alternate rows of trees for the
88 experiment and a 4 tree gap between treatments in the same row of trees. The fruits were left on the tree
89 until approximately 20% per bunch had ripened. The fruits were then harvested and only the middle hands
90 were used in the post-harvest analysis.

91 For the post-harvest dip mode of application, fruits were harvested at the mature green stage based on
92 degree of fullness of the fingers, as indicated by the disappearance of angularity and the number of days
93 after anthesis which was approximately 104 days. Only the middle hands of each banana bunch (a cluster
94 of fruits attached together at the stalk) were used in the analysis. The harvested fruits were packed in
95 cushioned crates, covered with wet magazine papers to reduce water loss, and immediately transported to
96 the post-harvest laboratory at Jomo Kenyatta University of Agriculture and Technology (JKUAT).

97 **2.3. Sample preparation**

98 In the post-harvest laboratory, the fruits were cleaned, dried, and selected for uniformity and freedom from
99 mechanical injuries. Pre-harvest spray-treated fruits were left to undergo normal ripening under ambient
100 room conditions of $25 \pm 1^\circ\text{C}$ and $\text{RH } 60 \pm 5\%$. Fruits for post-harvest treatment were dipped in one of the
101 two hexanal concentrations (2%, 3%) or plain water (control) for 5 minutes. The hexanal solution was
102 mixed with Tween 20 and ethanol to increase its solubility. The hexanal concentrations and application time
103 used was informed by a previous study done on 'Grand naine' variety [34]. All the fruits were left to undergo
104 normal ripening under ambient room conditions. Five banana hands from each treatment combination were
105 randomly sampled at 3-day intervals to evaluate respiration and ethylene evolution rates. Three fruits were
106 also randomly sampled to evaluate other ripening related parameters including pulp firmness, °Brix,
107 titratable acidity, ascorbic acid, simple sugars and sensory analysis evaluation.

108 **2.3.1. Shelf life**

109 The time taken by the fruits from harvesting to reach the optimal, edible ripe stage was counted and
110 reported in days. This was defined as stage 7 according to the standard banana ripening chart by Soltani *et*
111 *al.* [27].

112 **2.3.2. Analysis of physiological parameters**

113 Gas chromatographs models GC-8A and GC-9A, Shimadzu Corp., Kyoto, Japan were used to determine
114 the rate of respiration and ethylene production respectively. The gas chromatograph to determine rate of
115 respiration was fitted with a thermal conductivity detector and a Poropak N column while that for ethylene
116 determination was fitted with an activated alumina column and a flame ionization detector. The rate of

117 carbon dioxide production (used to estimate respiration rate) was expressed as mL/Kg/h while ethylene
118 production was expressed as $\mu\text{L/Kg/h}$. Five fingers of bananas were randomly sampled from each
119 treatment, numbered and their weights taken using a digital balance, Model Libror AEG-220, Shimadzu
120 Corp. Kyoto, Japan. Each of the five fingers were incubated for two hours in air tight containers fitted with
121 self-sealing rubber septa. Gas samples were taken from the headspace using an airtight 1 mL hypodermic
122 syringe and injected into the respective gas chromatographs

123 **2.3.3. Pulp firmness**

124 Firmness was measured along the equatorial region of the fruit using a penetrometer (CR-100D, Sun
125 Scientific Co. Ltd, Japan) fitted with an 8 mm probe. Four locations along the equatorial zone of the fruit
126 were used and average value of firmness calculated. The banana was peeled first, before allowing the
127 probe to penetrate the flesh to a depth of 8mm and the corresponding force required to penetrate this depth
128 determined. Firmness was expressed as Newton.

129 **2.3.4. Analysis of fruit quality parameters**

130 Quality parameters such as Total soluble solids, Total titratable acidity, ascorbic acid and simple sugars
131 were determined using standard operational protocols in the same set of fruit that were used for the fruit
132 firmness analysis. Total soluble solids (TSS) content was determined using a digital refractometer (Model
133 PAL-1, Atago, Tokyo, Japan) and expressed as $^{\circ}\text{Brix}$. Total titratable acidity (TTA) was determined by
134 titration with 0.1N NaOH in the presence of phenolphthalein indicator and expressed as percentage malic
135 acid, the predominant organic acid in banana fruit.

136 Ascorbic acid content (Vitamin C) was determined by use of high performance liquid chromatography
137 (HPLC) method. About 5g of sample was weighed and extracted with 0.8% meta-phosphoric acid under
138 subdued light conditions. The extract was made to 20 mL of juice and centrifuged at 10000 rpm at 4°C for
139 10 minutes. The supernatant was filtered and diluted with 10 mL of 0.8% meta-phosphoric acid. This was
140 passed through 0.45 micro filters. The samples were then set as a post-run into HPLC machine (Model LC-
141 10AS, Shimadzu Corp., Kyoto, Japan) where 20 μL of the micro filtered sample was automatically injected
142 into the HPLC machine on the same day of extraction. Various concentrations of ascorbic acid standards
143 were prepared at 10, 20, 40, 60, 80 and 100 ppm and a blank containing only degassed meta-phosphoric
144 acid and used to obtain a calibration curve. HPLC analysis was done using Shimadzu UV-VIS detector
145 fitted with phenomenex 250mm*4.6mm*5 μL C-18 ODS column. The mobile phase was 0.8% meta-
146 phosphoric acid, at 1.2 mL/min flow rate and wavelength of 266.0 nm.

147 Simple sugars were analysed using a high performance liquid chromatography (HPLC) (Model LC-20AS,
148 Shimadzu Corp., Kyoto, Japan) fitted with a refractive index detector (RID). Approximately, 10g of the
149 banana pulp was completely blended and 96% ethanol added. Refluxing was done for one hour at 100°C
150 and then cooled under running water. The solution was then filtered using 42mm whatman filter paper.
151 Rising was done using 5ml of 96% ethanol. The solution was rotary evaporated to dryness at 60°C. 5ml of
152 50% acetonitrile was then added and finally micro-filtered (0.45 μm). The HPLC was running under the
153 following conditions: oven temperature: 30°C, Flow rate: 0.5-1.0 ml/min, Injection volume: 20 μL , Column:

154 NH_2 (5.0 μl) Mobile phase: Acetonitrile: water (75:25). Sugars present in the solution including sucrose,
155 glucose and fructose were identified and their individual concentration calculated using the standards.

156 Sensory quality evaluation was done on the hexanal treated and untreated fruits once they were fully ripe;
157 stage 6, standard banana ripening chart by Soltani *et al.* [27]. The fruits were washed with clean water,
158 dried and diced into approximately equal-sized slices, avoiding the extreme ends. Three slices were placed
159 on white plates which were anonymously coded based on treatment to ensure objectivity. A panel of 36
160 untrained judges drawn from student population of faculty of Agriculture, University of Nairobi was guided
161 on the scoring procedure of the various sensory attributes which included; fruit colour, aroma, texture,
162 flavour, mouth feel and the general acceptability. The panellists scored for these attributes on five point
163 hedonic scale where 1 = dislike (worst), 2 = (dislike moderately), 3 = (neither like nor dislike), 4 = (like
164 moderately) and 5= Like extremely (Best). This was adapted from Galan *et al.* [9], but with few
165 modifications.

