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An Exploration of Possible Consequences of Indiscriminate Consumption of Bissap Drink in Ghana

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Author's contribution

The sole author designed, analyzed and interpreted and prepared the manuscript.

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ABSTRACT

Aim: To study the chemical and microbiological profiles of dried *Hibiscus sabdariffa* calyces and bissap drinks prepared from these calyces and relate the findings to current prevalence of noncommunicable diseases in the Cape Coast Metropolis.
Study Design: Experimental design.
Place and Duration of Study: The chemical aspect was carried out at the School of Agriculture Laboratory of the University of Cape Coast and the microbiological aspect was at the Food Research Institute, Accra from February to April, 2016.
Methodology: Standard methods and tests were used to determine the chemical and microbiological variables. The Kjeldahl method was used to determine protein, the amount of potassium and sodium were determined using a flame photometer, the complex metric titration was used to determine the amount of calcium and magnesium. The amounts of Iron, Copper and Zinc were also determined. The pH was measured using a pH meter, titratable acidity using NaOH titration and total sugars using the colorimetric method. Total Viable Cell count and the

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identification of probable presence of *Escherichia coli*, *Salmonella species* and Coliforms in the bissap drinks were also carried out.
Results: Â remarkable amount of potassium was found in the range of 10874.4 –1667. 1 ug/g). Other elements in the bissap were also determined in descending order as sodium (5425.3 - 6058.4 ug/g), iron (2052.9 - 2099.6 ug/g), zinc (90.6 – 94.1 ug/g) and phosphorus (1175.5 – 1187.2 ug/g). Coliforms and *Escherichia coli* were identified while *Salmonella species* were not identified. Low pH, titratable acidity and total sugars ranged from 2.39 – 2.52; 0.27 – 0.30 and 7.2 – 14.0 respectively.
Conclusion: *Hibiscus sabdariffa* with its chemical constituents may promote health when used in preparing bissap drink and consumed but the acidity and high sugars present in the drink may overshadow its health benefits and cause diseases especially if taken in large quantities indiscriminately. It's recommended that consumers drink bissap in moderation to avoid its probable health consequences.

Keywords: Hibiscus sabdariffa; bissap; non-communicable diseases; health; beverage.

1. INTRODUCTION

Non- alcoholic drinks form part of the daily diets of most Ghanaians. These include fruit juices, soft drinks/soda, carbonated drinks and Ghanaian local drinks. These drinks are often taken alone or with snacks and meals. Frequent and indiscriminate consumption of these drinks may be contributing to the dwindling or rise in non-communicable diseases like diabetes, hypertension and cancer in Ghana. There have been reports from the United States of America and other developed countries where half of the population of children above 2 years consume drinks on a daily basis leading to rising rates of weight gain, obesity, fatty liver disease, high blood pressure and type II diabetes [1]. Reports from one of the three major hospitals in Cape Coast- the University of Cape Coast hospital, indicates that a total of 1080 cases of diabetes were reported in 2010, 854 in 2011, 1115 in 2012, 507 in 2013, 702 in 2014 and 773 in 2015 [2]. Also reported cases of hypertension in the Central region were 12135 for 2009, 17797 for 2010, 25729 for 2011 and 30898 for 2012 respectively [3].

While the quality of fruit juices and other non-alcoholic beverages is strictly being maintained in the developed countries under several laws and regulations, unfortunately, in many developing countries including Ghana, manufacturers are not much concerned about the chemical, microbiological safety and hygiene of fruit juices because of lack of enforcement of law. For example, in North America, both the USA and Canada have Food Guides that guide people as to what to eat and drink to stay healthy. Others are the Healthy weights for Healthy kids in Canada and Chronic Disease Prevention Alliance of Canada, (CDPAC'S) policy, which regulate advertising of sweetened drinks especially to kids as a way of reducing obesity and other sugar related diseases [4,5]. Several health claims are made about fruit juices but the small amounts of vitamins and antioxidants in these juices do not make up for the large amount of sugar [6]. A single serving of the so-called healthy fruit juice has been found to contain the same amount of sugar as three-and-a-half doughnuts or 13 Hob Nob digestive biscuits [7]. There have also been reported cases of transmission of certain human diseases through the intake of juice and other drinks [8].

