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STUDIES ON PASTEURIZED AND CONCENTRATED LIME JUICE

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ABSTRACT

Lime juice is considered a rich source of many bioactive compounds which have numerous health benefits. The quality of the juice changes under the influence of processing and subsequent storage. The present study aimed to investigate the chemical composition, microbiological and sensory evaluations of fresh and pasteurized lime juice and its concentrates (30° and 50° Brix) stored at 4.0±1.0°C for eight weeks. Ascorbic acid, titratable acidity, pH value, reducing and non reducing sugars, free amino nitrogen, browning index, in addition to sensory evaluation (taste, colour, flavour, overall acceptability) varied with storage time. Gradual decreases in reducing and non reducing sugar, ascorbic acid, taste, colour, flavour and overall acceptability were observed in all stored samples of lime juice, the retention of ascorbic acid after storage at 4±1°C for eight weeks was 56.14% for fresh lime juice, 53.53% for pasteurized lime juice, 51.57% and 66.83% for reconstituted juice prepared from lime juice concentrate (30° and 50°Brix), respectively. During storage, there was a continuous increase in browning index in fresh, pasteurized and reconstituted juices. Moreover, an increase in titratable acidity was observed in fresh lime juice. There was a slight increase in free amino nitrogen content in fresh and pasteurized lime juice during storage time. pH value was increased in pasteurized and reconstituted juices prepared from lime juice concentrates (30° and 50° Brix) during storage time. As for the microbiological evaluation, most of lime juice samples showed microbiological count less than 30 CFU/ml, whereas the other samples did not contain any microorganisms, so it is considered not detected. Sensory evaluation of lime juices revealed that fresh, pasteurized, and reconstituted juice prepared from lime juice concentrate of 30° Brix had acceptable taste and colour up to eight weeks storage, but reconstituted juice prepared from lime juice concentrate of 50° Brix had acceptable taste up to the fourth week and acceptable colour up to the sixth week. The score of flavour was the highest in fresh and pasteurized lime juice up to the end of storage time. From the overall acceptability, all samples of lime juice showed decrement of overall acceptability during storage time.

Key words: Lime juice, pasteurization, concentration, quality characteristics and cold storage.

INTRODUCTION

Lime fruit is a member of Citrus fruits belonging to the family (*Rutacea*) which are an important source of antioxidants such as ascorbic acid, carotenoids, flavonoids, and other phenolic compounds (Abeysinghe *et al.*, 2007; Ghasemi *et al.*, 2009 and Rapisarda *et al.*, 2008).

Lime fruit (*Citrus aurantifolia*) is rich in vitamin C, a natural antioxidant, important for

boosting the immune system, protecting against colds and many bacterial illnesses. Additionally, the juice can do wonders to the body and it can relieve a person from numerous diseases, so lime has several of health benefits include weight loss, skin care, good digestion, eye care, peptic ulcer, respiratory disorders, gout, gums, urinary disorders, *etc.* (Abeysinghe *et al.*, 2007; Ghasemi *et al.*, 2009 and Rapisarda *et al.*, 2008).

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The annual world production of lemon and lime fruits in 2011 was reported by FAO (2013) to be 13.861.411 tonnes, of which 296.773 tonnes was produced in Egypt.

The quality of lime juice changes continuously under the influence of technological processes and storage time. Al-Zubaidy and Khalil (2007) investigated the kinetics of the loss of ascorbic acid in local lemon juice of 9° and 50° Brix stored at 25, 35 and 45°C for 4 months. The results indicated that increases of concentrations and temperatures increased the rate of ascorbic acid degradation.

El-Wakeil et al. (1986) observed a slight decrease in total acidity of lime juice due to thermal treatment in both pasteurized and concentrated juices. Whereas, the pH values were almost always constant, which could be attributed to the buffering action of the juice. During storage, gradual increases in total acidity was observed almost in all stored concentrates. They also revealed that there was a slight increasing trend in reducing sugars of lime juice due to pasteurization of lime juice concentrate. A corresponding decreasing pattern was also noticed concerning to the non-reducing sugars. However, the total sugars were almost always the same. Moreover, no appreciable changes in sugars contents due to processing techniques could be observed. Meanwhile, there was a slight increase in reducing sugars. Regarding storage, there was a slight continuous increase in reducing sugars in concentrated lime juice. They also found that there was a slight increase in free nitrogen content directly amino after pasteurization of lime juice. This could be attributed to the decrease in soluble non amino nitrogen substances due to heat treatments. On the contrary, a slight decrease was noticed in amino nitrogen content due to thermal concentration during processing.

