A STUDY ON LOW COST PRESERVATION OF LIME JUICE

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ABSTRACT

Lime (*Citrus aurantifolia*) is a seasonal fruit and during the glut period much of the produce goes waste. This study was undertaken to develop a low cost method of juice preservation which could be adopted by medium scale processors. The composition of fresh lime juice was determined by chemical analysis. Pasteurized (77 °C for 60 seconds) and non-pasteurized juice were treated with different levels of Potassium metabisulfite (500 ppm, 1000 ppm, 1500 ppm and 2000 ppm) and stored in glass bottles at room temperature. Physico-chemical, microbiological and organoleptic characteristics were determined at monthly intervals for a period of 3 months. Data showed that total soluble solids (TSS) increased during storage while sulfur dioxide (SO_2) and ascorbic acid contents gradually decreased. Incidence of browning and development of off flavour and off odour were progressive and parallel after ten weeks of storage in both pasteurized and non pasteurized juice treated with 500 ppm SO_2. Further storage caused microbiological spoilage. A significant difference ($p<0.05$) was observed in colour, odour, bitterness and overall acceptance of the different juice samples. Non–pasteurized juice was found to be the most acceptable in terms of organoleptic evaluation. Thus suggesting that lime juice could be preserved for a period of about 10 weeks without any undesirable changes by using SO_2 at an initial level of 500 ppm.

KEY WORDS: Lime juice, Low cost, Orgenoleptic quality, Storage.

INTRODUCTION

Lime (*Citrus aurantifolia*) is an economically important, seasonal fruit available in plenty during April-July while only limited quantities are available during the off-season *i.e.* July-Sep and Feb-April. Fruits can be stored at room temperature for about 7 days without any undesirable changes. They can be stored for 6-8 weeks at 9-10 °C, 85-90 % RH (Murata, 1997), but the incidence of chilling injuries markedly shortens storage life.

Lime fruit is processed into sweetened/unsweetened juice, frozen concentrates, squashes, cordials, pickles, marmalades and jelly. The juice is used as a flavour enhancer in many food products. Commercially, lime juice is pasteurized at 90 °C for a few seconds and the hot juice is filled into cans. Storage in cans at 1.7° C extends the shelf life to 15 months without any change in flavour (Ranganna *et al.* 1983).

Due to the seasonal availability of the fruit, technology is required to extend the shelf life of the juice while maintaining quality. However, medium scale processors are unable to utilize modern technology as they require heavy inputs. Hence the requirement for a low cost preservation method. Chemical preservation of juice/pulp during glut is an accepted method in the fruit processing industry. This study was carried out to determine the minimum dose of SO_2 required to preserve lime juice and the variations in physico-chemical and microbiological qualities during storage.

MATERIALS AND METHODS

Fresh mature juicy lime fruits were selected, washed in cold water and disinfected using 200 ppm chlorinated water for 2 minutes and then washed again in cold water. Fruits were cut in halves, juice extracted and filtered through a two-fold muslin cloth to obtain clear juice. Physico-chemical properties of the fresh juice were determined.

Preservation of lime juice

The extracted clear lime juice was divided into two batches, and one batch pasteurized at 77°C for 60 seconds. The pasteurized and non- pasteurized juice were treated with 500, 1000, 1500 and 2000 ppm SO_2 using 867, 1734, 2601 and 3468 ppm potassium metabisulfite respectively, and stored for 3 months in pre-sterilized glass bottles at room temperature. Completely Randomized design (CRD) was used with four replicates.

Determination of physico-chemical properties of lime juice during storage

Total soluble solids (TSS), titrable acidity expressed as % citric acid, ascorbic acid content and colour of juice were determined just before storage. Thereafter the tests were repeated at monthly intervals for a period of 3 months.

Determination of Sulfur Dioxide content

Sulfur Dioxide content of the stored samples was analyzed at monthly intervals using titrimetric method (James, 1995).

Microbial examination of the juice

Microbiological analysis was done at monthly intervals using pour plate method (AOAC, 1984) with both pasteurized and non-pasteurized juice treated and stored with an initial level of 500 ppm and 1000 ppm SO_2.