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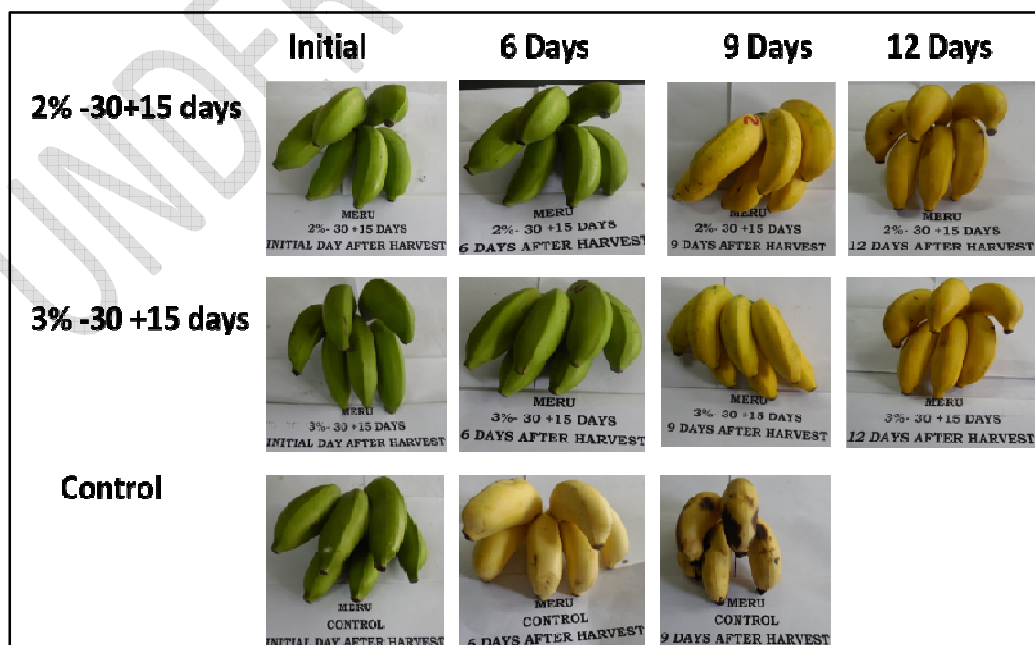
167 2.4 Statistical analysis

168 Data collected was subjected to analysis of variance (ANOVA) using Genstat statistical package (version
169 15). The means were separated by Least Significance Difference (LSD) at $p \leq .05$ using Fisher's protected
170 test. The sensory quality evaluation data was analyzed using Statistical Package for the Social Sciences
171 (SPSS) version 20.

172 3.0 Results

173 3.1. Shelf life

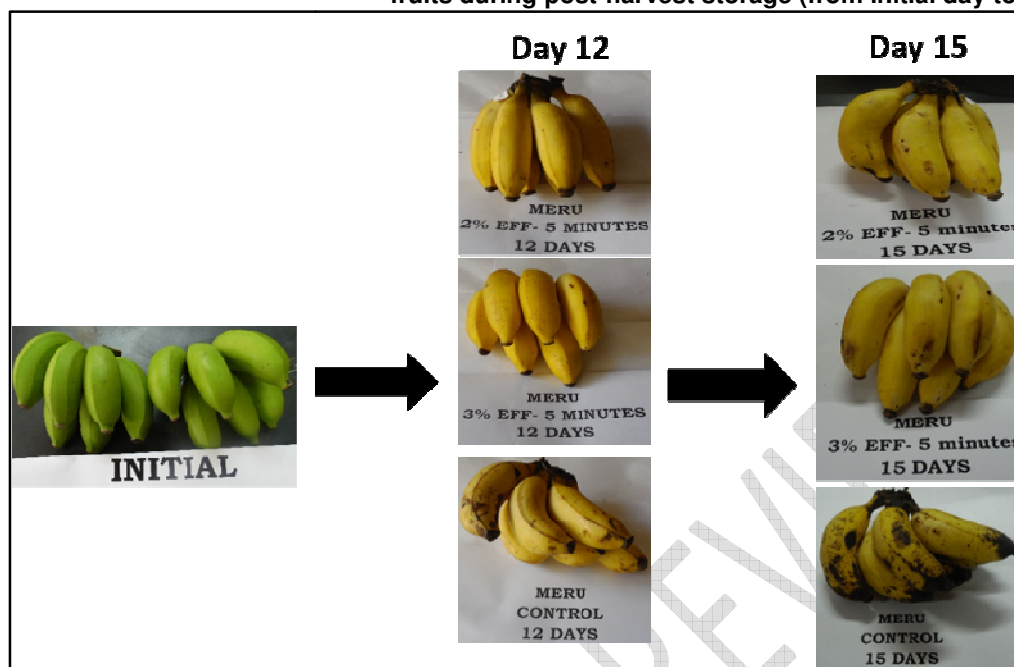
174 Hexanal treatment enhanced shelf life by 6 days (Plate 2) in the post-harvest dipped fruits, 6 and 3 days in
175 the sprayed fruits (Plate 1) from the drier AEZ IV and the wetter AEZ II respectively as compared to the
176 controls, irrespective of the production zone and hexanal concentration used.



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Plate 1: Ripening changes of 'sweet banana' fruits sprayed with 2% and 3% Hexanal and controls fruits during post-harvest storage (from initial day to day 12)



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Plate 2: Ripening changes of 'sweet banana dipped in 2% and 3% Hexanal for 5 minutes and controls fruits during post-harvest storage (from initial day to day 15)

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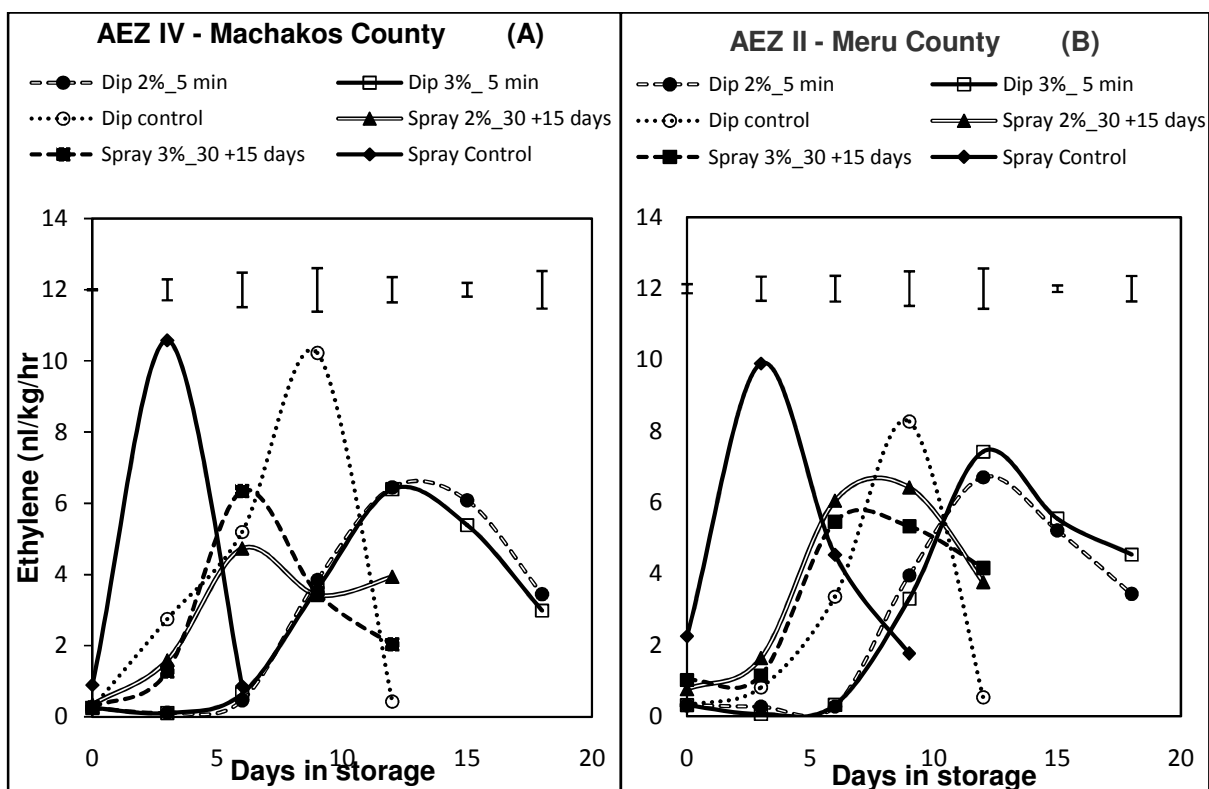
184 3.2 Rate of ethylene production

