

ZNANSTVENA BILJEŠKA / SCIENTIFIC NOTE

Influence of Storage on Quality and Sensorial Properties of Sports Drink with Lemon Juice and Isomaltulose

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Abstract

Lemon juice is a very common base in sports drinks because of its fresh and pleasant taste. Sugar isomaltulose is relatively new sugar suitable for sports drinks due to low glycemic index, but is still not widely used. Sports drink, the subject of this research, is only one produced in Croatia and present on Croatian market with added isomaltulose. Given this innovation in drink formulation the aim of this study was to monitor the quality and sensorial properties of the drink during six months storage at ambient temperature $20\pm 2^{\circ}\text{C}$. Researched drink was produced in industrial plant in Croatia in ordinary production for commercial purpose, and was formulated and declared as isotonic. To achieve the required properties drink besides mentioned ingredients (lemon juice and isomaltulose) contains, fructose, minerals (sodium, potassium, magnesium, calcium, zinc) and vitamins (C, E, niacin). Produced drink was filled in polyethylene terephthalate bottles and analysed after 0, 2, 3, 5 and 6 months of storage. Due to monitoring stability of quality properties of drink soluble dry matter, pH value, acidity, colour parameters (by CIELAB method), osmolality (by osmometer), content of isomaltulose and vitamins (by high-performance liquid chromatography), and minerals (by atomic absorption spectrometer) were determined. Sensory properties were determined by quantitative descriptive method.

Considering quality parameters significant changes were observed only in amount of vitamin C as well as colour parameters L^* , a^* , b^* while all others remained unchanged during six months storage at $20\pm 2^{\circ}\text{C}$. Further, among sensory attributes researched drink undergone the significant changes in colour (darkening) and taste (lower acidity and fresh fruit taste and higher sweetness). High correlation between sensory and instrumental determined colour was established. Despite mostly all quality parameters remained stable during all storage time without any trend of soon changes, six months of shelf life is maximal due to colour and taste are limiting factors.

Key words: Isotonic sports drink, Storage, Stability, Osmolality, QDA, Isomaltulose, CIELab

Introduction

The main purpose of sports drinks is to provide water, energy and electrolytes in a palatable and readily assimilable form. Sports drinks are intended to athletes and others exposed to strenuous physical activities to keep them hydrated and delay fatigue. It contains optimal level of carbohydrates as a source of energy, certain salts e.g. sodium chloride, sodium citrate, potassium chloride, potassium citrate and some other as magnesium and calcium to achieve target value of electrolyte content, some vitamins like A, C, E beside others (Ashurst, 2005; Lavalli et al, 2010). They should contain $40\text{--}80\text{ gL}^{-1}$ (optimally $60\text{--}80\text{ gL}^{-1}$) carbohydrate and more than 400 mgL^{-1} (optimally $>600\text{ mgL}^{-1}$) sodium (Brouns&Kovacs, 1997). Besides, considering its absorption osmolality of drink is very important. Sports drinks mostly are designed as isotonic (Ashurst, 2005). EFSA use term isotonic and slight hypotonic for solutions with osmolality i.e. $200\text{--}330\text{ mmolkg}^{-1}$. Osmolality is strongly influenced by the proportion of monosaccharides, disaccharides or polysaccharides besides carbohydrate content (Mettler, 2006) as well as electrolyte content (Brouns&Kovacs, 1997). Further, desirable sugars for such drinks are sugars with low glycemic index due to slow release energy (Burke et al, 1998; Siu&Wong, 2004). Recently, new sugar isomaltulose (disaccharide composed of $\alpha\text{-1,6}$ -linked glucose and fructose) attributed by low glycemic index is in use as replacement for sucrose (Jeukendrup et al, 2000; Lina et al, 2002; Achten et al, 2007;

Holub et al, 2010; Kawaguti&Sato 2011). It is completely metabolized in human body but blood glucose and insulin levels rise slower and reach lower maxima than after sucrose consumption. It helps improve regeneration of individuals exposed to physical exertion (Wu & Williams, 2006) thus it is suitable and valuable ingredient for sports drinks. Also, it can be helpful on mental concentration in humans (Kashimura et al., 2003), contains lower cariogenic potential and in some foods and beverages is more stable than sucrose (Kawaguti&Sato, 2011), which is susceptible to hydrolysis under acidic conditions (Babsky, 1986). In spite of all mentioned benefits and that has been increasing interest in production of isomaltulose in last two decades (Kawaguti&Sato, 2011), it is still not widely used in sports drinks.