Evaluation of Organoleptic quality of juice

During the first two months of strorage, organoleptic quality of juice was assessed in the 500 ppm and 1000 ppm SO_2 treated pasteurized and non-pasteurized batches. In the third month, organoleptic quality evaluation was done only in the 1000 ppm SO_2 treatments.

Sample preparation

Lime juice was diluted (1:10) and brix was increased to a constant value (15°Brix) by mixing with ground sugar. Organoleptic evaluation was carried out immediately after sample preparation.

Presentation of samples

Organoleptic evaluation was carried out by thirty untrained panelists. It was repeated at monthly intervals during the three-months storage period. Panelists were provided with a scorecard and were requested to evaluate intensity of colour, odour, sweetness, bitterness and sourness of the presented lime juice and rank the samples for acceptance.

Analysis of data

Main factor effects and interactions were determined using a two- factor Analysis of Variance (ANOVA) model Statistical Analysis System (SAS). Duncan's Multiple Range Test (DMRT) at 5 % probability level was carried out for mean separation. Sensory analysis data were analyzed using non- parametric Friedman test using MINITAB statistical package (Watts *et al.* 1989).

RESULTS AND DISCUSSION

Physico-chemical analysis of fresh and pasteurized lime juice

Both TSS and TA had significantly ($p < 0.05$) increased after pasteurization, while Brix/Acid, ph and ascorbic acid content had significantly ($p < 0.05$) declined (Table 1). The significant difference in TSS, TA, ph and ascorbic acid between pasteurized and fresh juice can be attributed to the effect of heat treatment.

Table 1. Physico-chemical constituents of fresh and pasteurized lime juice

Constituent	Fresh lime juice Mean ± SD	Pasteurized lime juice Mean ± SD
Juice (%)	40.00± 3,30	-
TSS (°Brix)	7.59± 0,12	8,34± 0.17
TA (% citric acid)	7.04± 0.26	8.00± 0.25
Brix/Acid	1.07± 0.17	1.04± 0.04
pH	2.00 ± 0.01	1.99 ± 0.01
Ascorbic acid (mg/100ml)	27.23 ± 0.44	24.50 ± 0.33

Cloud loss or clarification was visible in the non- pasteurized juice but was observed to a very low extent in the pasteurized juice (Table 2).

Table 2. Initial colour of the fresh and pasteurized lime juice.

Treatment	(RHC Chart)
Non pasteurized juice	Yellow-Green group 154 C (Clear)
Pasteurized juice	Grayed-Yellow group 160 C (Cloudy)

Physico-chemical properties of lime juice during storage
Total soluble solids (TSS)

Initially, there was a significant difference ($p< 0.05$) in TSS in both pasteurized and non-pasteurized juice at different levels of potassium metabisulfite (KMS) (Table 3). There was no significant difference ($p< 0.05$) in TSS content in the different treatments at any stage of storage. However, the pasteurized juice treatments showed a significant difference ($p < 0.003$) after 3 months of storage.

Table 3. Changes in Total Soluble Solids content of juice during storage at room temperature (29 ± 10 °C) *.

Treatment (SO_2 ppm.)	TSS content (0 Brix) Storage time in months			
	0	1	2	3
Pasteurized juice				
500	8.34 b	8.59 a	9.06 b	9.91 b
1000	8.59 a	8.72 a	9.53 ab	9.91 b
1500	8.59 a	8.97 a	9.73 a	10.03 b
2000	8.59 a	8.97 a	9.91 a	10.35 a
Non pasteurized juice				
500	8.09 b	8.34 a	8.65 a	9.16 a
1000	8.34 a	8.59 a	8.59 a	8.85 b
1500	8.34 a	8.59 a	8.47 a	9.03 ba
2000	8.34 a	8.59 a	8.78 a	9.03 ba

* Treatment means in a column with a common letter are not significantly different by DMRT at 5%. Each data point represents average of samples.

Sudden increase of TSS in lime juice stored with an initial level of 500 ppm of SO_2 after 3 months, may be due to microbial fermentation and chemical reactions.