185 In the pre-harvest spray mode of application, the control fruits had significantly ($P = .05$) high levels of
186 ethylene production with the climacteric peaks of approximately 10 nL/kg/hr occurring 3 days earlier
187 compared to the hexanal treated fruits (Fig. 1A & B). Hexanal treatment significantly ($P = .05$) reduced the
188 rate of ethylene production in both AEZ and delayed the climacteric peaks by 3 days compared to the
189 untreated fruits. The reduced climacteric peaks of 4.8- 6.3 nL/kg/hr and 5.6 – 6.3 nL/kg/hr in the hexanal
190 treated fruits from the drier AEZ IV (Fig. 1A) and wetter AEZ II fruits (Fig. 1B) occurred at day 6 of storage,
191 then ethylene levels drastically declining till the end of storage exhibiting a true climacteric pattern.

192 A significant difference ($P = .05$) was observed between the two modes of hexanal application where post-
193 harvest mode of application delayed the climacteric peaks by 6 days compared to the pre-harvest spray
194 (Fig. 1A & B). However, zone of production did not have any significant effect on the rate of ethylene
195 production.

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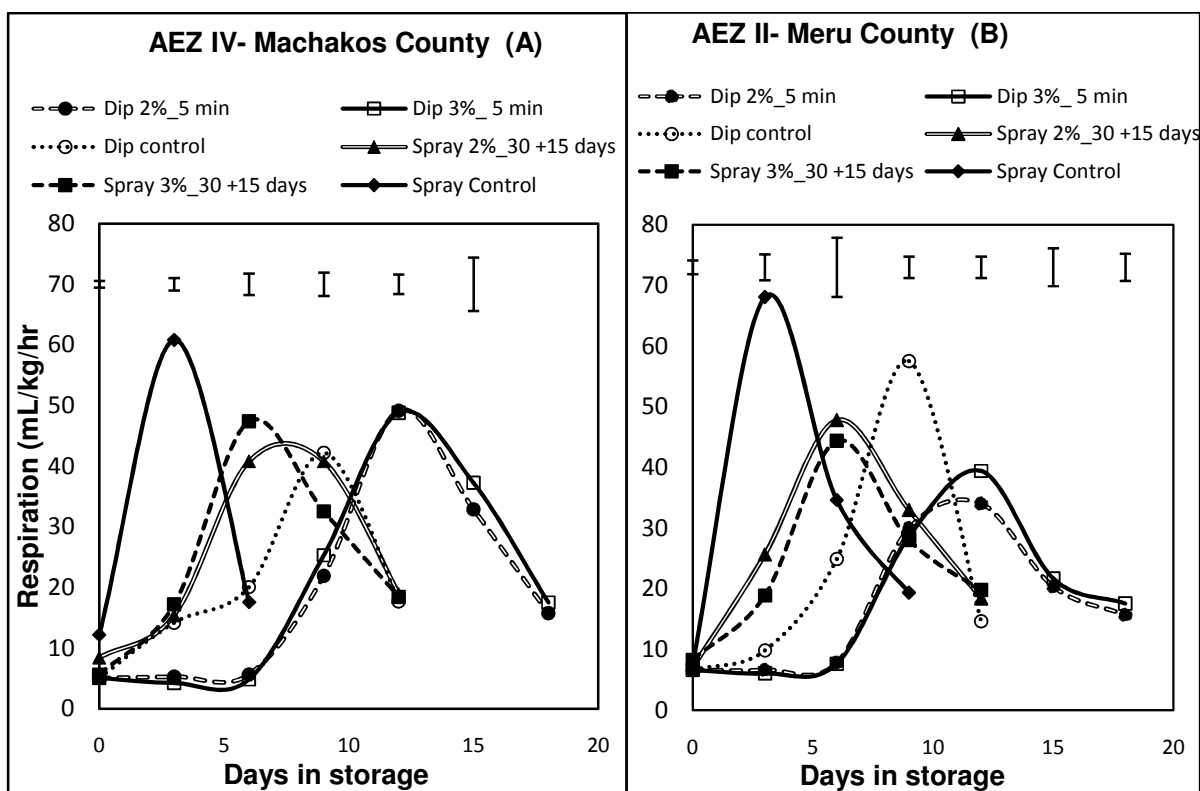


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199 **Fig. 1. Effect of pre and post-harvest application of Hexanal on rate of ethylene production in 'sweet**
 200 **banana' fruits from drier AEZ IV (A) and wetter AEZ II (B). Bars indicate least significant difference**
 201 **(LSD) between means at $p < 0.05$.**

202 3.3 Respiration rate

203 Respiration rate followed a similar pattern to the ethylene production. In both zones of production, hexanal
 204 treatment significantly ($P = .05$) reduced the rate of respiration, with a post-harvest dip mode of application
 205 exhibiting lower rates compared to pre-harvest spray (Fig. 2A & B). Just like in ethylene production, fruits
 206 from the pre-harvest spray mode of application had higher respiratory rate compared to the post-harvest
 207 dipped ones. The high respiratory peaks of 61 mL/kg/h and 69 mL/kg/hr in the controls occurred at day 3 of
 208 storage, compared to 41 -47 mL/kg/h and 44 -48 mL/kg/h in the hexanal treated fruits, 3 days later in drier
 209 AEZ IV and wetter AEZ II respectively (Fig. 2A & B). A similar trend was observed in the post-harvest dip
 210 mode of application experiment, where the treated fruits had lower levels of respiration compared to the
 211 pre-harvest spray experiment with respiratory peaks of 49 mL/kg/h and 34 -39 mL/kg/h, in drier AEZ IV and
 212 colder AEZ II, respectively, occurring 6 days later.

