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Artificial Ripening to Reduce Postharvest losses of Avocado

K.H.Sarananda[1], U.N.G.C.Kumari, J.P. Eeswara[2] and R.M.N.D. Rathnayaka[3].
[1]Food Research Unit, Department of Agriculture.
[2]Department of Crop Science, Faculty of Agriculture, University of Peradeniya.
[3]Department of Natural Resources, Faculty of Applied Sciences, University of Sabaragamuwa.

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ABSTRACT

Delay in ripening of mature avocado results in excessive weight loss and substantial postharvest loss due to anthracnose and stem-end-rot. Over-mature avocado ripen within a short time, but significant postharvest losses take place due to excessive level of anthracnose. Of the artificial ripening methods tested Ethral 2 ml/l of water and CaC_2 1g/kg of fruit showed 5 days shortening of ripening period of mature avocado. However, Ethral 1ml/l concentration did not induce the ripening process successfully. Percentage weight loss of artificially ripened avocado was significantly reduced compared to that of control. Occurrence of stem-end-rot was significantly reduced when fruits were artificially ripened. Removal of stem end immediately after harvest increased the stem-end-rot. However, if stem detached fruits are artificially ripened the level of stem end rot was reduced. Over-mature avocado had very high level of anthracnose. However, the anthracnose level was significantly reduced when fruits were artificially ripened. Inoculated studies with *Botryodiplodia theobromae* showed, artificial ripening reduced the stem-end-rot of avocado even in stem end detached fruits. However, when over-mature fruits were inoculated with *Colletotrichum gleoesporioides* no response to artificial ripening was observed. Sensory evaluation study showed the quality of artificially ripened fruits was similar to that of control.

INTRODUCTION

Avocado is considered a nutrient dense food, rich in fat, carbohydrates and vitamins. The crop is mainly grown in Mid Country Wet Zone and in Low Country Wet Zone. Although the crop is not grown on an orchard scale, a sizable volume of harvested crop from backyard gardens comes to the market. Postharvest loss of avocado is reported to be 46% (Anonymous, 1998). The excessive postharvest loss during its marketing results in increasing the price gap between producer and consumer. Higher weight loss, postharvest diseases are major causes for excessive postharvest loss of avocado (Coates *et al., 1995*). The major postharvest diseases associated with avocados are Anthracnose (*Colletotrichum gleoesporioide)* and stem-end rot a disease complex (*Botryodiplodia theobromae, Phomopsis sp and Fusarium spp.*) (Snowdon, 1990). Avocado is a climacteric fruit and produce ethylene during ripening (Biale, 1960). However, fruit ripening is not initiated as long as fruits are attached to the tree (Gazit and Blumenfeld, 1970). Delayed harvesting therefore leads to over mature fruits resulting more disease infection through cracks of the skin. Avocados have naturally occurring ripening inhibitors as long as they are attached to the plant (Gazit and Blumenfeld, 1970). However, due to climacteric nature of fruits the ripening process can be induced by exposing to either acetylene or ethylene (Seymour and Smith, 1986). Moistened calcium carbide easily generates acetylene gas. Ethylene gas however is not locally produced and the use of imported gas is not economical. Changing the pH of commercially available Ethral can readily generate ethylene gas (Sarananda *et al.,* 2000). If artificial ripening is possible the time taken for ripening can be shortened which eventually results lesser postharvest loss. Studies were therefore conducted to find out the possibility of inducing the ripening process of avocado and to evaluate the quality of ripened fruits.

MATERIALS AND METHODS

Experiment 1. Effect of ethylene and acetylene on inducing ripening process of avocado harvested at different stages of maturity.

