Development of mango (*Mangifera indica* L.) energy drinks

Desarrollo de bebidas energizantes de mango (*Mangifera indica* L.)

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ABSTRACT

The effect of two hydrocolloids, pectin and carboxymethyl cellulose (CMC), was evaluated in mango beverage stability (*Mangifera indica* L.) formulated and developed with caffeine at a concentration of 30 mg/100 mL. The physico-chemical and sensory characteristics of color, acidity, viscosity, total soluble solids, *pH*, flavor, aroma and texture were studied every three days over a 12-day period. The beverages were packaged in high-density polyethylene containers with a 250 mL capacity and were stored at 5 °C and 90% RH for the duration of the experimentation period. The drinks with added pectin showed greater stability and lower acidity values than the control, but higher values than those prepared with CMC. The drinks made with CMC had a significantly higher viscosity at a 95% confidence level than those made with pectin or the control beverages. The treatment that showed the lowest browning index was the one added with pectin. Concerning the sensory evaluation, the drinks showed significant differences at a 95% confidence level; the drink made with pectin was the most widely accepted. It was concluded that the most stable drinks were those made with pectin because they presented the lowest height in millimeters of precipitate solids over the storage period. No off-flavors in beverages were perceived by the judges.

Key words:
Agroindustry
Caffeine
Pectin
Carboxymethyl cellulose

Palabras claves:
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RESUMEN

Se evaluó el efecto de dos hidrocoloides, pectina y carboximetil celulosa (CMC) en la estabilidad de bebidas de mango (*Mangifera indica* L.) adicionadas con cafeína en concentración de 30 mg/100 mL. Se estudiaron las características físico-químicas y sensoriales; color, acidez, viscosidad, sólidos solubles totales, *pH*, sabor, aroma y textura bucal, cada tres días durante doce días. Los refrescos fueron empacados en envases de polietileno de alta densidad con capacidad para 250 mL y se almacenaron a 5 °C y 90% de HR durante el tiempo de experimentación. Las bebidas adicionadas con pectina mostraron mayor estabilidad y valores de acidez más bajos que el control, pero superiores a las preparadas con carboximetilcelulosa (CMC). Las bebidas con CMC presentaron una mayor viscosidad con un nivel de confianza del 95% respecto a las elaboradas con pectina y el control. El tratamiento que mejor color presentó fue el adicionado con pectina. Las bebidas en cuanto a su evaluación sensorial presentaron diferencias significativas con un nivel de confianza del 95%, siendo la de mayor aceptación de acuerdo a la evaluación sensorial la elaborada con pectina. Se pudo concluir que las bebidas con pectina presentaron menor altura de sólidos precipitados expresados en milímetros durante el tiempo de almacenamiento. No se percibieron sabores extraños en las bebidas por parte de los jueces.

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The food hydrocolloids are high molecular weight polysaccharides, which are water soluble, used in a variety of functions in food systems for increasing viscosity, forming gel structures and films, controlling crystallization, inhibiting syneresis, improving texture, encapsulating flavors and increasing physical stability, among others. Due to its functional properties such as water retention capacity, balance of rheological properties and ionization of aqueous solutions, it is used to control the instability of food and the suspension of insoluble particles (Figueroa, 2015).

In food industry, polysaccharides are commonly used as emulsifiers to stabilize beverages. The typical beverage emulsion usually contains an oil phase and aqueous phase. The oil phase is comprised of flavor (such as orange oil, lemon oil, among others) while the aqueous phase that includes emulsifiers, acids and antimicrobial agents, which need to be stabilized (Zhao et al., 2015).

The beverages should be stable, homogeneous and without particle aggregation. However, there are three physical phenomena that cause instability of a suspension: sedimentation, aggregation and coalescence. Sedimentation is the result of a density difference between the dispersed phase and the continuous phase, and produces two separate phases with different concentrations and viscosities. The suspended particles settle differently, depending on their characteristics and concentration. The discrete particles are those whose characteristics do not change (shape, size and density). In the free sedimentation (i.e., without interference between particles), the particles are held by hydraulic forces and the fall can be described by Stokes' law (Figueroa, 2015).

The technological problem of these juices is to stabilize particles during storage due to the presence of pectin methyl esterase (PME). The result is an unattractive beverage in its sensory properties. This separation is a serious quality defect, which reduces the attractiveness and marketability of the product. Usually, the heat treatment used for PME inhibition stabilizes the colloids and the suspended particles. However, hydrocolloids negative charge (like sodium alginate, CMC and Arabic gum) in concentrations as low as 0.05% completely inhibits juice clarification. Furthermore, gum absorption on particles may give rise to steric repulsion stabilizing the beverages (Ibrahim et al., 2011).

Energy drinks first appeared in the 1960s in Europe and Asia. However, the introduction and aggressive marketing of Red Bull in Austria in 1987, and in the U.S. in 1997, created a trend towards the consumption of energy drinks with high caffeine content. Energy drink production has seen exponential growth since its inception, with nearly 500 new brands reportedly launched around the world in 2006 and 200 brands launched in the US over a 12-month period ending in July 2007 (Reissig et al., 2009). A study by Aranda et al. (2006) found that the main active ingredient in energy drinks was caffeine, although the authors also found that substances including taurine, riboflavin, pyridoxine, nicotinamide and other B vitamins were present, along with various phytochemical derivatives.