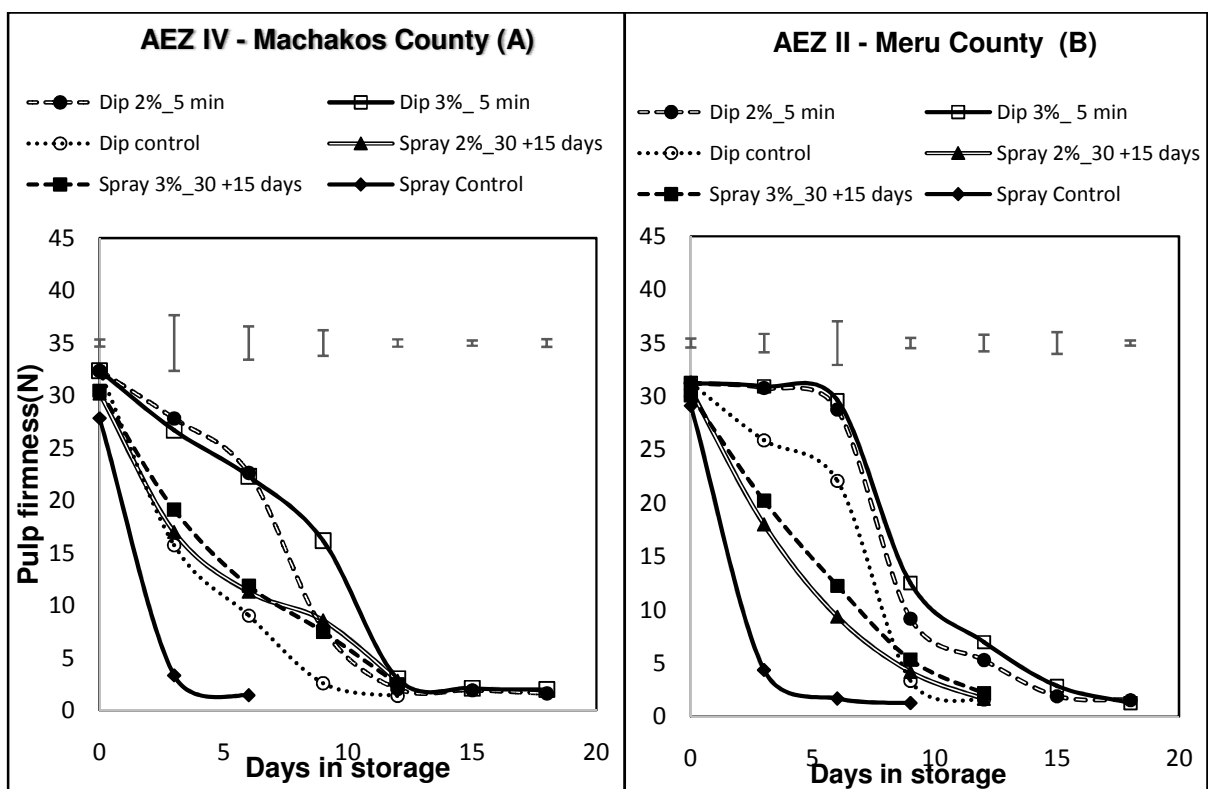


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214 **Fig. 2. Effect of pre and post-harvest application of Hexanal on the rate of respiration in 'sweet**
 215 **banana' fruits from drier AEZ IV (A) and wetter AEZ II (B). Bars indicate least significant difference**
 216 **(LSD) between means at $p < 0.05$.**

217 3.4 Pulp firmness

218 A general reduction in pulp firmness was observed in both the hexanal treated and control fruits as ripening
 219 progressed (Fig. 3A and B). Hexanal treatment applied either as a pre-harvest spray or post-harvest dip
 220 significantly ($P = .05$) delayed pulp softening in both AEZ. Interaction between mode of application and
 221 zone of production had a significant ($P = .05$) effect on the rate of softening with fruits from the drier AEZ IV
 222 (Fig 3A) softening faster compared to those from the colder AEZ II (Fig 3B). The control fruits drastically
 223 lost their pulp firmness by 96% in fruits produced in both AEZ, after 6 and 9 days of storage in the drier
 224 AEZ IV and wetter AEZ II, respectively in the pre-harvest spray mode of application. Similarly, in the post-
 225 harvest dip mode of application, the untreated control fruits had lost approximately 95% of their pulp
 226 firmness after 12 days of storage in both zones. By the 9th day of storage, pre-harvest sprayed fruits had
 227 lost approximately 72% -75% and 82%- 85% of their firmness compared to 50% - 76% and 60% - 71% in
 228 the post-harvest dip treated fruits in the drier AEZ IV and wetter AEZ II, respectively (Fig. 3A and B).



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230 Fig. 3. Effect of pre and post-harvest application Hexanal on pulp firmness in 'sweet banana' fruits
 231 from drier AEZ IV (A) and wetter AEZ II (B). Bars indicate least significant difference (LSD) between
 232 means at $p < 0.05$.

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3.5 Total soluble solids (TSS, °Brix)

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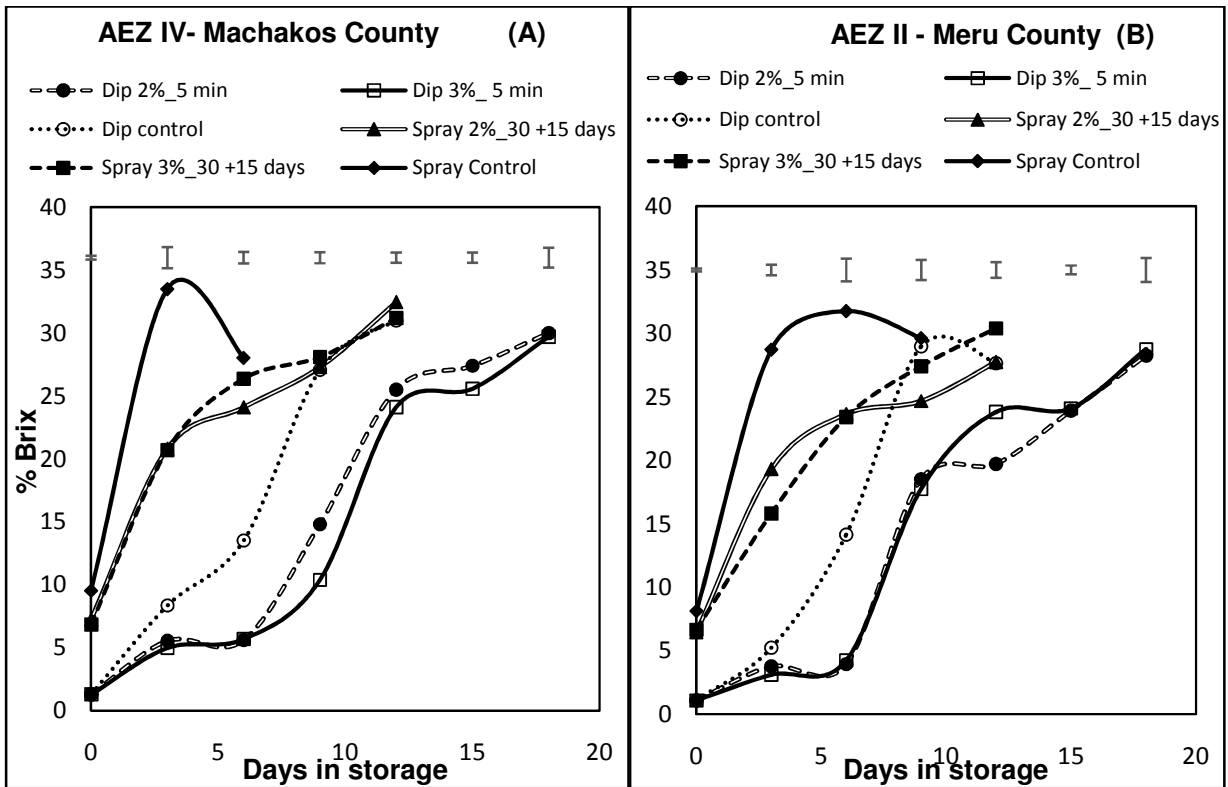
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Total soluble solid (TSS) levels were significantly ($P = .05$) affected by the interaction between zone of production and hexanal treatment. Generally, fruits from the drier AEZ IV (Fig. 4A) had significantly high TSS levels throughout storage compared to those from the colder AEZ II (Fig. 4B). The °brix levels of the untreated fruits from the drier AEZ IV, increased rapidly from an initial value of 1.3 and 9.5° brix to a peak value of 33.81° and 31° brix on day 12 and 3 of storage in the post-harvest dip and pre-harvest spray mode of treatments respectively (Fig. 4A). On the other hand, TSS levels increased gradually from initial of 1.1° brix to peak of 29° brix at day 9 of storage in the post-harvest dip mode of application in the wetter AEZ II (Fig. 4B).

