Formulation of Isotonic Drink Processed from Rambutan Fruit
(Nephelium lappaceum L) cv. Lebak Bulus

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Abstract
The formulation to develop an isotonic drink from rambutan fruits has been studied. The formula comprises of two types i.e. base formula dan advanced formula. The base formula are water, juice, sucrose, sodium benzoate, sodium bicarbonate, vitamin C, citric acid, sodium lactate, sodium nitrate, potassium chloride, sodium chloride. Various treatments applied were single strength rambutan juice and mixture of rambutan with pineapple juices (1:1). Advanced formula variation was with and without addition of 0.1% lychee flavour. Third variation was addition of 3.5% and 4.6% sugar. The best treatments was single strength rambutan juice, addition of 4.6% sucrose and 0.1% lychee flavour.

INTRODUCTION
Sweating is the way of human body to maintain body temperature to normal i.e. 37°C. This physical condition causes the body loss of the liquid and electrolytes, and at severe stage without any effort of replacement. It can cause the dehydration and circulation stroke. Sweat which evaporates through skin contains various types of electrolytes such as sodium, potassium, calcium, magnesium, chloride, bicarbonate, phosphate and sulphur (Winarno dan Kartawijayaputra, 2007). Three main minerals in the sweat are sodium, potassium, and chloride. The higher the rate of liquid losses from the body during sweating, the higher the rate of losses of sodium, pottasium, and chloride from human body (Irwan, 2007).

Sport drinks are designed so that human can regain the stamina after doing sports or other heavy physical activity. Sport drinks are classified as isotonic, hypertonic and hypotonic. Isotonic drinks contain salt and sugar similar to human body, while hypertonic contain higher salt and sugar. On the otherhand hypotonic drink contain lower salt and sugar (Wikipedia 2014). Normally, the isotonic drink contains electrolytes such as Na⁺, K⁺, Ca²⁺, Mg²⁺, Cl⁻ (Anonim, 2011a).

Depending on the purpose of physival activity, consuming sport drink is not only as electrolyte replacement but also supplying of substrate, prevention of dehydration, pre-exercise hydration, and post-exercise rehydration (Maughan 1998, Shireff 2003).

Important components for formulation of isotonic drinks are carbohydrate content (concentration and type), osmolality, electrolyte composition and concentration, flavouring components, and other active ingredients. All the above components can be modified depend on our purposes (Maughan, 1998).

In this research, the flavor component were modified to fulfill Indonesian preference. Indonesian tropical fruit that currently received high attention are rambutan and pineapple. Rambutan (Nephelium lappaceum L) is a tropical fruit which is belong to Sapindaceae family, originated from South East Asia. Beside has special flavor, rambutan characteristics
are high in vitamin C. i.e. as high as citrus. Rambutan cv. Aceh has vitamin C of 71.24 mg per 100 g. The character that can distinguish the rambutan cultivars are fruit pericarp, moisture content, shape, skin color, and fruit length. Rambutan cultivars which have high economic value are Rapiah, Aceh Lebak Bulus, Simacan, Silengkeng and Sinyonya (Astawan dan Leomitro, 2008).

Pineapple (Ananas comosus) is a tropical fruit originated in Brazil and Bolivia. Beside has special flavor, pineapple is rich in vitamin C which can act as antioxidant. Pineapple also rich in vitamin B1 which are required in helping human body to form an antioxidant (Astawan dan Leomitro, 2008).

The aim of the research was to develop a new isotonic formula from based formula by modification of the flavor with addition of rambutan and pineapple juice.

MATERIALS AND METHODS

Materials

Main ingredient

The main ingredient were rambutan cv. Lebak Bulus, pineapple cv. Jati, citric acid, sucrose, water. Fruit were bought at Pasar Anyar Market in Bogor.

Additional ingredient

Additional ingredient were benzoic acid, baking soda, vitamin C, calcium lactate, sodium citrate, potassium chloride, sodium chloride, artificial lychee essence.

