Preservation of lime fruits under modified atmospheric condition created in a sand bed

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***Abstract*- Fresh limes were taken and stored under modified atmospheric conditions created in eight sand beds pertaining to two factor factorial experimental design. Three variables Charcoal & $KMnO_4$ (Ethylene scrubbers) and $NaHCO_3$ (CO_2 releasing agent) were taken and mixed with fine river sand (FRS) at two levels, 0 & 5g, 0 & 3g and 0 & 3g respectively per 100g of sand. FRS was selected after measuring bulk density of 4 bedding materials, sea sand, granite powder, clay, fine and coarse river sand. Fresh limes were stored in these treatments and measured changes occurrence on color and pH weekly and titrable acidity biweekly for a period of 10 weeks. All treatments were replicated thrice and results were compared with the control treatment; which was fresh lime, stored under normal atmospheric condition without sand. Lime stored in fine river sand with 5g of Charcoal and 3g of $KMnO_4$ without $NaHCO_3$ was the best treatment as it was capable to preserve green to yellow color of lime for 9-10 weeks because this treatment contained two ethylene scrubbers $KMnO_4$ and charcoal. This treatment also had least changes on pH, titrable acidity and peel color. The next best treatment was lime stored with charcoal and without NaHCO3 & $KMnO_4$ because this treatment was capable to maintain color of lime from green to yellow for 7- 8 weeks. Treatments contained NaHCO3 had least green life (2 weeks) because $NaHCO_3$ can create an anaerobic environment.**

***Index Terms*- Lime, modified atmosphere, factorial design, green life, post harvest life**

I. INTRODUCTION

Lime(*Citrus aurantifolia*) is a citrus fruit typically round & green, 3–6 cm in diameter, and enclosed with an acidic pulp which is a good source of vitamin C, and is often used to intonation the flavors of foods and beverages. Lime is a non climacteric seasonal fruit and harvesting is confined to a particular time period of a year due to its seasonal behavior. The lime harvesting season is usually beginning in Sri Lanka in December and goes up to July while leaving August as the low harvesting season and September to November period as the off season. During February to March, the harvest reaches to its peak. Price of lime reduces sharply during January to May duo to it's over production (Department of Agriculture, 2006). However, lime is a perishable food product and bulk of it, is coming from small scale domestic farmers but, who do not posses user-friendly low cost technology to preserve the excess production; because small scale producers are not affordable to make use modern high cost storing methods. Thus, a large amount of lime is wasted by the end of season. Moreover, during off season, price of the lime is sky rocketing due to dearth of the production. Whereas, even if lime is stored under cold room condition, shelf life of which is lasted about 4-6 weeks due to low rate of perishability at the refrigerator condition. Therefore, aim of this study is to develop a farmer-friendly low cost storing method to store lime for a considerable period in order to cope with the market situation particularly during off season as well as to impart an additional income for the down trodden citrus farmer.

II. MATERIALS AND METHOD

2.1 Determination of porosity of different types of bedding materials

Five types of bedding materials namely fine river sand (sieved with 2.8mm mesh), Course river sand, dry clay powder, granite powder and sea sand were taken and dried in a hot air oven at 105 ± 3^0C for 3hrs in order to remove presence of moisture and to kill micro organisms. Thereafter, dried bedding materials were filled up to the standard level of the cup in Apparent bulk density tester and carefully poured water at 20^0C until level of water reaches to the top level of the bedding material using a graduated measuring cylinder. The volume of water used to fill up the bedding material was recorded and porosity of the material was calculated "volume of water poured" divide by volume of the material. Finally it was expressed as a percentage. The same procedure was repeated for 10 times and average porosity of each material was calculated. The material with moderate porosity was selected as the best storing material because a material with the moderate porosity is capable to entrapped least amount of atmospheric O2 while facilitating to occur natural aeration within the sand mass at appreciable manner.

2.2 Selection of lime fruits

Same color, size and uniformly matured about 2500 lime fruits were obtained from trees in a same locality and sorted out visually while taking into account of color, size and without physical damage. About 1500 of limes were sorted out and to be kept for subsequence using of the study. In order to validate the uniform maturity of selected lime, a random sample was drawn and peels color (Greenish-yellow), thickness and diameter of the fruits were measured.