Hexanal treatment significantly ($P = .05$) reduced the rate of TSS increase in both zones and mode of application. However, at the end of storage, the hexanal treated fruits attained almost the same TSS level of approximately 28° - 32° brix compared to the untreated controls.

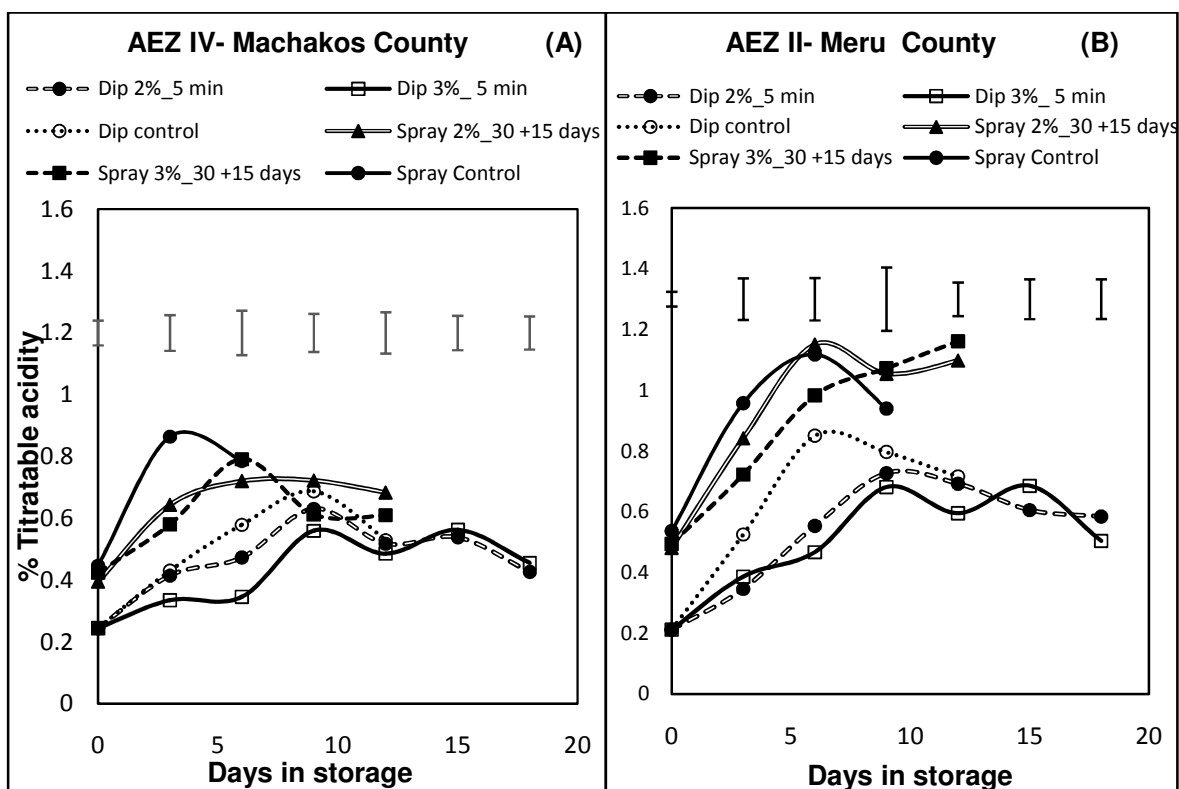


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247 **Fig. 4. Effect of pre and post-harvest application of Hexanal on Total Soluble Solids (TSS) in 'sweet**
 248 **banana' fruits from drier AEZ IV (A) and wetter AEZ II (B). Bars indicate least significant difference**
 249 **(LSD) between means at $p < 0.05$.**

250 **3.6 Total Titratable Acidity (TTA)**

251 As ripening progressed, total titratable acidity (TTA) increased up to a peak level then gradually decreased
 252 till the end of storage (Fig. 5A & B). A significant ($P = .05$) interaction was observed between zone of
 253 production and hexanal treatment with fruits from the colder AEZ II (Fig. 5B) having high TTA levels
 254 throughout the storage period compared to those from the drier AEZ IV (Fig. 5A). Hexanal treatment
 255 significantly ($P = .05$) slowed the rate of TTA increase in both zones of production, irrespective of the mode
 256 of application used (Fig. 5A & B).