Titratable acidity (TA)

After incorporation of potassium metabisulfite, TA increased in both pasteurized and non-pasteurized juice when compared to that in fresh juice. However, TA in the non-pasteurized juice remained relatively lower than in pasteurized juice throughout the storage period in all treatments.. Significant difference ($p < 0.05$) in TA was observed in both pasteurized and non-pasteurized juice at different levels of potassium matabisulfite at the initial stage of storage. Statistically the % TA was not significant ($p < 0.05$) in the non- pasteurized juice after one month whereas in the pasteurized juice it was significant ($p < 0.03$) after the second and third months of storage (Table 4).

Table 4. Titratable Acidity of Pasteurized and Non Pasteurized juice after storage at room temperature (29 ± 10ºC)*

Treatment SO_2 ppm	Titratable Acidity (% Citric Acid)** Storage time (months)			
	0	**1**	**2**	**3**
Pasteurized juice				
500	7.90 d	8.08 ab	8.29 a	8.55 a
1000	8.13 a	7.84 b	8.29 a	8.29 ab
1500	8.10 c	8.15 ab	7.84 b	8.19 b
2000	8.12 b	8.36 a	8.16 a	8.05 b
Non pasteurized juice				
500	7.00 d	6.96 a	6.88 a	8.19 a
1000	7.12 c	7.02 a	6.87 a	8.04 a
1500	7.23 a	7.09 a	7.34 a	7.84 a
2000	7.17 b	7.07 a	6.97 a	7.93 a

*Treatment means in a column with a common letter are not significantly different by DMRT at 5%.
** Values represent average of four replicates

Ascorbic acid

Ascorbic acid content gradually decreased during storage. After three months of storage, retention of ascorbic acid in non-pasteurized juice was higher than in the pasteurized juice. Loss of ascorbic acid was higher in juice stored with an initial level of 500 ppm SO_2 when compared to juice stored with an initial level of 2000 ppm SO_2 (Table. 5). This loss could be due to oxidative deterioration during heat treatment and storage (Johnson *et al.*, 1995; Lee and Chen, 1998; Kanner *et al.* 1982). Retention of ascorbic acid in juice increased with increasing concentration of added potassium matabisulfite. This may be due to the stabilization effects of the chemical.

Table 5. Ascorbic acid content of juice after 3 months of storage

Treatment (SO_2 ppm)	Ascorbic acid content (mg per 100 ml) * Pasteurized juice	Non-pasteurized juice
500	3.45 ± 0.01**	5.05 ± 0.00
1000	4.02 ± 0.19	5.73 ± 0.29
1500	6.10 ± 0.30	8.16 ± 0.21
2000	10.34± 0.18	15.49 ± 0.32

*Average of four replicates
** (Mean ± SD)

Sulfur dioxide

A gradual reduction of the sulfur dioxide content was observed during storage at all levels of added potassium metabisulfite. The 500 ppm level recorded the lowest value at the end of 3 months of storage (Table 6).

Table 6. Changes in sulfur dioxide content in juice during storage

Initial Level of SO_2 (ppm)	Pasteurized juice				Non-pasteurized juice			
	Storage time (months)							
	0	1	2	3	0	1	2	3
500	484	400	300	100	500	420	320	140
1000	960	760	640	440	1000	800	700	560
1500	1492	1260	1120	960	1500	1360	1240	1120
2000	1980	1820	1700	1420	2000	1860	1760	1600

Microbiological studies

Growth of microorganisms was not detected during the first two months and this may be due to the antimicrobial action of potassium matabisulfite. However, after three months of storage, a few bacterial and fungal colonies were visible in plates inoculated with juice treated with an initial level of 500 ppm SO_2 (Table 7). Further storage , growth of microorganisms was visible at the storage of the juice and they were identified as *Penicillium spp* and *Aspergillus spp.* Also microbial fermentation does not occur in lime juice because of its low sugar content and high acidity. The growth of fungi at the surface of the juice may be due to the aerobic nature of treatment.