Methods

There were 2 sets of experiment. First is analysing the chemical and organoleptical characteristics of isotonic drink available commercially. Second on the development of new formula of isotonic.

Commercial Isotonic Analysis

Commercial isotonic drink such as “mzne”, “vtzne”, “100 p” and “p s”, were bought from local store. They were then analysed for total acidity, total soluble solids, pH, Brix/acid ratio, and organoleptic characteristic.

Development of New Formula

There are three separate proses such as processing of rambutan juice and pineapple juice, and processing of isotonic. Extraction is the process of separating the juice from the fruit pulp. It is done for both rambutan and pineapple. This process can be done with or without the addition of water. The water serves to dilute the juice. Processing of isotonic beverage used pure juice of the fruit with no added water. The extraction process was using a juicer because this equipment is practical for separating fruit pulp, fruit juice and that will automatically separated.

Isotonic preparation was using a serial of process including formulation, cooking at 90 °C, pasteurisation at 90°C for 10 minutes, flavor addition and hot filling packaging. The processing was using a basic formula. Every 1000 ml of isotonic consisting of 3% fruit juice, Na-benzoate at 0.05%, sucrose 3.5%, 0.075% sodium bicarbonate, 0.1% vitamin C,
citric acid 0.0961%, 0.0109% calcium lactate, Na -citrate 0.0859%, 0.0297% KCl, and 0.0643% NaCl.

Processing new formula was to modify the basic formula with the addition of various additives. There were 8 types of formulas namely (1) R: Isotonic drink containing rambutan juice 30 ml, citric acid 0.961 g/l and sucrose 35 g/l; (2) RF: Isotonic drink containing rambutan juice 30 ml, citric acid 0.961 g/l, sucrose 35 g/l and lychee flavor 1ml/l, (3) RA: Isotonic drink containing rambutan juice 30 ml, citric acid 1.922 g/l and sucrose 46 g/l, (4) RAF: Isotonic drink containing rambutan juice 30 ml, citric acid 1.922 g/l, sucrose 46 g/l and lychee flavor, (5) RN: Isotonic drink containing rambutan juice 15 ml, pineapple juice 15 ml, citric acid 0.961 g/l and sugar 35 g/l, (6) RNF: Isotonic drink containing rambutan juice 15 ml, pineapple juice 15 ml, citric acid 0.961 g/l, sucrose 35 g/l and lychee flavor, (7) RNA: Isotonic drink containing rambutan juice 15 ml, pineapple juice 15 ml, citric acid 1.922 g/l and sucrose 46 g/l, (8) RNAF: Isotonic drink containing rambutan juice 15 ml, pineapple juice 15 ml, citric acid 1.922 g/l, sucrose 46 g/l and lychee flavor.

The isotonic drinks were then analysed for total acidity, total soluble solids, pH, Brix/acid ratio, and organoleptic characteristic.

Analyses

The analysis conducted was chemical and organoleptic analysis. Chemical analysis included pH, TSS, Total Acidity and Brix/acid ratio. pH was measured using a pH meter. Measurements were performed for one minute until the reading of the pH value was constant. Total soluble solids were measured using a refractometer.

Total acidity analysis was referred to AOAC (1995). Sample firstly stirred to obtain a homogeneous phase. Sample was weighed as much as 10 g, then diluted in 100 ml volumetric flask. A total of 25 ml of solution was pipetted into a 100 ml erlenmeyer flask and titrated with 0.1N NaOH using phenolphthalein indicator. Total acidity can be calculated with the formula:

\[
\text{Total acidity} = \frac{\text{Volume} \times \text{normality} \times \text{NaOH} \times \text{BE citric acid} \times \text{dilution factor}}{\text{Sample weight} \times 1000} \times 100\%
\]

Brix acid ratio was calculated as ratio of percentage of acidity with total soluble solids. Organoleptic analyses were conducted using hedonic scoring (Setyaningsih et al. 2010). Panelists was 10 trained panel. The scale was 1-5 from very dislike until very like.