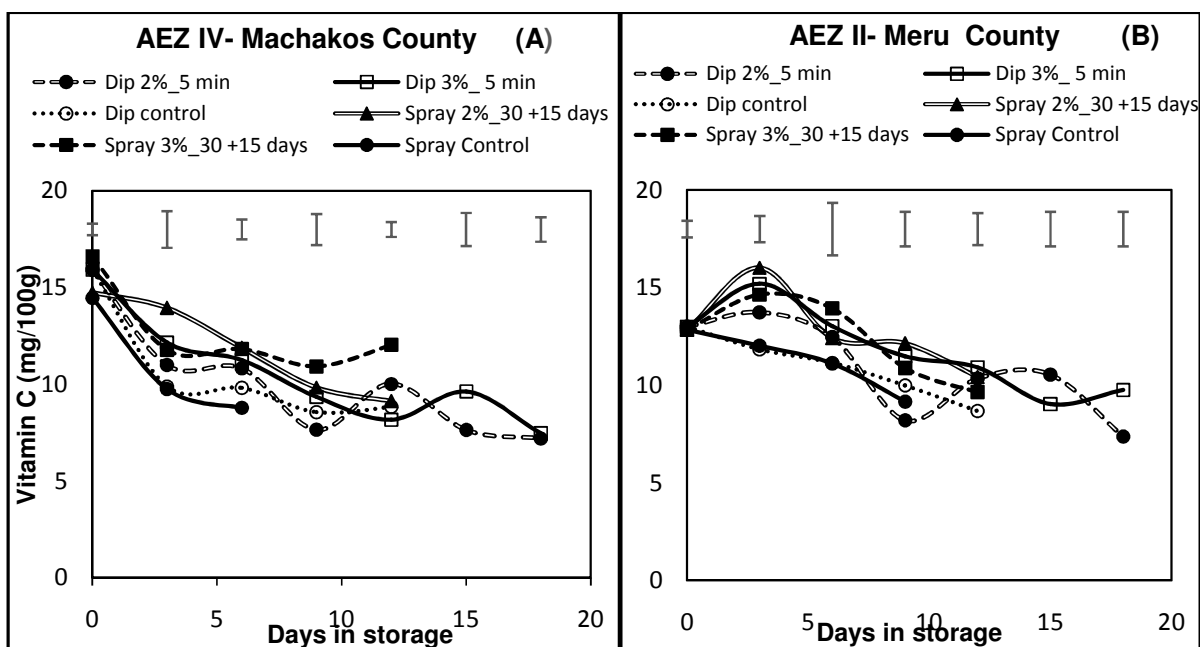
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258 **Fig. 5. Effect of pre and post-harvest application of Hexanal on Total Titratable Acidity (TTA) in**
 259 **'sweet banana' fruits from drier AEZ IV (A) and wetter AEZ II (B). Bars indicate least significant**
 260 **difference (LSD) between means at $p < 0.05$.**

261 3.7 Ascorbic Acid content

262 The ascorbic acid content decreased gradually during storage in all the fruits except in the hexanal treated
 263 fruits pre-harvest spray) from the wetter AEZ II, where an increase was observed up to day 3 of storage
 264 (Fig. 6B). The ascorbic acid levels were significantly ($P = .05$) affected by the interaction between zone of
 265 production and hexanal treatment. Generally, fruits from the wetter AEZ II had significantly ($P = .05$) high
 266 ascorbic acid levels (Fig. 6B) compared to those from the drier AEZ IV (Fig. 6A). Hexanal treatment
 267 significantly ($P = .05$) slowed the rate of ascorbic acid reduction with the treated fruits maintaining relatively
 268 higher levels throughout the storage period compared to the controls in both AEZ (Fig. 6A & B).

269 Ascorbic acid levels decreased rapidly in the control fruits from an initial of 14.5 mg/100g and 11.7 mg/100g
 270 to an average of 8.8 mg/100g and 9.2 mg/100g in the drier AEZ IV and wetter AEZ II respectively, by the
 271 end of storage (day 9) in the pre-harvest spray mode of application (Fig. 6A & B). Contrasting results were
 272 observed in the hexanal treated fruits where 2% concentration was more effective in the drier AEZ IV fruits
 273 where else in wetter AEZ II, 3% concentration was more effective. In the post-harvest dip experiment, the
 274 ascorbic acid levels decreased from initial values of 15.9 mg/100g and 13 mg/100g to 7.4 mg/100g and 7.3
 275 - 9.6 mg/100g in the treated fruits at the end of storage (day 18), 6 days later compared to the controls in
 276 AEZ IV and AEZ II, respectively.



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279 **Fig. 6. Effect of pre and post-harvest application of Hexanal on ascorbic acid content in 'sweet**
280 **banana' fruits from drier AEZ IV (A) and wetter AEZ II (B). Bars indicate least significant difference**
281 **(LSD) between means at $p < 0.05$.**

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283 **3.8 Simple sugars (Sucrose, glucose and Fructose)**

284 Sucrose, glucose and fructose increased gradually with ripening in all the fruits irrespective of production
285 zone and hexanal treatment (Table 1 and 2). Sucrose was the most abundant sugar in banana fruits
286 compared to glucose and fructose irrespective of zone of production and hexanal treatment. A significant
287 interaction ($P = .05$) was observed between hexanal treatment and zone of production in both glucose and
288 fructose (Table 1 and 2). Glucose and fructose levels were significantly high ($P = .05$) in the drier AEZ IV
289 fruits (Table 1) compared to those from the wetter AEZ II (Table 2) in both modes of application. However,
290 zone of production did not have any significant effect on the sucrose levels (Table 1 and 2). The increase in
291 sucrose, glucose and fructose content was significantly ($P = .05$) affected by hexanal treatment, were the
292 increase was lower in the treated fruits compared to the controls throughout the storage period. However,
293 no significant differences were observed between 2% and 3% hexanal concentrations evaluated for
294 sucrose and fructose while 3% significantly delayed the increase in glucose compared to 2% in the spray
295 mode of application.

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Table 1. Effect of pre and post-harvest application of Enhanced fresh formulation (EFF) on Glucose, fructose and sucrose content (g/100g) of 'apple' bananas from AEZ IV (Machakos County).

DAYS Dip	SUCROSE			GLUCOSE			FRUCTOSE		
	Control	2%	3%	Control	2%	3%	Control	2%	3%
0	6.4e	6.4h	6.4i	5.4e	5.4e	5.4e	4.8d	4.8de	4.8c
3	48.6d	7.8gh	9.7i	6.9e	3.6e	3.1e	5.5d	2.2e	2.9c
6	68.5d	29.1f	46.4f	15.2d	7.1e	6.7d	18.4c	5.1de	4.5c
9	113.7b	67.1e	64.1e	29.5c	18.6c	19.6c	41.6b	7.6de	6.9c
12	173.0a	97.4d	92.5d	51.1a	33.2b	39.2a	57.3a	31.9b	24.9b
15		191.7a	161.8a		42.4a	44.5a		37.3b	37.8a
18		160.5b	150.6b		45.4a	43.5a		45.1a	41.9a
Mean	82.0	80.0	75.9	21.6	22.2	23.1	25.5	19.1	17.7
Spray									
0	4e	1.9h	2.6i	14.7d	4.7e	4.3e	4e	1.9h	2.6i
3	97.4c	17.8g	19.7h	55.7a	15.6d	11.3d	97.4c	17.8g	19.7h
6	87.5c	36.1f	32.5g	44b	22.8c	17c.1	87.5c	36.1f	32.5g
9		87.1d	85.8d		38.4b	34.3b		87.1d	85.8d
12		111.5c	114.9c		42.5a	36.4b		111.5c	114.9c
15									
18									
Mean	63.0	50.9	51.1	38.1	24.8	20.7	35.8	22.8	23.1
LSD*	10.2			5.5			7.4		

305 *Values followed by the same letter within the same column are not significantly different between the treatments using*
306 *Fishers Least significant difference (($p < 0.05$)).*

307

308 **Table 2. Effect of pre and post-harvest application of hexanal on Glucose, fructose and sucrose**
309 **content (g/100g) of 'sweet banana' fruits from the wetter AEZ II (Meru County).**