Table 7. Observations of microbiological studies conducted after 3 months of storage

Treatment	No. of visible organisms per ml juice
Pasteurized juice with initial level of 500 ppm. SO_2.	3×10^1
Non–pasteurized juice with initial level of 1000 ppm. SO_2	4×10^1

Organoleptic evaluation

According to the Friedman test there was a significant difference ($p < 0.05$) in terms of colour, odour, bitterness, sourness and overall acceptance of juice after one month of storage (Table 8).

Table 8. Summary of sensory analysis of juice after one month of storage

Initial level of SO_2 (ppm)		Sensory property colour		odour		bitterness		sweetness		sourness		acceptance	
NPJ	Code	EM	SR	EM	SR	EM	SR	EM	SR	EM	SR	EM	SR
500	670	1	48	1	46	1.7	57.5	3	83	3	66.5	!	46
1000	256	1	48	1	64	2	76	3	75.5	3	74	2	60
PJ		2	102	2	102	2	77.5	3	67.5	3	76	3	84
500	831	2	102	2	101	2.3	89	3	74	3	83.5	4	110
1000	349	p= 0.00		p= 0.00		p= 0.00		p= 0.115		p= 0.045		p=0.00	

NPJ= Non pasteurized juice PJ= Pasteurized juice EM = Estimated median
SR= Sum of rank p=Probability.

Similarly there was a significant difference ($p< 0.05$) in terms of colour, odour, bitterness, sweetness, and overall acceptance of juice samples after two and three months of storage (Tables 9 and 10).

Table 9. Summary of sensory analysis of juice after two months of storage

Initial level of SO_2 ppm.		Sensory property											
		Colour		Odour		Bitterness		Sweetness		Sourness		Acceptance	
NPJ	Code	EM	SR	EM	SR	EM	SR	EM	SR	EM	SR	EM	SR
500	670	1.1	48	1.5	59	1.4	55	3	74	2	75.5	1.4	53
1000	256	1.4	74	1.8	66	1.9	74	3.3	87	2	64	2	60
PJ		2	85	2.5	92.5	2.1	85	2.8	59	2	81.5	2.9	88
500	831	2	93	2.2	82.5	2.1	85.5	3	79	2	74	3.8	98
1000	349	p= 0.00		p= 0.00		p= 0.00		p= 0.01		p= 0.461		p= 0.00	

NPJ= Non pasteurized juice PJ= Pasteurized juice EM= Estimated median
SR= Sum of ranks p= Probability.

Table 10. Summary of sensory analysis of juice after three months of storage

Initial level of SO_2 ppm		Sensory property											
		Colour		Odour		Bitterness		Sweetness		Sourness		Acceptance	
NPJ	Code	EM	SR	EM	SR	EM	SR	EM	SR	EM	SR	EM	SR
1000	256	1	32	3	38	1	35.5	3	54	2	38.5	1	33
PJ		2	58	3	52	2	54.5	2	36	3	51.5	2	57
1000	349	p= 0.00		p=0.00		p=0.00		p=0.00		p= 0.05		p=0.00	

NPJ=Non-pasteurized juice. PJ= Pasteurized juice EM= Estimated median
SR= Sum of ranks p=Probability

Ranking test for overall acceptance indicated that panelists prefer non-pasteurized juice to pasteurized juice. Out of four samples (831, 349, 670, 256), it was evident that the panelists preferred the non-pasteurized lime juice, treated with an initial level of 500 ppm SO_2 in the first and second months. In the third the preference was for non-pasteurized juice treated with an initial level of 1000 ppm SO_2. Pasteurization had enhanced the flavor of the juice thus resulting in the low acceptance .

CONCLUSIONS

Use of high levels of potassium metabisulfite (1500 and 2000 ppm SO_2) to preserve ascorbic acid in stored lime juice is successful to a considerable extent. Non-pasteurized lime juice is the more acceptable than pasteurized juice in terms of organoleptic properties. Lime juice with an initial level of 500 ppm SO_2 can be stored in glass bottles at 29 ± 1°C for ten weeks without any undesirable organoleptic and other quality changes.

ACKNOWLEDGMENT

The authors gratefully acknowledge Mr. H.C.G.K. Jayarathne for typing the manuscript.

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