RESULTS AND DISCUSSION

Materials Used in the Formulation of Isotonic Drink

Rambutan cv. Lebak Bulus has been chosen since it is more juicy than the rambutan cv. Rapiah. Rambutan cv. Sinyonya contains a lot of water but the flesh is not easily separated from the seeds so it is difficult to prepare the juice. Fruits used are ripe ones because they have a sweet taste and volatile substances that give a distinctive flavor to the fruit (Anonymous 2011b).

Pineapple cv. Bogor has the characteristics of small in size, yellow skin, delicate fibrous flesh and a sweet flavor. Based on these characteristics they were classified into Queen cultivar (Budi, 1998). Pineapples used were ripe pineapple. Pineapple fruit maturity level can be determined from the color of the peel. Fruits are ripe when the yellow color reached 25% of the total surface of the fruit. At this level the fruit has a high total soluble solids and low acidity (Anonymous, 2011d), because at the time of maturation the amount
of simple sugars is increased that give a sweet taste, as well as a decrease in organic acids and phenolic compounds that reduce pungent and acid taste. There is also an increase of volatile substances that give a distinctive flavor to the fruit (Anonymous, 2011c). Pineapple is a fruit that consists lots of sugar (Anonymous, 2011c). Some of simple sugars contained in pineapple are sucrose, fructose, and glucose (Anonymous, 2011c).

Water is one of the most important elements in the manufacture of beverages. The taste or smell of water will affect the taste of the final product (Buckle, et al. 2010). In the manufacture of isotonic drink the tap water was used which was then boiled and cooled. Water was heated to boiling temperature (100 °C) to kill the bacteria present in the water which are harmful such as Escherichia coli (Anonymous, 2011e). Water in addition to functioning as a release thirst while drunk also serves to dissolve the nutrients found in isotonic drinks (Winarno and Kartawijayaputra, 2007). For isotonic process the water was only heated to 90 °C since the water had already been boiled.

Commercial acid is an organic acid in the form of white crystals or powder form. Citric acid is easily soluble in water, spirits and ethanol. Its taste is very sour, and when heated will melt and then decompose into charcoal (Margono, et al.1993). According to Indonesia National Standard/SNI 01-4452 (1998), quality requirements for pH isotonic is 4. To lowering the pH of the isotonic drinks, citric acid is used as acidulant material (acidity regulator). It is a chemical compound that is acidic and serves to acidify, neutralize, and maintain the acidity of processed foods. In the processed fruit products, the addition of acidulant will a decrease the pH (Fachruddin, 1998). At low pH, required sterilization temperature will also be lower and the growth of harmful microbes will be smaller (Winarno, 2008).

Sucrose is a type of sugar that is used in isotonic drink which has the properties that rapidly emptying stomach and a high absorption in the gut. Both of these properties are affected by the levels and types of carbohydrates or sugar used in the drink. Types of carbohydrates, such as glucose, sucrose, maltodextrin have a relatively rapid absorption compared to fructose (Winarno and Kartawijayaputra, 2007). Isotonic drinks which have carbohydrates or sugar content of 6-8% will be absorbed by the body at the same speed as the body absorbs water (Winarno and Kartawijayaputra, 2007).

In beverages, sugar is not only giving a sweet taste, eventhough this taste is important. Sugar give perfection to sour taste, to other flavors, and also gives a sense of glut since it provides viscosity to the drink (Buckle, et al. 2010).

The minerals present in the body and especially in the food contained in the form of ions. There are minerals as positive ions, especially Na +, K +, Ca + and also the negative ion which is Cl-(Poedjiadi and Supriyanti, 2007). Ions of HCO3-, HPO4-and SO4 is including negative ions. In the human body, electrolytes will have a function, such as maintaining the osmotic pressure of the body, regulate the distribution of fluid into the body's fluid compartment, maintaining the pH of the body and also be involved in each oxidation and reduction reactions as well as play a role in every metabolic process (Irwan, 2007).

