

## ASCORBIC ACID RETENTION IN CANNED LIME JUICE PRESERVED WITH SULFUR DIOXIDE AND BENZOIC ACID

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### ABSTRACT

The effects of two levels each of sodium metabisulfite and sodium benzoate on the shelf-life of canned lime juice stored at ambient temperature was based on ascorbic acid degradation as an index. Sodium metabisulfite increased the shelf-life significantly ( $P < 0.05$ ) compared to sodium benzoate, whose effect was not significantly different from that of the control sample. Increasing sodium metabisulfite concentration from 150 ppm to 300 ppm had a significant stabilizing effect ( $P < 0.05$ ) on ascorbic acid although it did not prevent completely its

destruction during storage. Doubling sodium benzoate concentration from 150 ppm to 300 ppm had no significant increase in ascorbic acid stability. From linear regression calculations, 300 ppm sodium metabisulfite resulted in the longest shelf-life of 49 weeks while the control samples had the shortest shelf-life of 22 weeks.

**Key words:** *Ascorbic acid stability; Citrus aurantifolia; lime juice, sodium metabisulfite, sodium benzoate, shelf-life.*

### INTRODUCTION

Lime (*Citrus aurantifolia*) fruits are among the group of citrus fruits which are desired largely for their highly acidic juice. From a nutritional point of view, lime juice is an excellent source of ascorbic acid (vitamin C) [1]. Lime juice is also a moderate source of vitamin A and the minerals calcium, phosphorus, iron and potassium [2]. Ascorbic acid stability in citrus fruit juices, particularly orange juice during processing and storage has been studied extensively [3-5]. However, information on ascorbic acid stability in other citrus fruit juices is limited. Since the quality of lime juice is greatly affected by the method of extraction and preservation [6], it is important that the juice is processed, packaged and stored under conditions that maximize ascorbic acid retention.

In order to facilitate preservation and distribution, it is a technological practice to

package juices in metal cans, glass bottles or plastic containers. Although metal cans are expensive and require sophisticated machinery for container closure [7], they have been shown to effect ascorbic acid retention better than other packaging materials [8,9]. However, packaging alone cannot preserve the quality of juice [10], and in addition, therefore, juices are treated with chemical preservatives.

In Kenya, like in many developing countries in the tropics, fruit juices are preserved with chemicals mainly to prevent microbial spoilage during storage, both in the retail stores and consumer homes. Some of these chemicals are thought to have protective effects on ascorbic acid [8, 11-13]. While a large number of chemicals with preservative effects have been described, only a relatively small number is allowed for direct use in human food. Moreover, even use of these chemicals is regulated by the maximum allowable effective levels [14,15].

Sulfites and metabisulfites of sodium or potassium are added to fruit juices as potential sources of sulfur dioxide, which acts

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as an antimicrobial agent and also stabilizes ascorbic acid [11]. Use of metabisulfite providing up to 400 ppm of sulfur dioxide in orange juice has been reported [8,13]. However, such high levels of sulfur dioxide are likely to impart a characteristic pungent smell to fruit juices. The action of sulphur dioxide as an antimicrobial agent as well as a stabilizer of ascorbic acid depends on the pH of the food. It is, therefore, important to test its efficacy in a high acid juice such as lime juice. On the other hand, benzoic acid is also used in fruit juices and other acid products for its antimicrobial activity. Since the two chemicals exert either one or both of synergistic or concerted actions, it is possible that benzoic acid makes a significant contribution to protection of ascorbic acid.

This study was, therefore, designed to investigate the protective effects of sulfur dioxide and benzoic acid on ascorbic acid in canned lime juice stored under the conditions prevailing in retail shops and consumer homes in most parts of Kenya.

## MATERIALS AND METHODS

### *Juice processing*

Freshly harvested lime (*Citrus aurantifolia* cv. Tahiti) fruits were obtained from the Research Farm of Jomo Kenyatta University of Agriculture and Technology, Kenya. The fruits were washed in distilled water, peeled, cut into halves and the juice expressed using a hydraulic press. The expressed juice was passed through a single layer of muslin cloth to filter out the seeds, solids and pulp material, and the total solids content in the juice was adjusted to 14° Brix using refined sugar. The juice was pasteurized at 82°C for 15 min and while still hot, it was divided into five batches which were treated as follows: Batch I- No chemical treatment (control); Batch II- 150 ppm sodium metabisulfite; Batch III- 300 ppm sodium metabisulfite; Batch IV- 150 ppm sodium benzoate; Batch IV- 300 ppm sodium benzoate. The hot juice (about 70°C) was then poured into 303 x 406 cans (CMB Kenya Ltd.)

leaving a headspace of 1.5 cm and the cans were sealed using an O-type vacuum seamer (Hashimoto Canning Research Institute Ltd., Tokyo, Japan). The cans were then cooled in running water to about 23°C.