Mature and fully mature avocados were harvested from a single tree using an improved picking pole. Fruit stalk was trimmed; near stem end and fruits of each maturity stage were grouped into four. Each group contained 20 fruits. Plastic bins (60 l) were used as ripening chambers. Ten ml of diluted Ethral of 1 and 2 ml/ l of water respectively placed in a 25ml beaker and placed in separate bins. Fruits were placed in the bin just before closing the lids and 10ml of 0.1N NaOH were poured into both concentrations of Ethral. The bins were kept sealed for 24hrs at ambient temperature (25 $\pm 2^0$C). Bins were then opened and fruits were allowed to ripen. Same bins were used to treat with calcium carbide. The required quantity of calcium carbide (2g/Kg of avocado) was weighed, wrapped with newspaper and placed at the bottom of the container. The bin was sealed and kept at ambient temperature similar to Ethral treated bins. Control fruits were also stored in a closed bin for 24 hrs and stored at the same place. After 24 hrs all the bins were opened and fruits were allowed to ripen. Percentage weight loss and time taken for ripening was recorded. The statistical design used was Completely Randomized Design with 20 replicates.

Experiment 2. Effect of stage of maturity at harvest and removal of stem-end on postharvest disease development.

Mature and over mature avocados were harvested from the same tree using an improved picking pole. Forty fruits of each stage of maturity were divided into two groups. The fruit stalk of the first group (20) fruits was trimmed leaving the basal part of the stalk attached to the fruit. The fruit stalk of the remaining 20 fruits was completely removed. Each group of 20 fruits was again divided into two, while 10 fruits was used for Ethral ripening the remaining 10 fruits was allowed to naturally ripe. When fruits were ripened, table ripe stage, stem-end rot and Anthracnose levels were recorded. When 5% stem-end of the fruit became discoloured it was considered as stem-end rot infected fruit which has no market demand. Anthracnose levels were recorded based on black colour lesion development. The black coloured area presented as percentage area infected over the whole fruit surface. The experimental design used was Completely Randomized Design with 10 replications.

Experiment 3. Effect of stem-end removing on severity of stem-end rot development of avocado inoculated with *Botryodiplodia theobromae.*

Stem-end attached and stem-end detached 10 fruits from each mature and fully mature avocados were prepared as described in Experiment 2. *Botryodiplodia theobromae* isolated from stem-end-rot infected avocado fruit was grown on PDA. Once spores are formed spores released from pycnidia were used for inoculation. The spore suspension of 2.5×10^4 Spore/ml was prepared using sterilized water. 0.1ml of spore suspension was used to inoculate either the wounded scar of the stem-detached fruit or wounded area of the stem where the stem was trimmed. Sterilized pipettes were used for inoculation and inoculated fruits were incubated for 4 hrs. in a humid chamber. Ten fruits of each treatment were induced for ripening using Ethral as described in experiment 1 while the remaining 10 fruits served as control. At the table ripe stage the stem-end-rot and the Anthracnose levels were recorded as described in experiment 2. The experimental design used was Completely Randomized Design with 10 replications.

Experiment 4. Sensory parameters of ripe avocados as affected by ripening treatment.

Mature avocados were harvested using an improved picking pole. The stems of fruits were trimmed leaving a stem-base attached to the fruit. Of the 40 fruits harvested 20 were induced for ripening using 2ml/l Ethral concentration as described in Experiment 1. At full mature stage both naturally ripe and Ethral induced ripened fruits were given for 6 trained taste panel. Flesh colour, off odour, taste, texture and overall acceptability were recorded qualitatively and quantitatively using hedonic scale. The experimental design used was Completely Randomized Design.

RESULTS AND DISCUSSION

Calcium carbide and both concentrations of Ethral significantly reduced the time taken for ripening of mature avocados compared to that in controls (Table 1). Of the methods used to induced ripening, Ethral 2ml/l and CaC_2 were equally effective in inducing the ripening process of the mature fruits. The Ethral concentration 1ml/l was not as effective as 2ml/l in inducing the ripening process. In addition to slow ripening, non-uniform ripening was also observed with 1ml/l concentration of Ethral. However, when 1ml/l concentration was used to ripe over mature fruits the inducing process was similar to those with Ethral 2ml/l and CaC_2. Table 1 further shows that the time taken for ripening of mature avocado can significantly be shortened (5 days) by artificially treating with either CaC_2 or Ethral 2ml/l. However, no significant difference in time taken for ripening of over mature avocados was observed with the 3 methods of induction. In addition, although the time taken for ripening of over-mature avocados was significantly reduced by the treatments, the difference was around one day.