Gherardi *et al.* (1992) found that unpasteurized orange juice immediately showed complete pulp separation and was spoiled after 5 days at 10° C and after 21 days at 2° C. Juice prepared by recombination of pasteurized pulp and clear serum had steady turbidity and satisfactory sensory properties; microbial spoilage was observed after 10 days of storage at 10° C.

Balaban (2000) decided that the heat process is effective but, has a negative effect on the quality of the orange juice. Pasteurization not only kills microorganisms, but also, changes the flavour of the juices and the juice acquires a slightly cooked flavour. The shelf life of fresh squeezed orange juice under the best circumstances is about 14 days whereas, the shelf life of thermally processed, heat pasteurized orange juices is about 2 months.

The aim of the present study was to evaluate impact of some technological processes on physico-chemical, microbiological and sensory properties of lime juice products during storage.

MATERIALS AND METHODS

Materials

Fresh lime fruits (*Citrus aurantifolia*, Banzaheir variety) used in this study were obtained from El-Obour market, Cairo, Egypt, during September, 2011. Lime fruits were washed with tap water. The fruits were cut into two halves and the juice was extracted by hand reaming. The juice was strained through two layers of cheese cloth. The yield of fresh lime juice and its total soluble solids were 40-42% and 9.0°Brix, respectively.

The juice was divided into four parts, the first part was fresh lime juice that was packed in sterilized glasses bottles (250 ml), caped and kept at $4.0\pm1.0^{\circ}$ C. In the second part, pasteurized lime juice, the juice was filled in sterilized glasses bottles (250 ml) and put in water bath at 85°C for 5 min (Shouman, 1974). The bottles were cooled directly using tap water and stored at $4.0\pm1.0^{\circ}$ C. The third and fourth part were concentrated in a rotary evaporator RE 300 (Stuart Rotary evaporators) under vacuum at 40°C to 30° and 50° Brix, then the concentrates were packed in sterilized glass bottles (250 ml), caped and kept at $4.0\pm1.0^{\circ}$ C.

Methods

Total soluble solids were determined using a digital refractometer (Leica Mark II) at 20°C, titrtable acidity, pH value and free amino nitrogen were determined according to the method reported by AOAC (2007). Total and reducing sugars were determined according to phenol sulphuric acid method, non reducing sugars was calculated by difference between total and reducing sugars as described by Dubois

et al. (1956). Ascorbic acid content was determined using 2,6 dichlorophenol indophenol dye according to the method reported by Ranganna (1979). The results were expressed as mg/100ml juice. Browning index was measured according to Ranganna (1979).

Microbiological Analyses

Total count of bacteria, yeasts & moulds and acidophilic bacteria were determined according to the methods recommended by the American Public Health Association (1958). Spore forming bacteria was determined according to American Society for Microbiology (1982).

Sensory Evaluation

Lime juice samples were evaluated by 10 experienced panelists. The panelists were asked to give score from 10. The score 10 = excellent, 9 = highly acceptable, 8 = moderately acceptable, 7 = slightly acceptable and 6 =unacceptable (Khater, 2003)

RESULTS AND DISCUSSION

Ascorbic Acid

The highest ascorbic acid content was found in fresh lime juice (23.669mg/100ml), compared to pasteurized (23.554 mg/100ml) and the reconstituted juices prepared from lime juice concentrates of 30° and 50°Brix were 20.680 and 21.480 mg/100 ml, respectively. The results indicated that the increase of concentrations and temperatures increased the rate of ascorbic acid degradation especially after 6 weeks of storage (Al-Zubaidy and Khalil, 2007). Fig. 1 shows the content of ascorbic acid of lime juice samples during storage for 8 weeks at 4.0±1.0°C. It can be observed that ascorbic acid decreased with increasing storage time and retention of ascorbic acid in those samples was 56.145% for fresh lime juice, 53.536% for pasteurized lime juice, 51.595% for reconstituted juice prepared from lime juice concentrate of 30°Brix and 66.834% for reconstituted juice prepared from concentrate of 50°Brix. The retention of ascorbic acid in fruit juices decreased during storage, depending on storage conditions, such as temperature, oxygen and light access (Al-Zubaidy and Khalil. 2007; Kabasakalis et al., 2000 and Zerdin et al., 2003).