Besides all mentioned constituents beverage palatability also depends on kind of fruit juice used in its formula. Taste and aroma of sports drink usually originated from added juices and in majority it is lemon juice, because of its fresh, fruity and pleasant taste (Ashurst, 2005). On the other hand, lemon juice as orange and grapefruit juices are susceptible to darkening (Handerwerk et al, 1988) what is result of non-enzymatic browning during storage. (Robertson&Samaniego, 1986; Kacem et al, 1987; Rouseff et al, 1989; Koca et al., 2003). Non-enzymatic browning in juices could result from Maillard reaction (between α -amino groups and reducing sugars), and ascorbic acid browning processes, and degradation of pigments (Zhu et al, 2009). In spite of this phenomenon is well known

and well-studied through the many years (Clegg, 1964; Lee & Nagy, 1988; Nagy et al., 1990, Martins et al., 2001; Burdurlu et al., 2006), it is still attracting the attention of scientist with the main reason to overcome that unpleasant phenomenon (Sharma et al, 2006; Valdramidis et al, 2010; Fustier et al, 2011; Bharate&Bharate 2014).

All ingredients and desirable sensory attributes have to be preserved in sports drink during its shelf life. If only one ingredient or sensory attribute do not fulfil desired requirements, in spite of well microbiological findings, shelf-life should be shortened. According to literature findings susceptibility of citrus juices to darkening (Handerwerk et al, 1988), could be limiting factor of shelf-life of such drinks.

The subject of this research was a sports drink based on lemon juice but instead of usual sugars present in most drinks on the market, the researched drink contains isomaltulose. Due to kind of sugar could influence on intensity of browning (Oh at al, 2005), and isomaltulose has greater stability than sucrose in some foods and beverages (Kawaguti&Sato, 2011) the aim of this study was to investigate, through monitoring the quality and sensorial properties of the researched drink, if its predicted shelf life of six months is appropriate or should be shortened or could be prolonged.

Materials and Methods

Materials

Sports drink based on lemon juice was produced in industrial plant in ordinary production for commercial purpose in two batches. Produced drink was filled in dark polyethylene terephthalate bottles and stored in producer's regular storage room at ambient temperature $20\pm 2^\circ\text{C}$. Samples for analysis were taken out after 0, 2, 3, 5 and 6 months of storage from the same batches. Results were presented their average values.

Drink soluble solids was minimally 4 % and pH 3.6. Drink contained: water; lemon juice from concentrate (3 %); fructose from fructose syrup (2,2 %); isomaltulose (1.8 %); citric acid, calcium lactate, potassium citrate, magnesium carbonate, zinc citrate, Na-cyclamate, Na-saccharinate; ascorbic acid (150 mgL^{-1} vitamin E and niacin, sodium benzoate, and potassium sorbate. Electrolyte and sugar content in drink were formulated to obtain osmolality $300\pm 10\text{ mosmkg}^{-1}$ consequently to be characterized as an isotonic drink and with total sodium concentration 190 mgL^{-1} .

Methods

Content of isomaltulose was determined by high-performance liquid chromatograph (HPLC) (McAlister et al, 1990).

Osmolality was determined by cryoscopy method using osmometer, according to instructions of producers (Gonotec GmbH, Users Manual, 2009).

Minerals were determined by atomic absorption spectrometer (AAS):

Na, K, Mg and Ca were determined by HRN EN 1134:2001 (EN 1134:1994) and Zn by HRN EN 14084:2005 (EN 14084:2003).

Vitamins were determined by HPLC: Vitamin C was determined by IFU Method No. 17 a /1995; α -tocopherol was determined by HRN EN 12822:2003 (EN 12822:2000); niacin was determined by HRN EN 15652:2010 (EN 15652:2009).

Soluble solids were determined with a refractometer by IFU Method No. 8/1991 and results were expressed in Brix percentages ($^\circ\text{Brix}$), pH value by IFU Method No. 11/1989, and total acidity by HRN ISO 750:2001 (ISO 750:1998) and results were expressed as citric acid in gL^{-1} .

Colour was monitored according to CIELAB method (McGuire, 1992).

All measurements were done in duplicate.