At the time of sweating, the evaporates from the skin contains various electrolytes, such as sodium, potassium, calcium, magnesium, chloride, bicarbonate, phosphate and sulfur (Winarno and Kartawijayaputra, 2007). The function of minerals contained in isotonic drinks is to meet the needs of minerals and also replace minerals lost during sweating.

Benzoic acid is a preservative that is widely used and often used in acidic foods. This material is used to prevent the growth of yeasts and bacteria. According to Desrosier (1988), at levels of 1% or less than the amount allowed, benzoate is effective against yeasts and molds than bacteria. Benzoate is effective at pH 2.5-4.0 because the solubility of its salts larger, it is usually used in the form of salts Na-Benzolate. Benzoate salt breaks down into an
effective form, *i.e.* undissociated benzoic acid (Winarno, 2008). According to Estiasih and Ahmadi (2009), benzoic acid has an antimicrobial activity when in the form of undissociated. Benzoic acid is a preservative which is safe to use because there is a body detoxification mechanism against benzoic acid, so there is no build-up of benzoic acid. Benzoic acid will react with glycine into hippuric acid to be disposed of by the body (Winarno, 2008).

Lychee flavor used is a clear liquid, which is packed in dark colored bottles. Flavor components is relatively changed by the conditions of raw materials, processing and final products. Therefore, the flavor should always be kept at a low temperature to avoid damage. It can be stored in airtight and watertight container, also in dark colored bottles or aluminum bottles to avoid sun damage (Winarno, 2002).

Flavor is added to the isotonic drinks to replace the natural flavor lost during processing, and mask unpleasant flavors (Winarno, 2002). During pasteurization a little loss of smell and taste could happen due to evaporation. The smell of cooked flavor can be formed by the use of heat (Estiasih and Ahmadi, 2009).

Vitamin C is a white crystal soluble in water. In the dry state is fairly stable, but in a state of soluble, vitamin C is easily destroyed by exposure to air (oxidation) especially when exposed to heat. Oxidation is accelerated by the presence of copper and iron. Vitamin C is unstable in alkaline solution, but stable in acid solution. Vitamin C is the most labile vitamins (Almatsier, 2004).

Vitamin C serves to increase the nutritional value of the isotonic drinks as well as replace vitamins lost during heating. In the human body, vitamin C serves to help the absorption of calcium by keeping the calcium in the form of solution (Almatsier, 2004).

**Results of Commercial Isotonic**

Hedonic test performed on isotonic drinks from the market with brands such as: "Mzne", "Vtazne", "100 P" and "P S" aimed to determine which of the four brands most preferred by the panelists. The brand most preferred by the panelists will be used as benchmark in the processing of fruit isotonic drinks. The results (Table 1) of the assessment from 10 panelists showed that, the most preferred brand is "P S" with a value of saltiness 3.50, sweetness, 3.70, off taste 3.00, and overall liking 3.90. The least preferred is 100 P. The runner up of the brand is “Mzne". If product is preferred by the panelist in the overall liking, then all of salty taste, sweet taste and off taste have high rank. Thus, the most important thing in developing formulation is the balanced between sweet taste, salty taste and off taste.

Results on pH of 4 samples was similar with the average pH 4 (Table 2). Total acidity are almost the same for all parameters *i.e.* about 0.2%. However, the Brix level was about 6 except for the “Mzne” which is very low *i.e.* 4. The Brix-acid ratio range was very wide which was very low (23) to very high (33). Two samples which were similar in Brix-acid ratio were P S and 100 P. The most preferred isotonic organoleptically market was P S.

Referring to the data of chemical composition, it still not easy to make a bench mark when making a formula. At least the target should have a Brix-acid ratio of 26 when the Brix is around 6. However, when the Brix-acid ratio was 23 it was also acceptable when the Brix level is around 4. The difference between PS and 100 P is most probably on the level of added flavor. However, even though the Brix- acid ratio was 26, it was still needed to be tested organoleptically.
New Formula

Eight isotonic formulation aims to approach the benchmark of the product characteristics which has the chemical properties to "P S". Looking at the Brix -acid ratio of the eight characteristic of formula, the closest to "P S" was the formula RA and RAF. The value of the Brix -acid ratio was around 30. It seems that both formulation have degree of Brix which was too high while its pH was slightly too high (Table 3).