The population that consumes energy drinks is varied; it includes athletes, students and executives. Energy drinks are attractive to consumers because they provide the temporary benefits of increased alertness, improved mood and increased mental and physical energy (Costa et al., 2014). Consumers expect the consumption of these drinks to provide a stimulating effect, particularly of a physiological nature (Reissig et al., 2009). A study by researchers at the Pontificia Universidad Javeriana defined attention as a central processing control mechanism that acts in accordance with the objectives of the organism by activating and inhibiting processes; it can be directed towards the senses, knowledge structures in memory, and response systems. Within this framework, the researchers found that energy drinks make participants to react faster. This type of energizing beverage can also have an effect mainly on the organism’s peripheral nervous system, leading to increased tonic components in attentional processes, which are related to processes such as waking (arousal or mental), physical endurance and preparing the organism to respond faster (Aguilar et al., 2008).

Studies have shown that consumption of fruit drinks among teenagers declined from 1999 to 2008, while consumption of energy drinks tripled. Energy drinks contain added sugars and high amounts of caffeine, which is often supplemented with other natural stimulants to enhance its effects. They may also contain vitamins, minerals and proteins intended to increase concentration and physical performance (Larson et al., 2014).
Mango (Mangifera indica L.) is cultivated in 16 Colombian departments, of which Tolima is one of the main producers. It is a species of Indian origin planted at altitudes ranging from sea level to 1650 m (Corpoica, 2013). The color of mangoes varies depending on their stage of maturation and variety; some go from being green to purple and red, while others are yellow in their mature stage. Sixteen mango varieties exist in the Colombian market. These are classified into two groups: mangoes of creole origin and table mangoes, the latter of which are improved varieties. The grade Brix of mangoes depends on the altitude where they are cultivated (Garcia, 2009).

Mango is a commercial fruit widely consumed at a national and global scale. Consumers also accept processed products. These include fruit drinks, which are prepared from fresh mango pulp or reconstituted dried pulp, added with water, sweeteners, flavorings and other additives (according to national legislation). These are subjected to the preservation treatment deemed appropriate for the manufacture of these products. It should be kept in mind that the Colombian Institute for Technical Standards and Certification (ICONTEC) is responsible for defining the standards and quality parameters for products (ICONTEC, 1999).

In order to develop an energy drink from mango, it is necessary to consider the resolutions that regulate the ingredients used in commercial products. These include NTC 3549, regarding fruit drink processing, and resolution 4150 of 2009, which establishes the technical regulations on the requirements that energy drinks must meet for human consumption (ICONTEC, 1999).

The objective of the present study was to evaluate the stability of mango energy drinks with caffeine, added with hydrocolloids such as pectin and CMC.

MATERIALS AND METHODS

Materials and equipment
The energy drinks were prepared using unripe mangoes with a maturity index of 0 according to NTC 5210. The fruit was acquired from the Central Minorista market (Jose Maria Villa) of Medellin, Colombia. Mangoes that showed no signs of mechanical damage or deterioration and were homogeneous in size and color were selected and standardized. To characterize the pulp of the fruit, an Ohaus balance with a precision of 0.01 g and a Leica Abbe auto refractometer with a 0-32% scale were used by applying the AOAC 932.12/90 method. The acidity was determined by acid-base titration with 0.1N NaOH, and was expressed as a percentage of citric acid, by applying AOAC 942.05/90 method. The pH was measured with a CG-840B Schott digital potentiometer, and viscosity was measured with an Ostwald viscometer (Bernal, 1993).

Pulp preparation
The fruits were washed and sanitized by immersion in sodium hypochlorite (50 ppm) for 15 min. They were then rinsed, peeled, and processed, with a small amount of water added to facilitate this process. They were subsequently homogenized using an Oster blender and sieved to separate the pulp.

Production of the drinks
According to previous essays and NTC 3549, there were selected two stabilizers for the treatments: one with 0.1% CMC and the other one with 0.1% pectin. The energy drinks were prepared with a share of 25% pulp, 12° Brix total soluble solids, and a caffeine content of 30 mg per 100 mL. A third treatment without a stabilizer was used as control. To produce the beverages, sucrose and pulp were added to water and the mixture was then gently stirred until it became homogenized. After the hydrocolloids were added, the solution was homogenized again for five minutes. The products obtained were packaged in sterile 250 mL polyethylene bottles and refrigerated (5 °C) for 12 days which it is normal for fruit beverages without heat treatment time. Each beverage was manufactured six times (repetitions), and all of the analyses were performed on the six samples in each treatment for day of evaluation. The test were destructive analyzes for each sample and for each day. The drinks were evaluated for total soluble solids, pH, acidity, viscosity, color, and precipitate height in plastic bottles after 3, 6, 9 and 12 days of storage. Sensory quality was evaluated after the 12-day storage period. The effect of stabilizing agents in beverages was measured with the height of precipitate in centimeters using a calibrator, something that could be related to turbidity obtaining supernatant phase.