Sensory properties were evaluated by quantitative descriptive method (QDA) as described earlier (Marković et al., 2011). Briefly, sensory attributes were evaluated by sensory panel group of ten members from (25 – 50 years, 3 male, 7 female) in a sensory laboratory performed according to all of the requirements for the sensory evaluation (ISO, 2007). Sensory attributes that were studied were taste attributes (sweetness, acidity, bitterness, artificial, fruity, astringent, fresh), colour attributes (yellow, brown, opalescent) and intensity of odour. The intensity of each sensory attribute was rated from 0 = "none" to 10 = "high". Temperature of evaluated samples were $20\pm 2^\circ\text{C}$. All results were statistically analysed using ANOVA and the results of sensory evaluation, besides spider's web plots, were interpret by principal component analysis (PCA) (StatSoft Inc. Tulsa, SAD, Single User Version. University of Zagreb, 2014).

Results and Discussion

Results presented in Figure 1, 2 and 3 showed that concentration of minerals, osmolality and level of isomaltulose remained stable during six months of storage.

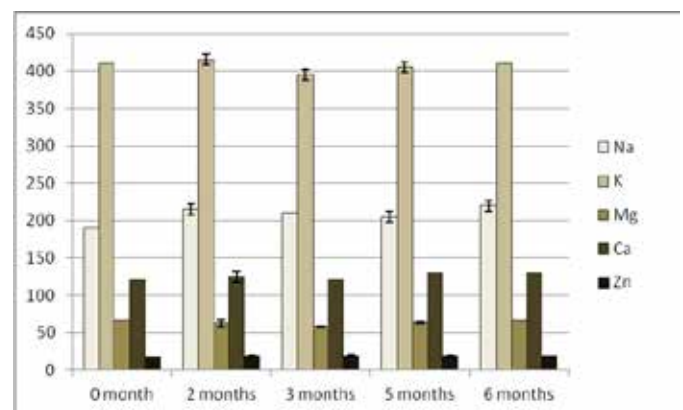


Figure 1. Minerals (mgL^{-1}) in sports drink during storage

Concentrations of vitamin E remained stable during storage with concentrations of $17.80\text{--}17.90\text{ mgL}^{-1}$, but niacin slightly decreased. (Table 1). Vitamin C decreased approximately for 30 % during six months of storage (Table 1).

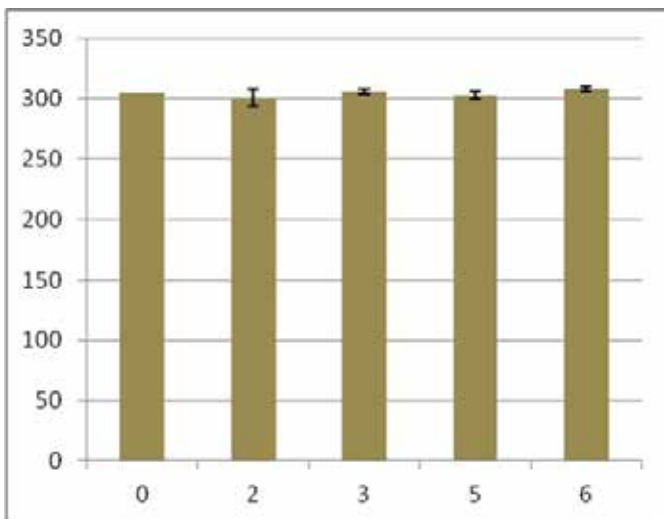
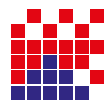