Hedonic test conducted on isotonic drinks formula aimed to determine which one is most preferred by the panelists. Scoring test aimed to assess the intensity of certain properties, such as sweetness, hardness, and color. Based on the hedonic test results, the most preferred formula was both formula RAF and RNAF which have color score of 3.40 (like), aroma 3.60 (like), and overall liking scores 3.30 (Table 4).

Further scoring showed that the sweet taste of RAF (score 2.9) which was close to a slightly sweet flavor and somewhat sour taste (3.0) (Table 5). Further organoleptic analyses indicated that it was still difficult to conclude since almost all formulas having a good balance between sweet taste and sour taste. Thus, further improvement of the formula (if it is necessary to be improved) should be from chemical and flavor point of view.

Only water alone can cross freely crossing the cellular membranes. Osmosis is defined as the crossing of water from cell part with a low concentration of solute to an area with a higher level of concentration. The final aim of this exchange of water is to balance out solute concentrations. In the human body, the optimal transfer of body liquids happens at normal extra-cellular liquids which is about 300 mOsm. This is also called isotonicity (Moughan 1998, Shirreffs 2003, Hopkins and Woods 2006, Pons Biescas et al. 2011).

Thus, an Osmole is defined as the Avogadro number of particles. One mole of glucose is one osmole. If one mole of NaCl is completely disassociated there will be 2 osmoles of particles i.e 1 osmole of Na⁺ and 1 osmole of Cl⁻. One milliosmole (mOsm) is $10^{-3}$ osmoles. The most common concentration unit is osmolarity, which is defined as osmoles of solute particles per l of solution. Osmolality on the other hand is defined as osmoles of solute particles per kg of pure solvent. Actually, most of solutes are not completely disassociated, and osmolality should be defined as:

$$\text{Osmolality} = \text{osmole/kg H}_2\text{O} = \phi n C$$

where: $\phi$: The osmotic coefficient that determines the degree of disassociation

$n$: The number of particles into which the molecule may disassociate

$C$: The molal concentration of the solution

One mOsm/kg is one mmole/kg in S.I. units (Pons Biescas et al. 2011).

The basic formula has proximate value of osmolality of about 300. Thus the modified of new formula should have similar value.

Application of the New Formula

Thus, the recommended formula of isotonic drink was containing rambutan juice 30 ml, citric acid 0.961 g/l, sucrose 35 g/l and lychee flavor. It is because the formula has the chemical composition closest to P S and has the the highest organoleptic scores.

Compliance with SNI, it is required to decrease the pH since the pH is required to be below 4 while pH of our formula was below 4.5 which is slightly higher than requirement. The pH on the new formula is sufficient for preservation using pasteurisation since pH facilitate the isotonic drink fall into the acid food. International standard such as Codex standard does not require any electrolyte requirement except for the requirement to claim the
excessive recommended daily nutrients. It only regulate the energy which is the maximum of 25% above the recommendation (Anonim, 2011).

CONCLUSION
A new formula of isotonic drink for small scale industry has been successfully developed. This formula is the basic formula with addition of rambutan juice and lychee flavor to make an acceptable isotonic drink. Compare to commercial isotonic drink (PS brand), this formula was comparable with hedonic score 3.30 lower than PS brand (3.90). The pH of this formula has to be lowering to comply to the SNI before can be applied to the small scale industry.