In Ghana, consumers believe locally produced drinks are healthier than their imported counterparts probably because they can easily identify the ingredients they are familiar with in these drinks [9,10]. According to [10], consumers follow the sweetness and tastes of these locally produced drinks and are mainly concerned about the relief these drinks provide which is mainly to put out their thirst and also to delay hunger as they commute in vehicles or on foot from one place to another. Sobolo or bissap is a popular traditional drink that has emerged in Ghana in recent times and is being produced and sold by several people. The drink is produced from the flowers or calyces of *Hibiscus sabdariffa.*

Hibiscus sabdariffa bears red flowers which have been used over centuries for several purposes including the preparation of juice and tea. It has different names in different places across the world. In Senegal, it is known as the "national drink" and is sold in all places in the country by various vendors. It's known in other Western African countries as Sorrel, Bissap and O'seille de Guinea and in places such as Egypt and

Sudan as Karkaday. In East Africa, it is infused into a drink called "Sudan tea" which is used to treat coughs. Several health claims have been made about *Hibiscus sabdariffa* and is known to be used as an antiseptic, aphrodisiac, astringent, cholagogue, demulcent, digestive, diuretic, emollient, laxative, cooler, sedative, and tonic. The Chinese use it to treat liver disorders and high blood pressure [11]. In Northern Nigeria the *Hibiscus sabdariffa* drink is traditionally used as a treatment for lowering high blood pressure [12].

Scientific studies indicate that the *Hibiscus sabdariffa* drink can indeed lower blood pressure [13] and inhibit the angiotension converting enzymes (ACE) that play a part in raising blood pressure [14]. In a study [15] suggested that the integration of daily consumption of hibiscus tea in the diet could lower high blood pressure in adults with a pre-and mild hypertension. As such it may help as an effective part of the dietary adjustments for a hypertensive. *Hibiscus sabdariffa* extract is known to have antioxidant, anti-diabetic, antibacterial, antihypertensive, lipid lowering properties and antimicrobial properties [16,17]. With all the amazing scientific reports that back the numerous health potentials of *Hibiscus sabdariffa*, promoting the consumption of the bissap drink requires a study of its constituents. Intriguing observation shows that the bissap drink easily stains any material it comes into contact with. The study looked at the chemical constituents of dried *Hibiscus sabdariffa* calyces and bissap drink prepared from these calyces. The pH, titratable acidity and total sugars of the bissap drink were also determined as well as the identification of probable presence of *Escherichia coli*, *Salmonella species* and Coliforms in the bissap drinks.

2. MATERIALS AND METHODS

2.1 Sample Collection

Two main samplings were carried out. First, *Hibiscus sabdariffa* calyces were purchased from the Kotokuraba market in Cape Coast. Equal amounts of the calyces were weighed using a Kitchen scale into four clean containers labeled as A, B, C, and D. The contents of A were milled into powder using a Kenwood blender. Sample B was soaked in hot water. Sample C was soaked in hot water and pineapple juice, and spices (ginger, peppercorn and clove) added. The fourth sample D was soaked in hot water, pineapple juice, spices and 12% sugar added. Chemical analysis of samples A, B, C and D were carried out at the School of Agriculture Laboratory of the University of Cape Coast to determine the various nutrients in each. Analysis was run in triplicate using different amounts as follows: 1 (ug/g), 2 (ug/g) and 3 (ug/g). Secondly, four different samples of already prepared bissap drinks were purchased from four different vendors and added to the self-prepared one sample D and analyzed. Titratable acidity, pH and temperature of these drinks were determined.

2.2 Determination of Protein

The Kjeldahl method for protein determination was used to determine the amount of protein in samples A, B, C and D. The digestion mixture contained 350 ml of hydrogen peroxide, 0.42 g of selenium powder, 14 g Lithium Sulphate and 420 ml Sulphuric acid. Between 0.10 to 0.2 g of the oven-dried ground sample was weighed into a 100 ml Kjeldahl flask and 4.4 ml of the digestion reagent added to digest samples at 360 $^{\circ}$ for two hours. The blank digestions without the addition of samples were obtained following the same procedure. Digests obtained after digestions were transferred quantitatively into 50ml volumetric flasks and distilled water added to make up the volume.