Figure 2. Osmolality (mosmkg⁻¹) in sports drink during storage

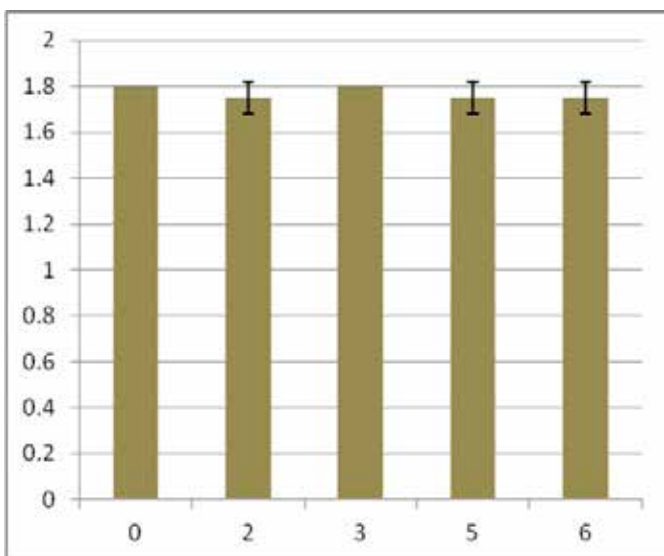


Figure 3. Percentage of isomaltulose in sports drink during storage

Total acidity and pH value was mostly unchanged, while soluble solids showed very slight increase during storage (Table 2). Observed increase could be explained as result of probable minor decomposition of some polysaccharides present like pectin in acidic media (Thakur et al., 1997).

Table 1. Vitamins (mgL⁻¹) in sports drink during storage

Month of storage	Vitamin C (mgL ⁻¹)	α -tocopherol (mgL ⁻¹)	Niacin (mgL ⁻¹)
0	158.50 \pm 0.71	17.80 \pm 0.00	23.00 \pm 0.00
2	133.00 \pm 1.41	17.85 \pm 0.07	23.50 \pm 0.71
3	120.50 \pm 0.71	17.80 \pm 0.00	24.50 \pm 3.54
5	113.50 \pm 0.71	17.80 \pm 0.00	23.50 \pm 2.12
6	108.50 \pm 0.71	17.90 \pm 0.00	21.50 \pm 0.71

Table 2. Soluble solids, pH value and total acidity of sports drink during storage

Months of storage	Soluble solids ($^{\circ}$ Brix)	pH value	Total acidity as citric acid (gL ⁻¹)
0	5.22 \pm 0.00	3.64 \pm 0.01	4.68 \pm 0.01
2	5.31 \pm 0.01	3.66 \pm 0.01	4.62 \pm 0.01
3	5.34 \pm 0.01	3.65 \pm 0.01	4.63 \pm 0.02
5	5.43 \pm 0.02	3.67 \pm 0.00	4.71 \pm 0.16
6	5.51 \pm 0.07	3.68 \pm 0.01	4.68 \pm 0.10

Analysis of variance showed significant influence of storage time on vitamin C, but there was not observed significant influence on osmolality, level of isomaltulose, other vitamins, all minerals, soluble solids, pH, and total acidity..

The most obvious changes of investigated sports drink were observed in colour. Lightness (L*) was between 65.7 at the end of storage and 71.8 before storage, what means that darkening occurred. Redness (a*) was very low but increase from 1.35 to 2.6 and yellowness (b*) were higher, 30.68, before storage and increased on 34.40 after six month (Table 3). Chroma (C*) also increased during storage what means that drink's colour was vividier after six month. Hue (h*) value showed that colour is in yellow part of colour wheel with decreasing trend during storage. That means that yellow colour of the drink became closer to the part of red colours, but was still yellow. Before storage investigated drink was very light yellow (Figure 3), so darkening was resulted only with increase of intensity of yellow colour. dE*, colour difference, is defined on the basis of theoretical conclusions, experience and experimental results as $dE^* = [(dL^*)^2 + (da^*)^2 + (db^*)^2]^{1/2}$ and present a total difference in colour among L*, a*, and b* values of started sample (drink before storage) and samples after 2, 3, 5 and 6 months of storage. dE increased as colour was changed. All colour parameters indicated a visual evaluation of drinks colour (Figure 3). Changes of colour during the first three months were not such remarkable. The most obvious changes occurred in the last months (Figure 3). Storage time significantly influenced on all colour parameters ($p \leq 0.05$).



Figure 4. Original image of investigated drink (S-0 –S-2, S-3, S-5 –, S-6 after 0, 2, 3, 5, 6 months of storage, respectively)

Table 3. Colour parameters of sports drinks during storage

Month of storage	L*	a*	b*	C*	H*	dE*
0	71.80±0.00	1.35±0.05	30.65±0.15	30.68±0.15	87.48±0.08	-
2	71.05±0.20	1.55±0.05	30.95±0.35	30.99±0.35	87.13±0.06	0.93±0.63
3	69.90±0.30	1.75±0.05	31.70±0.10	31.75±0.10	86.84±0.1	2.22±0.3
5	68.60±0.40	1.80±0.10	31.70±0.30	31.75±0.31	86.75±0.15	3.40±0.24
6	65.70±0.60	2.60±0.20	34.30±0.50	34.40±0.51	85.67±0.27	7.22±0.54

After six months of storage, what is labelled shelf life of investigated drink, change of colour was also observed by sensory evaluation, with lower increase of intensity of brown than intensity of yellow what is in accordance to measured colour parameters. Contrary, intensity of odour decreased during storage. Changes of taste were noticed, too. Intensity of fresh, fruity, bitter and acidic taste decreased, and intensity of artificial and sweet taste increase.