### *Ascorbic acid determination*

On the day of preparation and at two-week intervals, triplicate determinations of ascorbic acid were performed by visual titration with 2,6-dichlorophenolindolphinol [16].

### *Shelf-life stability evaluation*

Juice cans were stored on a laboratory bench at ambient temperature ( $23 \pm 2^\circ\text{C}$ ) simulating retail and consumer home conditions. Initially, then after every two weeks, the juice was analyzed for ascorbic acid content up to 16 weeks of storage. Mean separation by Duncan multiple range test was carried out to determine if there was any significant difference at the 5% level of significance between the treatments. Independent regression analysis was carried out to predict the ultimate shelf-life of each juice treatment in terms of ascorbic acid depletion.

## RESULTS AND DISCUSSION

Consumers consider citrus fruits as one of the best sources of ascorbic acid in their diet. The fruits are, however, seasonal and, therefore, their juices are becoming the main source of the vitamin. From a nutritional point of view, therefore, lime juice is an excellent source of ascorbic acid. Hence, it is important that the juice is processed, packed and stored under such conditions that ascorbic acid retention is maximized. The initial ascorbic acid content in the processed juice and the changes observed during storage for 16 weeks are shown in Table 1. The lowest ascorbic acid loss occurred in juice containing 300 ppm sodium metabisulfite. Loss of ascorbic acid in juice containing 300 ppm sodium benzoate was in the same range as the loss from juice containing 150 ppm sodium metabisulfite. Ascorbic acid loss in the control juice was the

highest followed by that from the juice containing 150 ppm sodium benzoate. It has been reported that sodium metabisulfite stabilizes ascorbic acid in other citrus juices for long periods of storage [8,11,13]. Although sodium metabisulfite and sodium benzoate are equally effective as antimicrobial agents, the results from this study indicate that the stabilizing effect of sodium metabisulfite on ascorbic acid was better than that of sodium benzoate. There was a general decrease in ascorbic acid content in all treatments during the entire storage period. At the end of the 16 weeks of storage, the control juice retained only 29% of its original ascorbic acid content, while the samples containing 150 ppm and 300 ppm sodium metabisulfite retained 52% and 64%, respectively, and the samples containing 150 ppm and 300 ppm sodium benzoate retained 38% and 48%, respectively. At higher sulfur dioxide levels (1000 ppm), there is only a slight decrease in ascorbic acid content [11]; however, such high levels of the chemical are likely to impart a pungent odor to the product [8]. Moreover, FDA has proposed a maximum residual sulfur dioxide level of 300 ppm in fruit juices [17]. The initial degradation of ascorbic acid could presumably have been due to oxidation by oxygen dissolved in the product or that contained in the headspace [[10,18] while subsequent decomposition could be attributed to a combination of oxygen penetration into the package probably due to poor closure and product specific decomposition [5,10,13,18-20]. This may lead to oxidation of ascorbic and dehydroascorbic acids to the biologically inactive forms. Although the degradation of ascorbic acid is known to be affected by pH, the change in pH during the storage period was not significant (data not shown). In addition, the degradation of ascorbic acid could have been due to anaerobic degradation which has been reported as the most common mechanism in canned fruit juices and the presence of certain sugars (e.g. sucrose at low pH) which are known to increase the rate of anaerobic degradation of ascorbic acid [21].