A steam distillation apparatus was set up and steam passed through samples for about 20 minutes. After flushing out the apparatus, a 100 ml conical flask containing 5 ml of boric acid indicator solution was placed under the condenser of the distillation apparatus. An aliquot of the sample digest was transferred to the reaction chamber through the trap funnel. Alkali mixture (10 ml) was added to commence distillation immediately and about 50 ml of the distillate was collected. The distillate was titrated against 1/140 Ml HCI from green to the initial colour of the indicator (wine red). Digestion blanks were treated the same way and subtracted from the sample titer value.

Calculation

$$N\ (\%) = \frac{(T-B) \times M \times 14.007 \times 100}{\text{Sample Weight (mg)}}$$

Where

M = Molality of Acid
S = Sample titer value

B = Blank titer value
Protein = %N *6.25

2.3 Determination of Potassium and Sodium

The amount of potassium and sodium in the digested samples were determined using a flame photometer. Working standards for both potassium (K) and sodium (Na) were prepared as: 0, 2,4,6,8 and 10 μg/ml and together with the sample solutions were aspirated individually into the flame photometer. Their emissions (readings) were then recorded. A calibration curve was then plotted using the concentrations and emissions of the working standards. Concentrations of the sample solutions were extrapolated from the standard curve using their emissions. The following calculations were obtained using [18].

Calculation

$$\mu gKg = \frac{C \times \text{solution volume}}{\text{Sample weight}}$$

$$\mu gNa/g = \frac{C \times \text{solution volume}}{\text{Sample weight}}$$

2.4 Determination of Calcium and Magnesium

The amount of soluble calcium and magnesium were determined by versenate titration method. The cations were chelated using ethylene di-amine-tetra-acetic acid (EDTA). The combined amount of calcium and magnesium were determined and the amount of only calcium was also determined. To obtain the amount of only magnesium, the difference between the combined calcium and magnesium, and calcium alone was used. For the determination of the combined amount of calcium and magnesium, an aliquot of 10 ml of the sample solution was placed in a 250 ml conical flask and the solution was diluted to 150 ml with distilled water. Buffer solution (15 ml) and 1 ml each of potassium cyanide, hydroxylamine hydrochloride, potassium ferro-cyanide and tri-ethanolamine (TEA) were further added. Five drops of Erichrome Black T (EBT) were also added and the solution was titrated against 0.005 M EDTA. Calcium was determined by pipetting 10 mL of the sample solution into a 250 conical flask and diluted with distilled water to 150 mL. Potassium cyanide (1 mL), hydroxyl-amine-hydrochloride (1 mL), potassium ferro-cyanide (1 mL) and TEA (1 mL) and 5 drops of calcon indicator were added, and the solution was titrated with 0.005 M EDTA.

Calculations

$$\% Ca = \frac{0.005 \times 40.08 \times T}{\text{Sample weight}}$$

$$\% Mg = \frac{0.005 \times 24.9 \times T}{\text{Sample weight}}$$

Where

T = titer value

2.5 Determination of Iron, Copper and Zinc

To determine the amount of Iron, Copper and Zinc. Standard solutions of 1, 2 and 5 μg/mL solutions of Fe, Cu and Zn were prepared. The standard solutions were aspirated into the atomic absorption spectrophotometer (AAS) and the respective calibration curves were plotted on the AAS. As the sample solutions were aspirated, their respective concentrations were provided.

Calculations

$$Fe\ (\mu g/g) = \frac{C \times \text{solution volume}}{\text{Sample weight}}$$

$$Cu\ (\mu g/g) = \frac{C \times \text{solution volume}}{\text{Sample weight}}$$

$$Zn\ (\mu g/g) = \frac{C \times \text{solution volume}}{\text{Sample weight}}$$

Where

C=microgram per ml of the individual element

The four bissap samples collected were labeled as E, F, G and H. The chemical constituents of these were not carried out because it was assumed that since these samples were all produced from the *Hibiscus sabdariffa* flowers and in the same way, these will have similar amounts of the elements determined in sample D. However, the amount of sugar added to these drinks could differ as well as the conditions under which they were produced. Thus, pH, Titratable acidity and temperature and the microbiological analysis of these drinks were determined.