Table 1: Mean time taken for ripening (days) of mature and over mature avocados treated with Ethral and CaC_2.

Treatment	*Mature*	*Over Mature*
Ethral 1ml/l	9.0[b]	7.0[b]
Ethral 2m/l	6.8[c]	6.5[b]
CaC_2	7.0[c]	6.8[b]
Control	12.0[a]	8.0[a]

Treatment means in a column having a common letter(s) are not significantly different by DMRT 5%.

The maximum percentage weight loss was observed in mature, control fruits when they were ripe (Table 2). The minimum weight loss at ripening was observed in fruits treated with Ethral 2ml/l and CaC_2. A similar pattern of moisture loss was recorded with over mature fruits. However, the weight loss values of over-mature fruits were lower than those of mature fruits. The minimum weight loss was observed at both maturity stages when Ethral 2ml/l and CaC_2 2g/Kg were used.

Table 2: Average percentage weight loss of ripe avocados harvested at mature and over mature stages and treated with Ethral and CaC_2.

Treatment	*Mature*	*Over Mature*
Ethral 1ml/l	8.9[b]	7.6[b]
Ethral 2m/l	7.8[c]	6.5[c]
CaC_2	7.8[c]	6.5[c]
Control	10.2[a]	8.7[a]

Treatment means in a column having a common letter(s) are not significantly different by DMRT 5%.

Stem-end-rot of all avocados was significantly reduced when fruits were ripened artificially (Table 3). Higher incidence of stem-end rot was recorded in all naturally ripe fruits compared to fruits ripened artificially. A very high incidence of stem-end rot was observed in stem-end detached, naturally ripe fruit. The stem-end rot in stem-detached fruit was higher when fruits were harvested at mature stage. However, when fruits were ripened artificially the incidence was very much reduced.

Table 3: Mean percentage stem-end-rot of ripe avocados as affected by stage of maturity at harvest and removal of stem-end.

Treatment	*Mature*		*Over mature*	
	Stem-end attached	*Stem-end detached*	*Stem-end attached*	*Stem end detached*
Natural ripening	8.2	18.6	3.1	6.3
Ethral 2ml/l	1.6	4.5	0.5	1.2
L.S.D	2.68	2.93	0.36	1.42

Artificial ripening of fruits significantly reduced the occurrence of Anthracnose on all avocados (Table 4). The difference in reduction was highly prominent with over-mature avocados. However, there was no effect of removal of stem-end on Anthracnose development of avocados. Results further showed the levels of Anthracnose were

extremely high when fruits were harvested at over-mature stage.

Table 4: Mean percentage of Anthracnose of ripe avocados as affected by stage of maturity at harvest and removal of stem-end.

Treatment	Mature		Over mature	
	Stem-end Attached	Stem-end detached	Stem-end Attached	Stem-end detached
Natural ripening	6.8	7.0	42.8	43.5
Ethral 2ml/l	3.2	4.1	21.7	20.6
L.S.D	1.21	1.62	3.12	3.96

The estimated median values recorded for stem-end-rot development were always significantly higher in naturally ripened fruits compared to those in artificially ripened fruits (Table 5). Although stem-end detached fruits showed a higher incidence of disease development, it was significantly reduced in artificially ripened fruits. In addition stem-end detached fruits were completely rot when fruits were allowed for natural ripening. Although all the fruits used for the experiment were treated with the same volume of *B. theobromae* the disease level was very low when stem-end attached fruits were artificially ripened.