Titratable Acidity and pH Value

The titratable acidity (expressed as % anhydrous citric acid) and pH value of fresh. pasteurized and concentrated lime juice at 30 and 50°Brix during storage time are shown in Figs. 2 and 3, respectively. Titratable acidity of fresh lime juice at zero time was 8.064% and increased to 9.600% at the end of storage time, the pH value of fresh lime juice at zero time was 2.351 and decreased to 2.224 after storage time. This indicates that during storage, the pH value decreased gradually, while titratable acidity increased gradually. These results are in agreement with the range of data obtained by Ziena (2000). On the contrary, titratable acidity of pasteurized lime juice and reconstituted juices prepared from lime juice concentrates (30° and 50° Brix) were decreased gradually during storage, from 8.000, 8.550 and 8.576%, respectively at zero time to 7.923, 8.320 and 8.384% respectively at the end of storage time. Also, pH value of pasteurized lime juice and reconstituted juices prepared from lime juice concentrates (30° and 50° Brix) were increased gradually during storage, from 2.360, 2.255 and 2.251, respectively, at zero time to 2.371, 2.282 and 2.277 at the end of storage time.

Sugars

Figs. 4, 5 and 6 show the contents of reducing, non reducing and total sugars of fresh, pasteurized and concentrated lime juices during storage time.

Reducing sugars of fresh lime juice at zero time was 0.812g/100 ml, whereas reducing sugars of pasteurized lime juice at zero time was 0.818 g/100ml, consequently pasteurized lime juice exhibited higher reducing sugars content compared to unpasteurized lime juice. These results are in accordance with the data obtained by Farnworth *et al.* (2001) on Mexican orange juice. It decreased sharply after the third week of storage in fresh and pasteurized lime juice.

Data in Fig. 4 show reduction in reducing sugars content in fresh and pasteurized lime juice during storage time. Non reducing sugars of fresh lime juice at zero time was (0.179g/100 ml) and it was decreased during storage time. Similarly, non reducing sugars content was decreased in reconstituted lime juice prepared

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Fig. 1. Effect of cold storage at 4±1°C on ascorbic acid content of lime juices



Fig. 2. Effect of cold storage at 4 ±1°C on titratable acidity of lime juices



Fig. 3. Effect of cold storage at 4±1°C on pH value of lime juices



Fig. 4. Effect of cold storage at 4±1°C on reducing sugar of lime juices

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Fig. 5. Effect of cold storage at 4±1°C on non reducing sugar of lime juices



Fig. 6. Effect of cold storage at 4±1°C on total sugar of lime juices

from 30° Brix concentrate being (0.170 g/100 ml) at zero time compared to fresh lime juice, and reducing sugars content increased in concentrated lime juice (30° and 50° brix) at zero time (0.820g/100ml) compared to fresh lime juice. These results are in accordance with Shouman (1974).

Fig. 5 shows the decrement in non reducing sugars content during storage time in fresh, pasteurized and reconstituted juices prepared from lime juice concentrates (30° and 50°Brix). The loss in total sugars in concentrated lime juice at 30°Brix after storage time for 8 weeks at 4.0±1.0°C was 4.05%, and in concentrated lime juice at 50° brix was 2.91% compared to fresh lime juice which was 12.92 % after storage time for 8 weeks at 4.0±1.0°C. These results are in harmony with results obtained by Akhavan and Worlstad (1980) who detected about 6% loss in total sugars in pear juice concentrate after storage for 16 weeks at 37 °C, while grapefruit juice stored at 30 °C for 15 weeks, only a loss of 5% in sugars was found by Lee and Nagy (1988). The loss of total sugar may be ascribed to the decrement of reducing and non reducing sugars during storage time.

Free Amino Nitrogen

From the results in Fig. 7 it could be observed that, the free amino nitrogen content at zero time was 18.928 mg/100 ml in fresh lime juice, 18.732 mg/100ml in pasteurized lime juice, 19.516 mg/100ml and 19.684 mg/100ml in reconstituted juices prepared from lime juice concentrates 30° and 50°Brix, respectively.