Browning index
The effect of the stabilizers on the browning rate in mango drinks with caffeine added was determined by measuring
color using a sphere spectrophotometer with a D65 illuminant and a 10° observation angle. The equipment used was a Model SP64 sphere spectrophotometer, X-RITE Inc., MI, USA. The evaluation was performed every 3 days during a 12-day period. The readings were obtained in the CIE-L“a“b” (Commission Internationale d’Eclairage) color coordinates, where “L” stands for brightness, scaled from 0 to 100, “a” represents the range of colors from green to red, and “b” represents the range of colors from blue to yellow. Measurements were taken for the six experimental units (EU) in each treatment. The browning index (BI) was calculated based on Equations 1 and 2 (Maskan, 2001).

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BI = \frac{100(X - 0.31)}{0.17}
\]

\[
x = \frac{(a + 1.75L)}{(5.645L + a - 3.012b)}
\]

Sensory analysis
The beverages were subjected to sensory evaluation by 15 panelists knowledgeable about the sensory characteristics of mango drinks. The panelists received approximately 20 mL of each sample in plastic cups coded with random three-digit numbers, at temperatures of 7 °C - 8 °C. They were also offered water to clean their palates. The panelists quantified the perceived intensities of each attribute using a scale unstructured without neutral, with a ranging from 1 (low) to 5 (high). The attributes evaluated were the following: appearance (color and consistency), aroma, flavor (sweetness and acidity) and texture in the mouth (viscosity) (Márquez, 2015).

**Statistical analysis**
The experimental design consisted in a randomized complete block, with three treatments and six replications, for 18 experimental units per day of evaluation, measuring were performed for days 0, 3, 6, 9 and 12. Statistical analysis of the mean, standard error and anova were applied, using the statistical program Statgraphics plus.

**RESULTS AND DISCUSSION**
The physicochemical measurements for the mango pulp used in the production of the energy drinks showed the following characteristic values: °Brix = 7 ± 0.1; pH = 2.29 ± 0.13; and acidity = 1.17% ± 0.15.

Figure 1A shows that the pectin and CMC treatments had higher soluble solid concentrations than the control; this was likely due to the action of these two substances as stabilizers that facilitated moisture retention and sugar generation in the energizing drink. Another relevant factor could be the temperature of the medium. Figure 1B shows that viscosity changed depending on the compound used as stabilizer; the drink containing CMC had the highest viscosity (19 to 23 cp), while the formulation containing pectin was less viscous, but had a higher viscosity than the control. This is because the CMC molecules generate negative charges due to the carboxyl groups, leading to electrostatic repulsion between the chains, which was manifested in the increased viscosity of the beverage (Fennema, 1993).
The acid content was found to increase for all treatments during the storage period. This was due to the acid synthesis produced by the reactions of phytochemical substances. As shown in Figure 2A, this change was for the treatment without stabilizer. The samples with the CMC treatment maintained the most stable acid content during the storage period. Figure 2B shows that the drinks with CMC as a stabilizer had the highest pH values (the least acidic drinks), followed by those containing pectin. The drinks with the lowest pH (highest acidity) were those containing no stabilizer; thus, the most acidic drinks had the lowest pH.

Figure 2. Effect of hydrocolloids on acidity (A) and pH (B).

Figure 3A shows the browning index, which is a function of the CIEL*a*b* parameters measured with the spectrophotometer. The shelf life of food is determined by color variation, respiration rate, pulp firmness, among others (Vignoni et al., 2006). The browning index in the energy drinks was analyzed as a factor of product lifetime. Temperature and light degrade components associated with the color of the drink, usually flavonoids. The drinks previously made were stored in a cool, dark room, which is why the browning index decreased, improving the visual appearance of the beverages. Figure 3B shows the stability of the beverages as a function of the level of precipitates. It can be observed that the pectin samples presented the best hydrocolloid action, showing significant differences from the CMC samples and the control, with a 95% confidence level. This is due to L-rhamnose and D-xylose functional groups, which are capable of reacting with the solutes in the beverages to cause stabilization (Fennema, 2010).

Figure 3. Effect of hydrocolloids on browning index (A) and stability of beverages with height of precipitates.
The sensory analysis established that the drinks with pectin had the highest acceptance level, with 95% significance, compared to the control and the CMC beverages, because the latter had a high viscosity. This parameter is directly related to the attribute of texture in the mouth, which was less acceptable for the panelists in the CMC beverages.

The sensory evaluation showed that the drink with added pectin was the most widely accepted, with a 95% confidence level, for the attributes of flavor, color, and texture in the mouth. For aroma, no significant difference was found between the treatments. In the sensory evaluation by judges they found no extraneous flavors therefore considered that caffeine did not affect drinks.

CONCLUSIONS

The best stabilizer for the mango drinks added was with pectin since the beverages manufactured with this stabilizer presented less precipitate solids with a height of 4.4 cm over the storage period. They also were the best formulation according to sensory characteristics and they can be consumed as energy drinks.

The browning index decreased for all treatments, which is related to the loss of brightness in the drinks.

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