2.6 pH and Titratable Acidity

2.6.1 Materials

pH meter or phenolphthalein, burette, burette clamp and stand, gram scale, graduated cylinder, beakers, 0.1 N NaOH solution B. Optional: magnetic stirrer & stir bar, automatic tiltrotor II.

2.6.2 Procedure

About 50 mls of clear bissap drink was measured at room temperature. The pH of each sample was measured using a pH meter and the value recorded. For each sample, 6 grams of bissap drink was weighed out into a 100 ml beaker and 50 mls of water was added to each sample. Each sample was titrated with 0.1 N NaOH to an end point of 8.2 (measured with phenolphthalein indicator) and the milliliter (mls) of NaOH used was recorded. Titratable acidity was calculated using the following formula:

> % acid = [mls NaOH used] x [0.1 N NaOH] x [milliequivalent factor] x [100]) / grams of sample
>
> Commodity Predominant Acid Milliequivalent Factor Stone fruit, Apples, Kiwifruit - Malic Acid 0.067
> Citrus -Citric Acid 0.064
> Grapes - Tartaric Acid 0.075
> Source: [19]

2.7 Determination of Soluble Carbohydrates (Total Sugars)

The Anthrone colorimetric method was used to determine the concentration of the total sugars in bissap drinks. The anthrone reagent was prepared by adding 760 ml concentrated H_2SO_4 to 330 ml water in a boiling flask and kept cool while mixing. A gram each of anthrone and thiourea were added and dissolved using a magnetic stirrer. The solution was transferred into a dark bottle and left at 1℃ for 2 hours prio r to use. Stock solution (1 ml is equivalent to 0.25 mg glucose); 0.250 g D-glucose was dissolved in water and diluted to 1 liter. Working standards ranging from 0 – 20 ml stock solution were pipetted into 50 ml flasks such that 2 ml of each standard gave a range from 0-0.20mg glucose and diluted to volume.

2.7.1 Procedure

For the extraction, 50 g of the sample was weighed into a 50 ml conical flask, 30 ml of distilled water was added and a glass bubble placed in the neck and left to simmer gently on a hot plate for 2 hours. This was topped up to 30 ml, allowed to cool slightly then filtered into a 50 ml volumetric flask using No. 44 Whatman paper. This was diluted to volume after cooling. A blank solution was prepared through the same procedure.

2.7.2 Colour development

A set of boiling tubes were each filled with 2 ml of the standard. Other boiling tubes were filled with 2 ml each of extract and water blank. Standards and samples were treated the same way. Anthrone solution (10 ml) was added rapidly to mix and tubes immersed in running tap water. Tubes were then placed in cold water and again placed in a beaker of boiling water in a dark fume cupboard, and boiled for 10 minutes. Tubes were then placed in cold water and allowed to cool. Sugars reacted with the anthrone reagent under acidic conditions to yield a blue-green colour. There was a linear relationship between the absorbance and the amount of sugar that was present in the original sample. This method determines both reducing and non-reducing sugars because of the presence of the strongly oxidizing sulfuric acid. Optical density was measured at 625 nm and a calibration graph was prepared from the standard and used to obtain mg glucose in the sample aliquot. The blank sample was treated in the same way and subtractions done.

2.7.3 Preparation of bissap drink (5 litres)

Ingredients

- 3 cups of dried hibiscus leaf
- 3 cups of sugar
- 3 cups of boiling water (for the sugar syrup)
- 5 litres of boiling water (for the brewing of the leaves)

Procedure

- Pour the leaves into a big bowl
- Pour in boiling water
- Let it sit for about an hour or more (cover it while it brews)
- Make a syrup from sugar and boiling water then stir
- Mix the syrup and sobolo juice.
- Add some squeezed pineapple juice.
- Add some spices, (cloves, ginger, peppercorns

2.8 Microbiological Analysis

The Ghana Standard Authority uses the following categories as standards for analyzing drinks:

1. Carbonated soft drinks/Carbonated malts - recommended tests are yeast, molds and APC
2. Concentrated fruit juices preserved exclusively by physical means- recommended tests are yeasts, molds, APC and Coliforms
3. Fruit juice prepared without physical or chemical preservatives - recommended tests are APC, *Escherichia coli*, and *Salmonella species.*
4. Fruit juices, squashes and cordials preserved by physical and chemical means – recommended tests are APC, Coliforms, yeast and molds

The bissap drink was identified to belong to the third group and thus the following were determined. Bacteria, Coliforms, *Escherichia coli,* yeast and molds were cultured, enumerated and isolated for samples, D, E, F, G and H.