2.3 Preparation of treatments for lime storing

Eight treatment combinations were prepared by resorting two factor factorial experimental design using 3 variables namely charcoal, $NaHCO_3$ and $KMnO_4$ at two levels - low and high. Fine river sand was selected as the best bedding material because it has the moderately low porosity. Thus, 320kg of well dried and fine sand was taken and divided into two portions. One portion was mixed with charcoal at the ratio of 5g of charcoal per 100g of sand and the rest portion was kept untreated. Thereafter, the sand with charcoal portion was divided in to two portions again and one portion was mixed with $NaHCO_3$ at the ratio of 3g of $NaHCO_3$ per 100g of sand. The rest portion was kept without incorporation of $NaHCO_3$. The two portions coming out this exercise were divided in to two portions again and one portion of each was mixed with $KMnO_4$ at the ratio of 3g of $KMnO_4$ per 100g of sand. The rest two portions were kept without incorporation of $KMnO_4$. Similar procedure was adapted to the sand portion without incorporation of charcoal too. Prepared 8 treatments with sand were used to store lime fruits in 96 clay pots (12 pots for each treatment) and each pot contained 3 replicates of 15 numbers of limes (5 lime fruits for each replicate). Finally all clay pots were filled with the respective treatment of sand completely and stored in normal in-house condition (RH 76-80% and 26-28^{0}C). Samples were drawn from each treatment weekly until color of the lime peel turn into scant brown in order to determine changes occurrence on pH value and color of the fruit peel and biweekly to determine titrable acidity. Results obtained from the study were compared with a control treatment; which was also same lime, but stored under normal atmospheric condition without using sand.

2.4 Treatment combinations as per two factor factorial design

Treatment A - No Charcoal, No $NaHCO_3$, No $KMnO_4$

Treatment B - With Charcoal, No $NaHCO_3$, No $KMnO_4$ (Mixing was done at the ratio of 5g of charcoal per 100g of sand)

Treatment C - No Charcoal, With $NaHCO_3$, No $KMnO_4$ (Mixing was done at the ratio of 5g of $NaHCO_3$ per 100g of sand)

Treatment D - With Charcoal, With $NaHCO_3$, No $KMnO_4$ (Mixing was done at the ratio of 5g of charcoal and 3g of $NaHCO_3$ per 100g of sand)

Treatment E - No Charcoal, No $NaHCO_3$, With $KMnO_4$ (Mixing was done at the ratio of 3g of $KMnO_4$ per 100g of sand)

Treatment F - With Charcoal, No $NaHCO_3$, With $KMnO_4$ (Mixing was done at the ratio of 5g of charcoal and 3g of $KMnO_4$ per 100g of sand)

Treatment G - No Charcoal, With $NaHCO_3$, With $KMnO_4$ (Mixing was done at the ratio of 3g of $NaHCO_3$ and 3g of $KMnO_4$ per 100g of sand)

Treatment H - With Charcoal, With $KMnO_4$, With $NaHCO_3$ (Mixing was done at the ratio of 5g of charcoal, 3g of $NaHCO_3$ and 3g of $KMnO_4$ per 100g of sand)

Treatment I - Control treatment (Lime stored under normal atmospheric condition, without sand)

2.5 Determination of physical and chemical properties of stored lime

Physical properties namely color of the lime peel was measured using a food color chart while taking photographs from a digital camera (Canon EOS 700D) weekly. Chemical Property such as pH value was measured using HANNA bench top pH meter weekly while titrable acidity was determined biweekly by titrating 10ml of filtered lime juice with 0.1N NaOH using Phenolphthalein as the indicator. Results were expressed as gram of citric acid per100 ml of fruit juice.

III. RESULTS AND DISCUSSIONS

3.1 Selection of a suitable bedding material in storing of Lime

Suitability of five types of bedding materials namely river sand (sieved with 2.8mm mesh), coarse river sand, dry clay powder, granite powder and sea sand were determined in terms of bulk density and results were used to plot a bar chart "bulk density verses different types of bedding materials" as shown in figure 1