According to ANOVA moreover, storage time significantly influenced on intensity of yellow and brown colour, intensity and fruity odour, fruity and fresh taste, bitterness, sweetness, acidic, and artificial impression ($p \leq 0.05$). Also, there was high correlation between sensorial scores and measured colour parameters ($0,762 < r < 0,885$). Although, total acidity, pH, soluble solids, level of isomaltulose did not significantly changed during storage, increase of sweetness and decrease of acidity during storage through sensory evaluation was obvious.

Such changes probably are result of non-enzymatic browning what is known phenomenon in citrus juices and well-studied through the many years (Clegg, 1964; Lee & Nagy, 1988; Nagy et al., 1990, Martins et al., 2001; Burdurlu et al., 2006). It is known that non-enzymatic browning is responsible for this unpleasant darkening of lemon, orange or grapefruit juice during storage (Robertson & Samaniego, 1986, Kacem et al, 1987, Rouseff et al, 1989, Koca et al., 2003). Authors studied causes and chemical reactions with the aim to over-

come that unpleasant phenomenon. It is found that vitamin C, L-ascorbic acid, is main precursor in these reactions and that carbonyl compounds formed from its degradation are responsible for color changing (Roig et al, 1999; Johnson et al, 1995). But mechanism of its degradation is still not fully understood, and both pathways aerobic and anaerobic are widely accepted (Manso et al, 2006). Burdurlu et al. (2006) found that losses of ascorbic acid correlated with accumulation of hydroxymethylfurfural (HMF) in citrus juices during storage. But according to Clegg (1964) HMF in lemon juices is not an active compound in the development of brown pigments. Furthermore HMF is characterized by sweet and caramel taste (Edris et al, 2007) so its accumulation could influence on taste of drink. In our study content of vitamin C significantly decrease ($p \leq 0.05$). At the same time, all colour parameters significantly changed and suggested darkening what the image (Figure 4) clearly shows. Further taste also significantly changed, and drink became sweeter, probably accumulated HMF was responsible for such taste changes, as already was mentioned. Those sensorial changes are limiting factor for shelf life of researched sports drink, because there were not observed significant influence of six months storage on quality properties of drink. In spite of all noticeable changes the drink was still potable during investigated period, and its shelf life. However, longer shelf life is not recommended.