2.8.1 Total viable cell count

Bissap drinks in bottles were shaken vigorously to mix up well before opening. The caps were removed and the tops briefly flamed using a Bunsen burner. The preparation of a series of six fold dilutions from the initial dilution was made. From each of the samples, aliquots of 1.0 ml was collected and inoculated into 9 ml of peptone water in screw capped test tubes. Plate Count Agar (PCA), Tryptone Soya Agar (TSA) and Dichloran Rose-Bengal Chloramphenicol Agar (DRBC) were used to culture, enumerate and isolate bacteria; coliforms and *Escherichia coli,* yeast and molds respectively. Each dilution was then dispensed into sterile Petri dishes containing 15 ml of medium. The plates were then rotated to mix the molten medium and the sample dilution then allowed to solidify before incubation at 30℃ for 48 hrs. After incubation, the visible colonies on selective plates (≥ 15 to ≤300 colonies) were counted and the results were calculated based on the count and the dilution factor.

2.8.2 Confirmation of *Escherichia coli* and coliforms

For coliform detection, pre-enrichment of organisms into test tubes containing Lauryl Tryptose broth with inverted Durham tubes as described by Speck (1976) was used. Tubes were incubated for 24 hours at 37℃. About 0.2 ml of the sample was inoculated by spread plate methods onto Violet Red Bile Agar (Oxoid), *Escherichia coli* Broth (SPS) and coliform count respectively. Plates were incubated for 24 – 48 hours at 37℃ for colony formation, except for Sabouraud Dextrose agar (SDA) that was left for 24-72 hours at 28 ± 2℃. After incubation, the colonies were counted using a colony counter (Stuart Scientific, UK). After pre-enrichment, samples were plated out onto Nutrient agar, MacConkey agar and Sabouraud Dextrose agar for 24 hours incubation at 37℃ and 28 ± 2℃. Colonies were isolated and purified by repeated sub-culturing for further identification. Distinctive morphological properties of each pure culture such as colony form, elevation of colony and colony margin were observed. Further identification of bacterial isolates was carried out. Fungal isolates were identified based on macroscopic and microscopic features [20].

3. RESULTS AND DISCUSSION

Through literature review, different names were found for the bissap drink in different countries and places. These names have been catalogued in Table 1 below.

Table 1. Different names given to the bissap drink by different countries

Country	Name(s)
Indonesia	Rosella, Rosella fruit
Ghana	Bissap, Sobolo
Senegal	Bissap
France	Bissap
Mali	Dah, Dah Blendi, Datou,
Gambia	Furundu
Nigeria	Wongo
Iran	Zobo
Egypt	Chaye-Torosh
Saudi Arabia	Karkade
Sudan	Karkade
Namibia	Karkade
Caribbean	Omutete
Latin America	Sorrel
Mexico	Sorrel
Panama	Flor de Jamaica
Burkina Faso	Saril
Niger	Bikalga Dawadawa-botso

The *Hibiscus sabdariffa* flowers (bissap.) were found to contain high amounts of potassium, sodium, iron and phosphorus (Table 2). When

the flowers were soaked in hot water and analyzed, the amounts of potassium, sodium, iron and phosphorus reduced drastically (Table 3) but with subsequent addition of pineapple and spices to the soaked flowers the amounts of these elements increased slightly over that soaked in water without any added pineapple and spices (Table 4).

When sugar was added to the soaked flowers containing pineapple and spices, there was not much change in the amount of potassium, sodium, iron and phosphorus as reported in Table 4. For each of the samples A, B, C and D, three different amounts were analyzed 1 (ug/g), 2 (ug/g and 3 (ug/g). On the average the amount of potassium found in the triplicate of sample "A" was 11298.0 ug/g, sodium 5725.17 ug/g, iron 2069.6 ug/g and phosphorus 1181.5 ug/g.