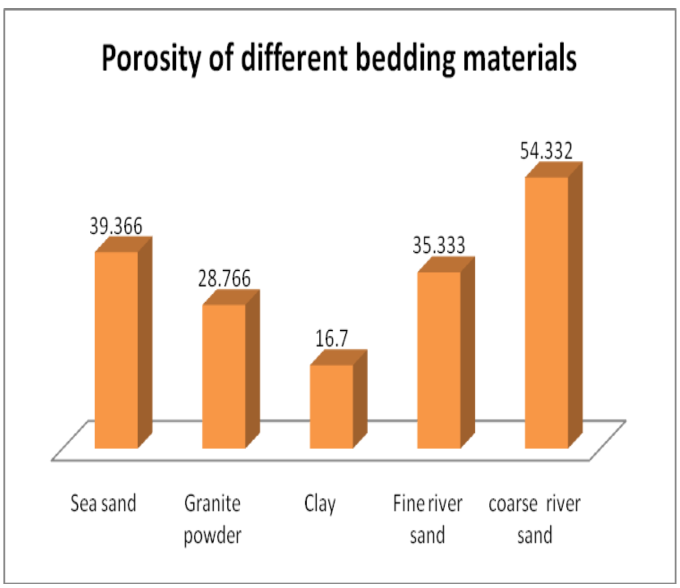

Figure1: Porosity of different types of bedding materials

The bar chart given in figure 1 clearly indicates that coarse sand having highest porosity around 50% and clay having least around 12%. The rest bedding materials are behaving in between, of which fine river sand having moderate porosity around 30% which is very important for natural aeration process within the sand mass itself; because, natural aeration process would facilitate to prevent anaerobic respiration as well as to flush out accumulated CO_2. Moreover, fine sand is capable to entrapped low amount of atmospheric oxygen. Thus, fine river sand was selected as the appropriate bedding material in storing of lime. Whereas, finding out of granite powder and sea sand is somewhat difficult in lime growing areas in Sri Lanka.

Porosity of a material usually depends on several factors that include packing density, the breadth, shape, and cementing property of the particles. For coarse river sand this has the highest porosity comparatively other bedding materials due to its large particle size and low cementing property. In the case of clay, this has the least porosity because of its highest cementing property. As far as fine river sand is concerned, this has the moderate porosity due to its low particle size and less cementing property. Similar finding has been reported by Nimmo, 2004 in variation of porosity of different types of soil materials.

3.2 Determination of de-greening process of lime stored in eight treatment combinations along with the control treatment

De-greening process of lime under 8 treatment combinations was monitored for period of 10 weeks and initial color of them was recorded. Results revealed that lime stored under fine river sand with charcoal and potassium permanganate ($KmNO_4$) was taken 10 weeks to complete the de-greening process as shown in figure 2. Thereafter, limes in this treatment gradually turned into brown color.

Treatment	Period of storage in week									
	1	2	3	4	5	6	7	8	9	10
Charcoal+ $KMnO_4$ + Sand										

Fig.2 De-greening process of lime stored under sand bed with charcoal and $KmNO_4$

The next best three treatments were lime stored under sand with Charcoal, sand with $KmNO_4$ and sand itself (without charcoal and $KmNO_4$) as these three treatments were capable to preserve lime from green color to yellow for 8, 7and 6weeks respectively. However, treatments with sodium bicarbonate as well as control treatment were taken least time to accomplish the de-greening process because lime in all of these treatments were turned into brown color within a matter of 2-3 weeks period.

Reason for this consequence is; lime is a non-climacteric fruit and produced least amount of ethylene during ripening and ethylene response for the ripening is also minimal. However, least response of lime for the ethylene causes to de-greening of the peel due to activating of chlorophyllase enzyme, which metabolized the chlorophyll pigments. Due to metabolizing of chlorophyll pigments, the masked yellow color (Carotenoid pigments) is gradually emerging from the lime peel and eventually skin turn into yellow color completely. Hence introduction of ethylene scrubbers into stored environment is an important aspect in order to control de-greening process of lime. Under optimal storage conditions, respiration rate of lime is <10mg CO2 Kg^{-1} h^{-1}. The rate of ethylene production is <0.1µl Kg^{-1} h^{-1} at 20^0C (Arpaia and Kader, 2000).

Moreover, usually fruits need adequate amount of oxygen to produce ethylene. If there is inadequate amount of oxygen in the stored environment, the product itself tends to use oxygen only for respiration rather than producing of ethylene. Hence, if presence of oxygen in the stored environment can be restricted, like storing fruits under fine sand bed, green life of lime can be extended. It has also been demonstrated by Grierson and Newhal, 1960 that oxygen is necessary for the de-greening action of lime because oxygen is an essential ingredient to produce ethylene from methionine amino acid. Formed ethylene trigger the activity of chlorophyllase enzyme to metabolize the green color pigments (Burns et al., 1995). Moreover, if stored environment contain ethylene scrubbers such charcoal and $KMnO_4$ green life of the produce can be extended.

Whereas, treatments stored under sand with $NaHCO_3$ had least shelf life because, de-greening process as well as conversion of peel color into brown was rapid. Reason for this consequence is liberating of CO_2 gas by $NaHCO_3$ and liberated CO_2 gas caused to anaerobic respiration that results in degradation of the green color of lime. Amir-Shappira et al., 1987 were also reported that lime is turning into brown color rapidly if they are stored under anaerobic condition.

3.3 Changes occurrence on pH value of lime stored under sand bed with different treatment combinations

Changes occurrence on pH value of lime stored under sand bed with 8 treatment combinations along with the control treatment were measured until peel color of the lime turn into brown. Results are given in the table 1.