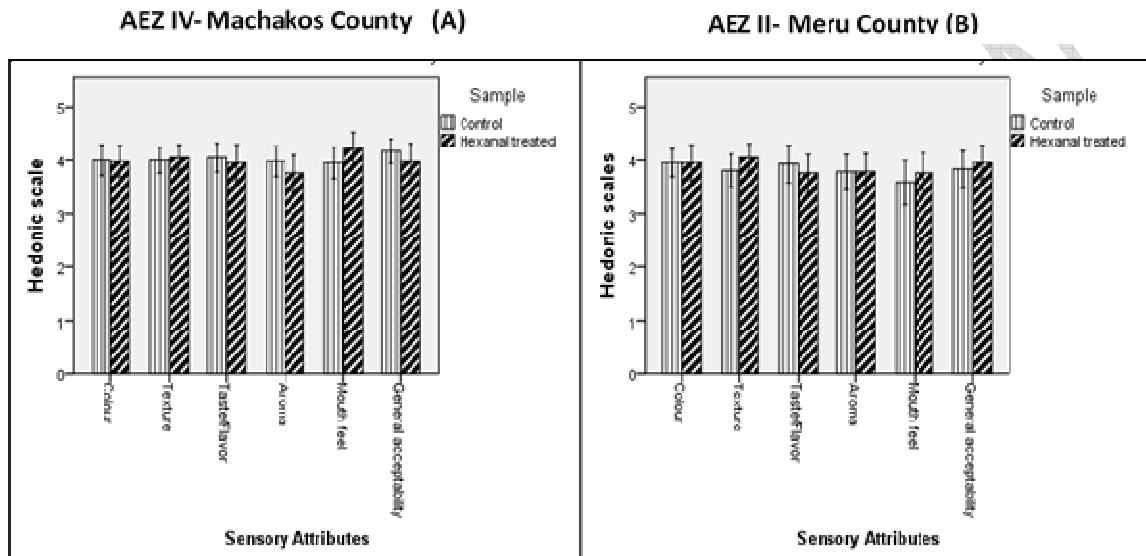
DAYS Dip	SUCROSE			GLUCOSE			FRUCTOSE		
	Control	2%	3%	Control	2%	3%	Control	2%	3%
0	3.8c	3.8e	3.8dc	4.1f	4.1f	4.1d	4.4e	4.4d	4.4e
3	27.5c	3.4e	7.5dc	7.5ef	5.0f	5.6d	4.5e	3.0d	3.6e
6	41.1c	12.0d	5.4dc	29.1d	5.9f	3.7d	15.7d	6.6d	3.7e
9	95.9b	31.5d	56.3c	28.5d	23.3d	32.4b	41.4a	18.9c	28.0c
12	155.0a	79.3c	109.3b	44.8a	34.0c	49.3a	46.2a	32.3a	36.1b
15		155.5a	152.2a		34.1c	53.3a		31.8a	42.4a
18		147.7a	135.9a		51.7a	32.2b		36.6a	33.7b
Mean	65.6	61.9	67.2	22.8	22.6	25.8	22.4	19.1	21.7
Spray									
0	6.1c	3.5e	5.1dc	10.2e	3.3f	3.1d	4.3e	2.8d	1.4e
3	93.1b	39.2d	28.8c	34.3b	14.6e	7.8d	25.3c	20.6c	14.8d
6	101.5b	119b	92.3b	41.2a	27.6d	23.2c	46.1a	16.2c	18.3d
9	107.9b	93b	126.4a	33b	31.2cd	29.2b	30.7b	27.5b	33.3b
12		119.5b	109.2		43.3b	35.1b		30.7b	35.3b
15									
18									
Mean	77.2	74.8	72.4	34.1	24	19.5	26.6	19.6	20.6
LSD*	35.6			4.8			4.9		

310 *Values followed by the same letter within the same column are not significantly different between the treatments using*
311 *Fishers Least significant difference (($p < 0.05$)).*

312

3.9 Sensory Quality Evaluation

Generally, there was no significant ($P = .05$) differences observed in all the quality attributes scores in both zones between the hexanal treated and control fruits (Fig. 7A & B). The treated and control fruits from both AEZ scored almost the same scores for peel color, texture in AEZ IV fruits (Fig. 7A) and aroma in AEZ II (Fig. 7B). On the other hand, hexanal treated fruits scored slightly high for taste/flavour in both AEZs (Fig. 7A & B) while general acceptability and aroma scored highest in AEZ IV (Fig. 7A) fruits though this was not significantly different.



320

321 **Fig. 7. Hedonic scores for sensory quality attributes of 'sweet banana' variety harvested from**
322 **Machakos and Meru Counties and treated with Hexanal or left untreated to act as the control. The**
323 **values on Y-axis represent scores on a 5-point hedonic scale (1 = dislike (worst), 2 = (dislike**
324 **moderately), 3 = (neither like nor dislike) 4 = (like moderately) and 5= (Like extremely/best)). The**
325 **vertical bars represent means \pm SE.**

326 4.0. DISCUSSION

327 Application of appropriate post-harvest technologies in banana fruits is of paramount importance in order to
328 minimize losses after harvest and maintain the best possible quality. Over the past decades, different post-
329 harvest technologies have been developed and tested in various fruits [1, 21]. However, the adoption rate
330 of most of these technologies depends on its appropriateness, cost, versatility and value of the commodity.
331 Moreover, most of the consumers and other actors in the value chain in the recent past have high affinity
332 for naturally-occurring post-harvest preservative compounds which are environmentally friendly, pose no
333 health hazard and are easy to use. Therefore, there is need to test the suitability of biological compounds
334 such as hexanal to enhance banana shelf life while preserving its quality. The objective of this study was to
335 evaluate the efficacy of hexanal, a naturally-occurring compound in enhancing shelf-life and quality of
336 'sweet banana' fruits in Kenya when applied as a pre-harvest spray or post-harvest dip.

337 Overall, zone of production had a significant effect on fruits shelf-life and quality. Fruits from the drier AEZ
338 IV, ripened faster and had high content of $^{\circ}$ brix and simple sugars as compared to those from the wetter
339 AEZ II. This could be as a result of differences on the prevailing environmental conditions such as