Although, the amount of the various elements was reduced, when *Hibiscus sabdariffa* flowers soaked in water and pineapple juice, spices and sugar were added, the amounts of potassium, sodium and iron were quite remarkable (Table 5). Bissap has become so popular a drink in Ghana that competes with imported international non-alcoholic drinks like coca cola, pepsi, and sprite among others. Reports by [21] show that Sobolo (Bissap) has certainly come to add to other locally brewed drinks such as pito, asana, tose and palm wine. The *Hibiscus sabdariffa* flowers (bissap) prior to preparing the drink seems to have more protein (Table 2). Once water is added the amount of protein reduced. However, with the addition of pineapple juice and spices, the amount of protein increased slightly again to 5.6 ug/ml, 5.9 ug/ml and 6.1 ug/ml. Probably, the pineapple and spices added could also contribute some amount of protein to the drink. This cannot be said about the sugar which was added to sample D. The addition of sugar rather reduced the amount of protein in the drink to 4.1 ug/ml.

Optimal levels of potassium have been reported to promote good heart function, skeletal and smooth muscle contraction but could shut down the heart and nervous system if it is too low or too high [22]. Patronizing soft drinks has been a contentious subject for both public health and public policy over the years. Often studies focus on finding probable connection between consumption of soft drinks and their related health problems. Several health claims have been made in reference to the health benefits of consuming bissap but looking critically at the supposed benefits, one foresees the possibility of these same nutrients causing health problems if not taken in moderation. For example with all the

Table 2. Determination of the chemical constituents of *Hibiscus sabdariffa* flowers (bissap) in microgram/gram (ug/g)

Constituents	Experiment 1 (ug/g)	Experiment 2 (ug/g)	Experiment 3 (ug/g)
Protein	5.8	5.9	5.9
Potassium (K)	10874.4	11667.1	11352.5
Sodium (Na)	5425.3	6058.4	5691.8
Iron (Fe)	2099.6	2056.3	2052.9
Zinc (Zn)	92.8	90.6	94.1
Copper (Cu)	14.6	14.3	13.4
Phosphorus (P)	1175.7	1181.6	1187,2
Calcium (Ca)	0.9	0.9	1.1

Table 3. Determination of the chemical constituents of *Hibiscus sabdariffa* flowers (bissap) soaked in water microgram/milliliters (ug/ml)

Constituents	Experiment 1 ug/g	Experiment 2 ug/g	Experiment 3 ug/g
Protein	4.8	4.8	4.8
Potassium (K)	559.0	541.5	541.5
Sodium (Na)	254.0	238.0	254.0
Iron (Fe)	30.2	30.2	31.2
Zinc (Zn)	2.2	2.4	2.4
Copper (Cu)	0.8	1.0	0.8
Phosphorus (P)	54.3	53.7	54.3
Calcium (Ca)	0.04	0.03	0.03

Table 4. Determination of the chemical constituents of *Hibiscus sabdariffa* flowers (bissap) soaked in water with added pineapple and spices in microgram/milliliters (ug/ml)

Constituents	Experiment 1 (ug/g)	Experiment 2 (ug/g)	Experiment 3 (ug/g)
Protein	5.6	5.9	6.1
Potassium (K)	628.8	646.2	628.8
Sodium (Na)	285.7	301.6	285.7
Iron (Fe)	32.0	31.6	31.6
Zinc (Zn)	2.6	2.8	2.6
Copper (Cu)	1.8	1.8	1.8
Phosphorus (P)	66.0	68.1	67.4
Calcium (Ca)	0.03	0.03	0.03

Table 5. Determination of the chemical constituents of *Hibiscus sabdariffa* flowers (bissap) soaked in water with added pineapple, spices and sugar in microgram/milliliters (ug/ml)

Constituents	Experiment 1 (ug/g)	Experiment 2 (ug/g)	Experiment 3 (ug/g)
Protein	4.1	4.1	4.1
Potassium (K)	611.4	602.6	611.4
Sodium (Na)	285.7	285.7	254.0
Iron (Fe)	31.8	30.4	30.8
Zinc (Zn)	2.4	2.2	2.4
Copper (Cu)	1.8	2.0	2.0
Phosphorus (P)	66.7	67.4	66.7
Calcium (Ca)	0.03	0.03	0.04

health benefits of potassium discussed earlier in this paper, the fact still remains that excessive intake of bissap could raise the levels of potassium which could become detrimental to the health of consumers. As reported [23] there is absolute proof to support the reality of health risks associated, especially with over consumption of soft drinks.