Table 5: Estimated median values of stem-end-rot development of inoculated avocados with *B. theobromae* as affected by stage of maturity and removal of stem-end.

Treatment	Mature		Over mature	
	Stem-end Attached	Stem-end detached	Stem-end Attached	Stem-end detached
Natural ripening	2.2	4.0	1.2	4.0
Ethral 2ml/l	1.6	3.2	0.6	2.8

Disease index: 1=1-10% surface area discoloured, 2=11-20% surface area discoloured, 3=21-30% surface area discoloured and 4=31%< surface area discoloured.

The estimated median values of Anthracnose were higher in naturally ripe fruits of both mature and over mature (Table 6). However, the severity of the disease was significantly reduced with the artificial ripening of fruits. Over mature fruits were more susceptible to Anthracnose than that of mature fruits.

Table 6: Estimated medians of Anthracnose of avocados inoculated with *C. Gleoesporoides* as affected by artificial ripening.

Treatment	Mature	Over mature
Natural ripening	4.0	3.8
Artificial ripening	2.6	3.2

Disease index: 1=1-10% surface area discoloured, 2=11-20% surface area discoloured, 3=21-30% surface area discoloured and 4=>31% surface area discoloured.

The qualitative aspects of organoleptic parameters, flesh colour, off odour, taste, texture and overall acceptability were equal both in naturally ripe and artificially ripened fruits (Table 7). The quantitative analysis of all the parameters tested above also showed no significant difference.

Table 7: Sensory evaluation of Ethral induced and naturally ripe mature avocados.

Parameter	Ethral 2ml/l	Naturally ripe
Flesh colour	Cream yellow (85%)	Cream yellow(87%)
Off odour	Absent (0%)	Absent (0%)
Taste	Typical avocado(90%)	Typical avocado(93%)
Texture	Smooth (83%)	Smooth (85%)
Overall acceptability	High (95%)	High (97%)

Data in parenthesis shows the quantitative value of each parameter.

Although avocado belongs to climacteric group, the fruit does not ripe as long as it is attached to the tree (Gazit and Nlumenfeld, 1970). Due to this reason fruit may remain over mature until they are harvested. Longer the period the fruits are attached to the tree higher the level of latent infection caused by both *C. gloesporioides* and *B. theobromae.* Although over mature fruits naturally ripe earlier, the disease incidence associated with these fruits increased the postharvest loss and reduced the external appearance. Mature fruits, however took about 12 days to ripe at ambient temperature resulting excessive weight loss.

Zauberman and Schiffmann-Nadel, (1972) reported that mature avocado took lesser time than less mature fruit to ripen which confirm the results observed in this experiment. Excessive respiration during prolonged exposure to ambient condition reduces the external quality of the fruit. In addition, a greater proportion was lost due to direct weight loss. Lesser time taken for ripening of Ethral induced bananas compared to that in naturally ripe has been reported by Sarananda *et al.,*(2000). The same effect would have caused for the lesser weight loss in artificially ripened avocados.

Mature avocados are artificially ripened by exposing them to 100-ppm ethylene at 16-18C for 3 days (Fitzell and Coates, 1995). Since ethylene is a natural ripening hormone the quality of the fruit is not impaired by the treatment. Ethral also release ethylene when the pH is increased above 6.5. Exposing the ethylene released from 2-ml/l concentration had induced ripening process of mature avocados. However, slow and uneven ripening observed with 1 ml/l concentration showed that the ethylene generated from the particular concentration might not be sufficient to initiate autocatalytic ethylene production. Although the ethylene liberated from the two different Ethral concentrations used not quantified, the response to ripening clearly showed that 2-ml/l concentration was satisfactory. Ripening process of 'Cavendish' banana fruit can be induced by exposing to 10ppm Ethylene (Seymour *et al*., 1993). Induction of ripening process of 'Embul' banana has been reported using 1ml/l Ethral (Sarananda *et al.,* 1990). No proper response to 1ml/l Ethral in avocados showed the fruit requires a higher concentration of Ethylene for induction. This observation confirms with the results reported that avocado needs 100 ppm Ethylene and exposure period of 24 hrs to induce ripening process (Zauberman *et al .,* 1988).