During storage at $4.0\pm1.0^{\circ}$ C, the level of free amino nitrogen in both fresh and pasteurized lime juice showed gradual increment. These results are in accordance with Trifiro et al. (1995). On the other hand, there was decrement of free amino nitrogen in both reconstituted juices prepared from lime juice concentrates (30° and 50° Brix). The loss of free amino nitrogen in reconstituted juices prepared from lime juice concentrates (30° and 50° Brix) was 1.87% and 2%, respectively. Beudo et al. (2001) found that the decrease in total amino acid content in peach juice concentrate was observed to be 8, 35 and 60%, at temperature 15, 30 and 37°C after 112 days of storage, respectively. The loss of total amino acid concentration was

attributed to the Maillard reaction with glucose and fructose which is naturally present in the juice (Beudo *et al.*, 2001 and Urbicain *et al.*, 1999).

Browning Index

Fig. 8 shows the increasing of browning index in fresh, pasteurized and reconstituted juices prepared from lime juice concentrates (30° and 50° Brix) during storage time. These results are in harmony with the results obtained by Ziena (2000) who detected that the browning index was increased gradually during storage. The non enzymatic browning was higher in pasteurized and concentrated of lime juices compared to fresh lime juice. This indicates that the browning reactions increased by increasing storage time and temperature just as Golden delicious and Amasya apple juice concentrates (Burdurlu and Karadeniz, 2003), and the non enzymatic browning reactions was increased during storage in citrus juice concentrates as reported by Koca et al. (2003). It had been suggested that the development of browning in lemon and grapefruit juice concentrates may be attributed to their higher acidity (Eskin, 1990).

Microbiological Evaluation

Thermal treatment is by far the most common method used in food preservation being very effective against enzymes and microorganisms, but can generate undesirable changes in the nutritional value as well as in sensory properties of foods (Guerrero *et al.*, 2001). Data obtained in the present study showed that bacteria, yeasts & moulds and acidophilic and spore forming bacteria were not detected in all samples. Some samples contained less than 30 CFU/ml, therefore it was considered not detected. This may be attributed to the acidity of lime juice.

Results obtained by Oronsaye and Ighodaro (2000) showed that the antimicrobial activity exhibited by lime juice is highly significant and compared favorably with the action of most antibiotics on bacteria. They ascribed the antimicrobial effect to the contributing effect of both organic acid *i.e.*, the 4-hydroxy benzoic acid limonene and the citric acid.

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Fig. 7. Effect of cold storage at 4±1°C on free amino nitrogen of lime juices



Fig. 8. Effect of cold storage at 4±1°C on browning index of lime juices

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Sensory Evaluation

From the results shown in Fig. 9, it could be observed that fresh. pasteurized, and reconstituted juice prepared from lime juice concentrate 30°Brix had acceptable taste up to the eighth week, but reconstituted juice prepared from lime juice concentrate 50°Brix had acceptable taste up to the fourth week. Pasteurized lime juice had the highest score in taste after eight weeks compared to the fresh and reconstituted juice prepared from lime juice concentrate 30° brix, in spite of fresh lime juice had a higher score at zero time than pasteurized one.

Fig. 10 shows the score of colour during storage time and it could be observed that fresh, pasteurized and reconstituted juice prepared from lime juice concentrate 30°brix had acceptable colour up to the end of storage period, and the pasteurized lime juice had the highest score in colour through the 8 weeks of storage, but reconstituted juice prepared from lime juice concentrate 50°Brix had acceptable colour up to the sixth week.

On the other hand, the score of flavour was the highest in fresh and pasteurized lime juice up to the end of storage time, contrary to reconstituted juices prepared from lime juice concentrates (30° and 50° Brix) which showed acceptable flavour up to the fourth week in reconstituted juice prepared from lime juice concentrate 30°brix and up to the second week in reconstituted juice prepared from lime juice concentrate 50°brix, (Fig. 11). The obtained results are in harmony with the results reported by Shouman (1974) who found that with every increase in concentration of lime juice, the colour and flavour qualities were lowered, during storage at 4.4°C. This was ascribed to the low concentrations of soluble solids which retained colour quality and flavour constituents more than high concentrations of soluble solids.