Table 6 shows the "total" or viable bacterial growth of the bissap samples analyzed. With the exception of sample F, the rest produced remarkable viable bacterial growth. Though remarkable, the growth levels were above that recommended by the Ghana Standards Board (1.0×10^2) [24]. The absence of *Salmonella species* in any of the samples is good; at least it shows that the drink is not likely to cause Salmonella food poisoning. However, Coliforms were present and could be a concern for the safety of drinking bissap. Coliform bacteria may not necessarily be a reason for diseases but could be indicators of pathogenic organisms that cause diseases such as dysentery, hepatitis, typhoid fever, cholera and other illnesses [25]. With the exception of sample G, *Escherichia coli* levels were quite low in all the samples. These organisms belong to the Coliform group and are used as an indicator to examine the possible presence of other more harmful microbes such as Shigella [25]. The presence of yeasts and molds in the bissap samples may not necessarily be a serious food safety issue but could reduce the quality of the bissap by producing off flavors in the drink. The yeast and mold levels in all the samples were above that recommended by the Ghana Standards Board (5.0×10^1) [24] Thorough cleaning of equipment used for producing the bissap drink could help reduce the incidence of yeast and molds in the bissap.

Table 6. Microbiological analysis of different samples of bissap drinks

Bissap drink	PCA	*Salmonella spp.*	Yeasts	Molds	Coliform	*Escherichia coli*
D	132×10^6	Absent	152×10^4	0×10^4	336×10^1	0×10^1
E	494×10^6	Absent	23×10^6	1×10^6	0×10^1	0×10^1
F	20×10^6	Absent	0×10^1	0×10^1	0×10^1	0×10^1
G	482×10^6	Absent	52×10^3	0×10^1	113×10^3	188×10^3
H	177×10^6	Absent	48×10^6	1×10^1	0×10^1	0×10^1

Table 7. Measured pH, titratable acidity and brix for all tested samples

Bissap drink	pH	Temperature °C	Titratable acidity g/100 ml	°Total sugar (Carbohydrate)
D	2.40	23.1	0.50	14.0
E	2.42	23.8	0.36	9.2
F	2.52	23.5	0.27	7.2
G	2.40	23.7	0.42	13.6
H	2.39	23.8	0.40	11.0

Low pH and high titratable acidity of juices and cola-based beverages have been reported to be significant causes of dental erosion [26]. Further, they added that demineralization of enamel by drinks with pH levels below 4.0 tend to be more severe. From Table 7 above, the average pH for the bissap samples was 2.40 indicating that when consumers often take bissap drink they may end up with demineralization of enamel and dental erosion. Also, from Table 7 below, the total sugars (measured as total carbohydrates) obtained from the bissap drinks ranged from 7.2 to 14.0 which may be considered as quite high. Although it would have been ideal to determine the Brix of the bissap drinks so as to get the right concentration of sugars in the drinks and relate it directly to probably incidence of high blood glucose, this was not possible because of non-availability of a brix meter or refractometer. Rather total carbohydrates was determined to give an idea of total sugars in the drinks. With high concentration of potassium in bissap drink, consumers need to drink moderate amounts of it to avoid dehydration and possible acquisition of diabetes [27]. It has been reported that food safety and factors that affect it have been key in rising issues in the food supply chain and have drawn various governmental and regulatory bodies to show interest [28].

4. CONCLUSION

In conclusion, bissap prepared from *Hibiscus sabdariffa* calyces do have rich constituents that could be beneficial to health. However, with the high acidity nature and level of total sugars often added to these drinks in the Cape Coast Metropolis could override the health benefits when taken in large volumes indiscriminately. Thus, instead of promoting health, it could rather promote diseases in consumers who over consume bissap. It is recommended that consumers should drink bissap responsibly and in moderation in order to obtain the maximum health benefits it comes with. Also, to obtain the maximum benefits from bissap, it could be prepared with very little sugar or no sugar.

COMPETING INTERESTS

Author has declared that no competing interests exist.

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