Acetylene can also induce the ethylene biosynthesis of climacteric fruits at higher concentration (Seymour *et al*., 1993). Cheaper and readily available acetylene source, commercially available calcium carbide was also equally effective in inducing ripening process. Although calcium carbide is widely used to induce ripening process of mango and banana no information is available on inducing ripening process of avocado.

B.theobromae causing stem end rot and anthracnose caused by *Colletotrichum gleoesporioides* are living on avocado tree hence pre-harvest infection is very common (Coates *et al.,* 1995). Although the infection of stem end rot is pre-harvest, the disease development of naturally infected fruits can be reduced to very low level if the fruit stalk is allowed to remain attached until fruits are artificially ripened. Detaching the stem from the fruit immediately after harvest causes a fresh wound, which synthesizes wound ethylene causing the wounded area to initiate faster ripening. Higher incidences of stem-end-rot reported previously in stem broken fruits during harvesting (Coates *et al*., 1995) confirm the results of this experiment. A significantly less incidence of stem-end-rot even in stem detached avocados may be due to faster ripening before the disease spreads. Early ripening provides lesser time period for the disease development hence that the fruit can be consumed. Fully mature fruits however, naturally ripe within a short period of 8 days and hence, relatively low disease incidence was observed. The disease level can further be reduced by artificial ripening due to further reduction of time taken for ripening.

Anthracnose caused by *C. gleoesporioides* is an essentially a quiescent infected disease (Coates *et al.,* 1995). When over mature fruits were allowed to ripe naturally a very high incidence of anthracnose was observed because the longer time of attachment of the fruits to the tree possibly increase the infected inoculum level compared to those harvested at correct maturity stage. Although artificial ripening significantly reduced the Anthracnose level a higher incidence was observed in over mature fruits. Leaving fruits on the tree beyond the harvest maturity can therefore increase the level of Anthracnose on the fruits.

When the wounded area after stem detaching was inoculated with spore suspension of *B. theobromae* all the fruits were severely infected when they were naturally ripe. If fruits are artificially ripened the disease level can significantly reduced. Artificial ripening of over mature fruits provided shorter time for the pathogen to multiply and make lesion. Inoculation of stem attached fruits showed relatively low level of stem-end-rot when they were allowed for natural ripening. The disease level however was further reduced when inoculated fruits were artificially ripened. This

observation clearly shows that *B. therobromae* penetrates through the wound caused during stem end detachment. The delay in penetration of the pathogen through the attached stem end delays the stem-end-rot development, in stem attached fruits. If those fruits are artificially ripened, ripening process of the fruit is completed before the pathogen enters the flesh.

Artificial ripening significantly reduced the anthracnose disease of mature avocados when they were artificially ripened. This may be due to early ripening of the inoculated fruit before the pathogen multiply and cause lesions. However, when fruits were over mature, very high level of anthracnose was found even with artificial ripening. This may be due to very high natural infection level before harvest together with introduced inoculum that can cause rapid disease development.

Sensory evaluation on flesh colour, off odour, taste, texture and overall acceptability of artificially ripened fruits showed no significant difference to those of naturally ripe fruits (Table 7). The taste panel was not able to identify artificially ripened fruits from naturally ripe fruits.

Based on these observations, harvesting avocados at mature stage, trimming the stem having the base of the stem attached to the fruit and subjecting to artificial ripening using ethral 2ml/l can reduce the post harvest loss to a greater extent. If these technologies are applied percentage weight loss is reduced, development of anthracnose and stem end rot are delayed. No impairment of the taste of artificially ripened fruits showed the possibility of using the technology in commercial scale.

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