Fig. 12 shows the score of overall acceptability, whereas all samples of lime juice revealed decrement of overall acceptability during storage time, it is may be due to the decrement in taste, colour and flavour during storage time. Gómez-López *et al.* (2010) found that the sensory quality (colour, flavour, aroma and overall quality) of orange juice which was untreated decreased during storage time up to 10 days. Pasteurized lime juice had the highest score of overall acceptability during the storage period.

It can be concluded that the quality parameters of fresh, pasteurized and concentrates of lime juice were slightly changed during storage for 8 weeks at 4.0 ± 1.0 °C. The results suggested that marketing of lime juice in pasteurized form under cooling conditions.



Fig. 9. Effect of cold storage at 4 ±1°C on sensory evaluation (Taste) of lime juices



Fig. 10. Effect of cold storage at 4±1°C on sensory evaluation (Colour) of lime juices



Fig. 11. Effect of cold storage at 4±1°C on sensory evaluation (Flavour) of lime juices



Fig. 12. Effect of cold storage at 4±1°C on sensory evaluation (Overall acceptability) of lime juices

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يعتبر عصير الليمون مصدر غني بكثير من المركبات الحيوية التي لها العديد من الفوائد الصحية، وتتغير جودة العصير تحت تأثير التصنيع والتخزين فيما بعد، وتهدف هذه الدراسة إلى تقدير المركبات الكيميانية والتقييم الميكروبيولوجي والحسي على عصير الليمون الطازج والمبستر والمركز على ٣٠ و ٥٠ بركس المخزنة على ٤ ± ٥١م لمدة ٨ أسابيع، وقد حدث تغير أثناء فترة التخزين لكل من فيتامين ج والحموضة وقيمة الـ pH والسكريات المختزلة وغير المختزلة والنيتروجين الأميني الحر ومدلول التلون البني بالإضافة إلى التقييم الحسي (الطعم، اللون، النكهة و القبول العام). وجد أثناء فترة التخزين أن هناك إنخفاض تدريجي لكل من السكريات المختزلة وغير المختزلة وفيتامين ج والطعم واللون والنكهة والقبول العام والذي لوحظ في كل عينات عصير الليمون المخزنة، وكانت نسبة الاحتفاظ بفيتامين ج في فترة التخزين على٤ ± ١ °م لمدة ٨ أسابيع هي ٢,١٤% في عصير الليمون الطازج ٣٣,٥٣% في عصير الليمون المبستر، ٥١,٥٧% و ٦٦,٨٣% للعصير المسترجع المحضر من مركز عصير الليمون ٣٠ و٥٠ بركس على التوالي، وأثناء التخزين أيضاً كان هناك ارتفاع مستمر لمدلول التلون البني في عصير الليمون الطازج والمبستر والعصائر المسترجعة. بالإضافة إلى ذلك وجد ارتفاع في الحموضة في عصير الليمون الطازج، وكان هناك ارتفاع بسيط في محتوى النيتروجين الاميني الحر في عصير الليمون الطازج والمبستر أثناء فترة التخزين، أيضاً ارتفعت قيمة الـpH أثناء التخزين في كل من عصير الليمون المبستر والعصير المسترجع المحضر من مركز عصير الليمون ٣٠ و ٥٠ بركس، وبالنسبة الى التقييم الميكروبيولوجي فإن أغلب عينات عصير الليمون أظهرت عدد ميكروبي أقل من ٣٠ خلية في المل بينما لم تحتوي العينات الأخرى على أية مجموعات ميكروبية، ولذلك يعتبر التقييم الميكروبيولوجي لم يظهر بعد. أظهر التقييم الحسي لعصائر الليمون أن عصبير الليمون الطازج والمبستر والعصبير المسترجع المحضر من مركز عصبير الليمون ٣٠ بركس، لهم قبولا في الطعم واللون حتى ٨ أسابيع من التخزين، ولكن العصير المسترجع المحضر من مركز عصير الليمون ٥٠ بركس له قبول في الطعم حتى الأسبوع الرابع وقبولا في اللون حتى الأسبوع السادس، من ناحية أخرى فقد سجل عصير الليمون الطازج والمبستر أعلى درجة في النكهة حتى نهاية فترة التخزين، أما بالنسبة للقبول العام فقد أظهرت جميع عينات عصبر الليمون انخفاضا في القبول العام أثناء فترة التخزين.

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