Literature Cited
Tables:

Table 1. Results of Hedonic Scale Scoring on Commercial Isotonic Drink

<table>
<thead>
<tr>
<th>No.</th>
<th>Brand of isotonic drink</th>
<th>Parameters</th>
<th></th>
<th></th>
<th></th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>Salty taste</td>
<td>Sweet taste</td>
<td>Off taste</td>
<td>Overall liking</td>
</tr>
<tr>
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<td>2.60</td>
<td>3.30</td>
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<tr>
<td>2</td>
<td>Mzne</td>
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<td>3.80</td>
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<tr>
<td>3</td>
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<td>3.90</td>
</tr>
<tr>
<td>4</td>
<td>100 P</td>
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<td>2.56</td>
<td>2.00</td>
<td>2.60</td>
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Table 2. Results of Chemical Analyses on Commercial Isotonic Drink

<table>
<thead>
<tr>
<th>No.</th>
<th>Brands of isotonic drink</th>
<th>pH</th>
<th>Total acidity (%)</th>
<th>⁰Brix</th>
<th>Brix Acid Ratio</th>
</tr>
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<tr>
<td>1</td>
<td>Vitzne</td>
<td>4.05</td>
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<tr>
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<tr>
<td>4</td>
<td>100 P</td>
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<td>0.2323</td>
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<td>26.3425</td>
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</table>

Table 3. Results of Chemical Analyses of New Formula of Isotonic Drink

<table>
<thead>
<tr>
<th>No.</th>
<th>Type of isotonic drink</th>
<th>pH</th>
<th>Total acidity (%)</th>
<th>⁰Brix</th>
<th>Brix acid ratio</th>
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<tbody>
<tr>
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<td>R</td>
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<tr>
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<tr>
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</tr>
</tbody>
</table>

Remarks: R: Isotonic drink containing rambutan juice 30 ml, citric acid 0.961 g/l and sucrose 35 g/l; RF: Isotonic drink containing rambutan juice 30 ml, citric acid 0.961 g/l, sucrose 35 g/l and lychee flavor; RA: Isotonic drink containing rambutan juice 30 ml, citric acid 1.922 g/l and sucrose 46 g/l; RAF: Isotonic drink containing rambutan juice 30 ml, citric acid 1.922 g/l, sucrose 46 g/l and lychee flavor; RN: Isotonic drink containing rambutan juice 15 ml, pineapple juice 15 ml, citric acid 0.961 g/l and sugar 35 g/l; RNF: Isotonic drink containing rambutan juice 15 ml, pineapple juice 15 ml, citric acid 0.961 g/l, sucrose 35 g/l and lychee flavor; RNA: Isotonic drink containing rambutan juice 15 ml, pineapple juice 15 ml, citric acid 1.922 g/l, sucrose 46 g/l and lychee flavor; RNAF: Isotonic drink containing rambutan juice 15 ml, pineapple juice 15 ml, citric acid 1.922 g/l, sucrose 46 g/l and lychee flavor.
Table 4. Results of Hedonic Scale Scoring of New Formula of Isotonic Drink

<table>
<thead>
<tr>
<th>No.</th>
<th>Type of isotonic drink</th>
<th>Parameter</th>
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<th>Taste</th>
<th>Overall liking</th>
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<tr>
<td>2</td>
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<tr>
<td>3</td>
<td>RA</td>
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<tr>
<td>4</td>
<td>RAF</td>
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<tr>
<td>5</td>
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<td>3.40</td>
<td>3.60</td>
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</tr>
</tbody>
</table>

Remarks: 1: very dislike; 2: dislike; 3: slightly like; 4: like; 5: very like

Table 5. Results of Scale Scoring of New Formula of Isotonic Drink

<table>
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<tr>
<th>No.</th>
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<th>Sour taste</th>
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<td>3.10</td>
<td>3.40</td>
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<td></td>
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</tr>
<tr>
<td>5</td>
<td>RNA</td>
<td></td>
<td>3.10</td>
<td>2.70</td>
</tr>
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<td>6</td>
<td>RNAF</td>
<td></td>
<td>3.10</td>
<td>2.60</td>
</tr>
</tbody>
</table>

Remarks: Sweet taste: 1: very not sweet; 2: not sweet; 3: slightly sweet; 4: sweet; 5: very sweet; Sour taste: 1: very not sour; 2: not sour; 3: slightly sour; 4: sour; 5: very sour