Table1. Changing of pH value of lime stored in eight different treatment combinations

Treatment (Lime stored under sand bed with)	Period of storage (week)									
	1	2	3	4	5	6	7	8	9	10
Sand itself	1.9	2.00	2.04	2.10	2.28	2.53				
Charcoal	1.88	1.92	2.01	2.04	2.18	2.22	2.36	2.50		
$KMnO_4$	1.84	1.93	2.06	2.12	2.28	2.40	2.54			
Charcoal + $KMnO_4$	1.87	1.90	1.99	2.00	2.02	2.05	2.09	2.25	2.33	2.48
$NaHCO_3$	1.93	2.12	2.30	2.38	2.46					
Charcoal + $NaHCO_3$	1.91	2.11	2.22	2.36	2.47					
$KMnO_4$ + $NaHCO_3$	2.03	2.35	2.60							
Charcoal+$KMnO_4$ + $NaHCO_3$	2.02	2.09	2.28	2.62						
Control	2.04	2.25	2.58							

The data given in the table 1 clearly indicates that best treatment in storing of lime is, storing with charcoal and $KMnO_4$ because lime in this treatment takes more than 9 weeks to become the pH value 2.40 or above, because which is the pH value of already de-greened lime.

However, changing of pH value of lime fruit is directly related to the peel color. When fruit is in green, pH value remained in between 1.5 and 2.1 and it increases with the de-greening process. When the color is changed from green to yellow, pH value is also increased concurrently and finally it reaches to around 2.40. The deteriorated fruits (Fruits in brown color) having the highest pH values usually in between 2.50 and 2.65. Reason for this phenomenon is; when acid lime fruit is becoming to commercial maturity state, citric acid content is gradually increasing and it reaches to its maximum level when onset of ripening (Selvaraj and Raja, 2000). Thereafter, it slowly goes down with the beginning of de-greening process and as a result of that pH of the fruits is gradually moving up. Further, Ascorbic acid content in lime fruit is reached to its highest at commercial maturity state (58.08mg/100ml) and it decline to 53.0mg/100ml, after de-greening process (Ladaniya and Singh, 2000).

3.4 Changes occurrences on titrable acidity of lime stored under sand bed with different treatments.

Titrable acidity of lime stored under 8 treatment combinations along with the control treatment were measured fortnightly until peel color of the lime turn into brown. Results are given in the table 2.

Table 2 Titrable acidity level of lime (%) stored under eight treatment combinations

Treatment (Lime stored under sand bed with)	Period of storage (week)				
	2	4	6	8	10
Sand itself	0.7360	0.7040	0.5568		
Charcoal	0.7600	0.7360	0.7232	0.6592	0.5696
$KMnO_4$	0.7552	0.7232	0.6592	0.5504	
Charcoal + $KMnO_4$	0.7700	0.7424	0.7296	0.6720	0.5952
$NaHCO_3$	0.7040	0.6208	0.5695		
Charcoal + $NaHCO_3$	0.7296	0.6528	0.5888		
$KMnO_4$ + $NaHCO_3$	0.5184				
Charcoal+$KMnO_4$ + $NaHCO_3$	0.6976	0.5632			
Control	0.6720				

The data given in the table 2 shows that best two treatments were lime stored with charcoal or mixture of charcoal and $KMnO_4$ because these two treatments take more than 9 weeks to reach the acidity level 0.5696 and 0.5952 respectively. However, other treatments take comparatively lesser time to reach the affosaid acidity level. Reason for this consequence is also ethylene governed ripening process; of which acids in the lime fruit gradually and slowly tuned into neutral and thus, titrable acidity level becomes low (Gold Schmidt etal.,1993). But, since best two treatments contained ethylene scrubbing agents such as charcoal and $KMnO_4$ the acid degradation process becomes very slow and as a result of that limes in these two treatments contained high acidity level even after 9 weeks period of storage. Grierson et al., 1986 also reported that production of ethylene in lime causes for de greening process and it may be badly affecting for the declining of acid level.

IV. CONCLUSION

The most important quality indices in acid lime fruit is the dark green color. The green color is due to presence of chlorophylls in mature green fruit and these pigments are located in the flavedo of the feel and the juice vesciles. Maintenance of green or yellow peel color is required throughout the post harvest supply chain to attract a premium price. Therefore, results of this study revealed that lime can be preserved under modified atmospheric condition created in a fine river sand bed with charcoal and $KmNO_4$ at the ratio of 5g of charcoal and 3g of $KmNO_4$ per 100g of sand for a period of 9-10 week. If lime stored under sand bed either with charcoal or KmNO4 or sand itself (At the ratio of 5g of charcoal or 3g of KmNO4 per 100g of sand) green life of the produce can be maintained for about 8, 7 and 6 weeks respectively. The worst treatment in storing of lime with fine sand is, with $NaHCO_3$ because; Sodium bicarbonate causes to create an anaerobic environment within the sand mass itself.

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