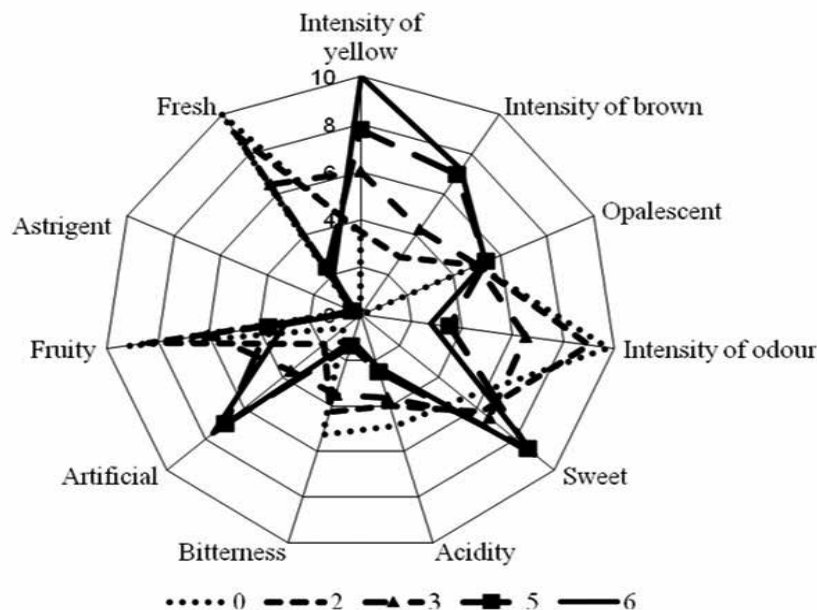


Figure 5. Spider's web plot of investigated drink

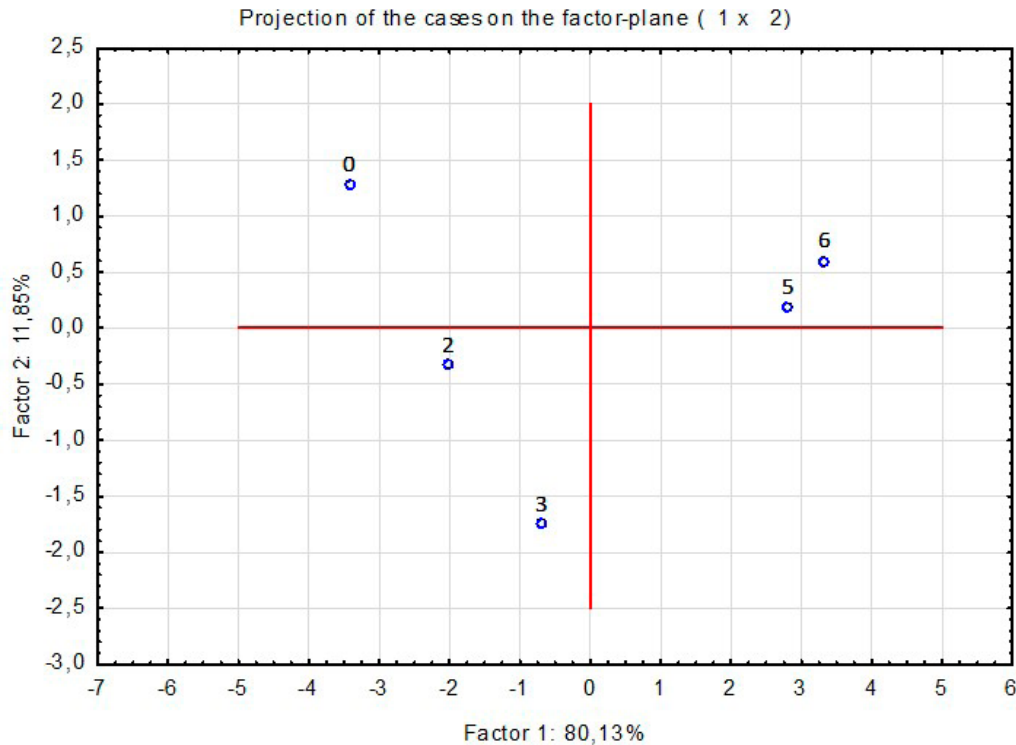
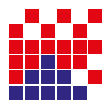


Figure 6. Principal component analysis (PC1 vs. PC2) of sport drink. Sample labels for the score plot: 0-start sample; 2-2 month of storage; 3-3 month of storage; 5-5 month of storage; 6-6 month of storage

Additionally, principal component analysis (PCA) was performed on all samples and variables to determine whether storage time of investigated sports drink with lemon juices had influenced the sensory attribute profile. In this context, 11 sensory attributes were the investigated variables, while sports drink were the cases under investigation. The first two factors (PC1 and PC2) represent 80.13% of the initial data variability (Fig 6). Samples with 0, 2 and 3 month of storage were positioned on the left, while samples of 5 and 6 month of storage on the right side of the PC1. All investigated sensory attributes, except astringent and fresh taste, strongly correlated with the PC1, while those two attributes showed better correlations with the PC2. Therefore, samples with 0, 2 and 3 month of storage were characterized with yellow intensity and fruity taste, and samples with 5 and 6 month of storage were mostly characterized with brown intensity and sweetness.

Conclusions

Investigated sports drink after six months storage at $20 \pm 2^\circ\text{C}$ undergone significant changes in colour ($L^*a^*b^*$ -parameters and sensory evaluated) and taste properties as well as content of vitamin C ($p \leq 0.05$). Contrary, vitamin E and niacin, all minerals (sodium, potassium, calcium, magnesium, zinc), level of isomaltulose, and osmolality as well as soluble solids, pH, an total acidity remained stable during six months without any significant changes.

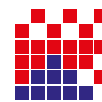
Future research should be addressed to improving formula of drink due to achieving more successful preventing or delaying of non-enzymatic browning with aim to obtain drink with more stable colour and taste.

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