340 temperatures and light as well as cultural practices which have all been reported to impact on the
341 physiology and post-harvest quality of fruits [13]. Hexanal treatment significantly extended shelf-life by 6
342 days in the post-harvest dip mode of application in both zones compared to the controls. On the other
343 hand, fruits sprayed with hexanal had a shelf life of 6 and 3 days in the drier AEZ IV and wetter AEZ II fruits
344 respectively compared to the controls irrespective of the concentration used. This observed increase in
345 shelf life is very significant especially to small scale farmers who will benefit by gaining an extra time to
346 source for better market and minimize exploitation by middlemen along the value chain. Banana fruit
347 especially the 'sweet banana' variety when ripe goes from marketable to unmarketable state rapidly,
348 leading to huge post-harvest losses. The observed extended shelf-life of up to 6 days in this study could be
349 as a result of the observed lower rates of ethylene production and respiration in the hexanal treated fruits.
350 Physiologically, an increase in respiration rate leads to a quick utilization of substrates, such as free sugars
351 that contributes to post-harvest losses as previously reported by [25]. Similar findings of extended shelf-life
352 have been reported in other banana varieties such as 'Grand naine' [33], [34] and in other fruits including
353 mangoes [2], papaya [12], Lime [6] and tomatoes [5]. The observed reduced rate of ethylene evolution in
354 the treated fruits may be as a result of hexanal being a weak inhibitor of ethylene as previously reported by
355 **Tiwari and Paliyath**, [31]. A study at molecular level in tomato fruit by **Tiwari and Paliyath**, [31], showed that
356 hexanal treatment in tomato fruit caused moderate down regulation of 1-aminocyclopropane-1-carboxylate
357 synthase 6 (ACS6) and 1-aminocyclopropane-1-carboxylate synthase (ACS) genes. The expression of
358 ACS6 and ACS genes are responsible for the biosynthesis of 1-Aminocyclopropane-1-carboxylic acid
359 (ACC) synthase enzyme, which converts the S-Adenosyl-L-methionine (SAM) to ACC in the ethylene
360 biosynthesis pathway. Hexanal inhibition of ACS genes will lead to a reduction in the evolution of ethylene,
361 and this may explain the low levels of ethylene production observed in this study.

362 Excessive softening is one of the main factors limiting fruit shelf life, transportability and storage in banana
363 fruit resulting to high levels of post-harvest losses. In the present study, the rate of fruit softening was
364 greatly delayed in the hexanal treated fruits compared to the controls throughout the storage period.
365 Softening in banana fruits is majorly as a result of textural changes due to disassembly of the primary cell
366 wall by various hydrolases such as pectin methylesterase, polygalacturonase and pectate lyase among
367 others [32]. However, other mechanism may also be active in determining the overall textural
368 characteristics of banana fruit such as loss of turgor and breakdown of starch to sugar [14]. The observed
369 delayed softening in the treated fruits might be as a result of hexanal reducing the activity of the various
370 enzymes involved in cell wall degradation and modification. A study in tomato [31], showed that hexanal
371 treatment down regulates the expression of genes involved in pectin and hemicellulose degradation which
372 are the major components of the plant cell wall. Additionally, the delay in fruit softening may also be as
373 result of the observed low rate of ethylene production and respiration in the hexanal treated fruits.
374 Ethylene, being a ripening hormone has a strong participation in modulating enzymes involved in fruit
375 softening [16]. Degradation of starch during respiration in fruits such as banana results into pronounced
376 textural changes. Similar results have been reported in banana fruits by **Venkatachalam et al.** [33] in India.
377 Zone of production had a significant effect on fruit firmness with fruits from the drier AEZ IV (Machakos
378 County), softening faster compared to those from the wet AEZ II (Meru County), irrespective of the

379 treatment. This could be attributed to differences in temperatures and rainfall in the different zones; both
380 having been reported to affect fruit softening [13].

381 Total soluble solids (TSS) increased gradually with ripening in all the fruits irrespective of zone of
382 production and hexanal treatment. The observed increase in TSS during ripening may be associated with
383 the breakdown of stored carbohydrates into simple sugars [14]. Fruits from the drier zone IV had higher
384 TSS levels compared to those from the wetter zone II. This could be attributed to high temperatures and
385 longer periods of exposure to sunlight characteristic of AEZ IV which led to increased accumulation of dry
386 matter content. Similar results have been reported in papaya [12] and mangoes [20]. In general, the rate of
387 TSS increase was significantly low in the Hexanal treated fruits throughout the storage duration and could
388 be attributed to the observed low rate of respiration and ripening process. Low rate of respiration leads to a
389 decrease in metabolic activity and slow conversion of starch to sugars, a possible explanation of delayed
390 increase in TSS content in the hexanal treated fruits. Our results concur with those of Anusuya *et al.* [2],
391 who reported similar results in mango fruits. Changes in simple sugars such as sucrose, glucose and
392 fructose followed a similar trend to the one observed in TSS. In the present study, levels of this individual
393 sugars increased drastically during the ripening process in all the fruits. However, hexanal treatment
394 significantly slowed down the increase rate of glucose and fructose. This might be as a result of the
395 observed delayed ripening and reduced rate of respiration in the hexanal treated fruits. During ripening
396 process, starch, which is the major form of carbohydrates in banana fruit, is usually catabolized into simple
397 sugars, which enters the metabolic pool where they are used as respiratory substrates or further converted
398 to other metabolites. Similar findings have been reported in hexanal treated banana fruits by
399 Venkatachalam *et al.* [33].

400 Banana is one of the few fruits whose TTA levels increases with ripening up to a maximum value then
401 decreases in the fully ripe stage as reported by Lechaudel and Joas, [13]. This is as result of increase in
402 malic acid from 1.8 meq/100g to 6.2 meq/100g during ripening [14]. In the present study, hexanal treatment
403 delayed the rate of TTA increase as compared to the drastic increase in the control fruits which peaked at
404 day 3 of storage. This could be attributed to the observed reduced rate of ripening in the hexanal treated
405 fruits. Additionally, reduced activities of enzymes such as malate dehydrogenase, which influence the level
406 of malic acid in banana could further explain the delayed rate of TTA increase by hexanal treatment.

407 Ascorbic acid is an important quality trait in fruits. In the present study, ascorbic acid levels decreased
408 gradually in all the fruits as ripening advanced during storage. The decrease in vitamin C during ripening is
409 partly due to degradation of ascorbic acid through oxidation [3]. The decrease in ascorbic acid was less
410 rapid in the hexanal treated fruits compared to the untreated controls. Higher retention of ascorbic acid
411 observed in the hexanal treated fruits may be as a result of reduced enzymatic oxidation by hexanal.

412 Various quality attributes such as peel color, firmness, aroma, taste, mouth-feel and general acceptability
413 were evaluated during the sensory evaluation analysis. The sensory evaluation results showed that,
414 hexanal treatment did not have any significant effect on the various quality parameters scored. Further,
415 there was no significant difference on the general acceptability of the treated and the control fruits. This
416 indicates that hexanal's effect on shelf life of banana fruit did not have detrimental effects on the various
417 quality parameters. These results are in agreement with a study by Siriboon and Banluisilp, [28], who

418 reported that hexanal treatment does not affect the expression of genes involved in quality development
419 pathway of tomato fruit.

420

421 **5.0. CONCLUSION**

422 Overall, results of this study indicated that, the use of Hexanal has the potential to increase 'sweet banana'
423 shelf-life by at least 6 days in case of post-harvest dip, 6 and 3 days in pre-harvest sprayed fruits from drier
424 AEZ IV and the wetter AEZ II respectively, without affecting the quality attributes. This results have also
425 showed that hexanal efficacy might be influenced by zone of production and further studies need to be
426 conducted to validate this. However, there was no significant difference between the 2% and 3% hexanal
427 concentrations tested and both concentrations were equally effective. Therefore, this technology shows
428 great promise in enhancing the shelf life while preserving quality attributes of banana fruits. This in turn can
429 reduce the huge post-harvest losses currently being incurred in developing countries such as Kenya.

430

431 **COMPETING INTERESTS**

432 Authors have declared that no competing interests exist regarding the publication of this paper.

433

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