On subjecting the data to a two-way analysis of variance [22], we found that sodium metabisulfite had a significant stabilizing ( $p = 0.05$ ) effect on ascorbic acid (Table 2). Doubling sodium metabisulfite level significantly ( $p = 0.05$ ) increased protection on ascorbic acid. No significant ( $p = 0.05$ ) stabilizing effect was observed between the two levels of sodium benzoate used. The predicted shelf-life was defined as the time it takes for complete exhaustion of ascorbic acid in the container package. The shelf-life depended on the levels of the stabilizers. These observations are in close agreement with reports by other investigators [8,11,13]. Preliminary experiments indicated that in the absence of added stabilizers, the shelf-life of canned lime juice at ambient conditions is limited to 19 weeks [9]. Ascorbic acid content in processed citrus fruit juices depends to a large extent on the storage time and temperature and the level and type of stabilizer used [8,23,24]. Thus, although sodium metabisulfite and sodium benzoate are both antimicrobial agents, sodium benzoate does not seem to protect juices against ascorbic acid losses, at least, not in lime juice. Due to lack of refrigerated facilities both at the retail shop and in consumer homes in most developing countries, it seems that sulfites are still the chemical of choice in the preservation of ascorbic acid in most fruit juices. The correlation coefficients shown in Table 2 are high suggesting that the best lines of fit are suitable for predicting the shelf-life of fruit juices. This, as pointed out by Squires and Hanna [3] and Maeda and Mussa [8] could be a useful guide to manufacturers for declaration of ascorbic acid shelf-life on the package label, thereby, enhancing consumer confidence in their products. Since ascorbic acid is a labile nutrient, its loss in processed juices can be used as an index of the levels of other nutrients in juices held at ambient temperature during retail marketing.

**Table 1**

*Effect of storage time on the mean ascorbic acid content in canned lime juice preserved with sodium metabisulfite and sodium benzoate*

| Treatment        | Time (Weeks) | Ascorbic acid content (mg / 100 ml juice) <sup>a</sup> |          |          |          |          |                 |          |          |          |
|------------------|--------------|--|----------|----------|----------|----------|-----------------|----------|----------|----------|
|                  |              | 0  | 2        | 4        | 6        | 8        | 10              | 12       | 14       | 16       |
| Control          |              | 26.6±1.2   | 21.3±2.1 | 19.0±0.8 | 16.8±1.1 | 13.3±0.7 | ND <sup>b</sup> | 10.0±0.3 | 8.9±0.5  | 7.7±0.8  |
| S M <sup>c</sup> | 150 ppm      | -  | 24.8±2.6 | 22.1±1.7 | 20.9±1.4 | 20.2±1.0 | ND              | 16.0±0.6 | 14.2±1.2 | 13.6±0.8 |
|                  | 300 ppm      | -  | 25.2±1.9 | 24.5±2.5 | 22.8±0.9 | 23.2±1.3 | ND              | 20.3±2.1 | 18.6±1.4 | 17.0±1.2 |
| S B              | 150 ppm      | -  | 23.4±1.2 | 21.0±2.0 | 19.8±1.4 | 16.9±0.7 | ND              | 13.8±1.1 | 12.7±0.9 | 10.0±0.5 |
|                  | 300 ppm      | -  | 23.8±1.9 | 22.4±0.8 | 20.6±2.6 | 18.2±1.3 | ND              | 14.6±1.0 | 13.8±0.8 | 12.4±0.9 |

<sup>a</sup> Values are means ± SD of three replications.

<sup>b</sup> ND: Not determined.

<sup>c</sup> SM: Sodium metabisulfite; SB: Sodium benzoate.

**Table 2**

*Ascorbic acid depletion rates and probable shelf-life (weeks) as predicted from independent linear regression equations.*

| Treatment            | Correlation coefficient (r) | Ascorbic acid depletion rate in mg/100 ml of juice per week (slope) | Number of weeks to attain 'zero' ascorbic acid content | Mean ascorbic acid content over the storage period |
|----------------------|-----------------------------|---|--|--|
| Control              | -0.972                      | 1.11  | 22   | 15.5a  |
| Sodium metabisulfite |                             |   |  |  |
| 150 ppm              | -0.991                      | 0.82  | 32   | 19.8b  |
| 300 ppm              | -0.984                      | 0.57  | 49   | 22.3c  |
| Sodium benzoate      |                             |   |  |  |
| 150 ppm              | -0.989                      | 0.96  | 27   | 18.0b  |
| 300 ppm              | -0.988                      | 0.87  | 30   | 19.1b  |

Means subscripted by the same letter are not significantly different ( $p = 0.05$ ). Mean separation by Duncan multiple range test.

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