Evaluation of the chemical contents of *Zobo* (*Hibiscus sabdariffa*) flavoured with natural spices

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Abstract

Zobo is an important healthy drink made from the dried calyces *of Hibiscus sabdariffa by boiling water extraction. An equal volume of the basal extract was flavoured separately with conventional strawberry essence (Zobo-S), alligator pepper (Zobo-A), and clove (Zobo-C) and evaluated for chemical and sensory properties using established methods. The obtained range of values for mineral (4.70-578 mg/L) and vitamins (0.05-0.29 mg/100 g) were in moderate amounts with potassium being the most abundant mineral element (ranged 522-578mg/L) and highest in Zobo-A (578 mg/L). Zinc, an antiviral and important element for the body immune system was significantly ($p<0.05$) higher in Zobo-C (13.34 mg/L) and Zobo-A (8.33 mg/L) than Zobo-S (7.14 mg/L) making Zobo with spices a better choice drink for a preventive measure against infectious viral-diseases like COVID-19. Total titratable acidity ranged from 0.93-1.06 g/100 ml and was highest in Zobo-S (1.06g /100 ml) implying the Zobo made with spices were less acidic. Also, tannin content was improved with the spices and highest in Zobo-C (0.44 mg/100 g) while Zobo-A was the most preferred in overall acceptability. Thus, the natural spices improved the minerals, phytochemical contents and sensory acceptability of Zobo drink. This could proffer potential health benefits to the consumers and encourage the use of natural spices in the preparation of food and drinks rather than the chemical-based conventional flavour essence.*

Keywords: *Zobo drink, natural spices, conventional essence, minerals, vitamins,*

1.0 Introduction

One important vegetable, potent in naturally occurring antioxidant and antiviral compounds is *Hibiscus sabdariffa* (Roselle) called 'Isapa' in the Yoruba language (Chen *et al*., 2013; Aworh, 2014). *Hibiscus sabdariffa* belongs to the superorder *Malvaceae* and it is believed to originate from East Africa (Ilondu and Iloh, 2007). There exists credible documentation on the culinary and medicinal properties of the leaf and calyx extracts of *Hibiscus sabdariffa* due to the presence of a high level of polyphenolic compounds and anthocyanins with notable health benefits (Oboh *et al*., 2011; Okereke *et al*., 2015; Zhen *et al*., 2016; Adeooye *et al*., 2019). The green leaf of the *H. sabdariffa* plant is employed in soup preparation among many tribes in the countries of the world like the Yoruba tribe in Nigeria (Zhen *et al*., 2012). However, the popular dried calyces of the red cultivars are mainly employed in the preparation of a refreshing drink popularly referred to as *Zobo or Soborodo* which though, originated from the northern part is

now known among all tribes in Nigeria (Adeooye *et al*., 2019). As such, robust research has been done on Z*obo* drink; its nutritive importance, health benefits, and safety; Olayemi *et al*. (2011) assessed the nutritional quality of three varieties of *Zobo* (*Hibiscus sabdariffa*) subjected to the same preparation condition; Oboh *et al*. (2011) reported the medicinal value and low glycemic index of *Zobo* drink with good acceptability by all age groups, Awe *et al*. (2013) reported the antioxidant properties of cold and hot water extracts of Cocoa, *Hibiscus sabdariffa* extract (*Zobo*), and ginger beverage blends while Adeniji (2017) assessed the nutritional, sensory and microbial quality of *Zobo* fortified with soybean milk. Thus, *Zobo* drink was found to have tremendous nutritional and health benefits.

Consequently, some Nigerians prefer *Zobo* drink to carbonated drinks because it is a drink close to nature and rich in minerals, vitamins and antioxidants for the acclaimed health benefits (Ogiehor and Nwafor, 2004). The health benefits of *Zobo* drink include; ability to boost the immune system; treat high blood pressure, high cholesterol, disturbed digestive, and inflammatory problems (Olayemi *et al*., 2011). Other phases of previous works also expressed the antioxidant, phytochemical capacity; medicinal values, and other health benefits of *Hibiscus sabdariffa* extracts; the water extraction of which is equivalent to *Zobo* drink (Yadeng *et al*., 2005; Okereke *et al*., 2015; Zhen *et al*., 2015). The drink is gaining wide acceptance, being consumed by several millions of people from different socio-economic classes and backgrounds in the West Africa sub-region, especially amongst the Nigerian youths and other age groups who see *Zobo* drink as an alternative source of cheap and relaxing nonalcoholic drink in social gathering (Ogiehor and Nwafor, 2004; Oboh *et al*., 2011).

Conventionally, *Zobo* drink is flavoured with commercial flavouring essence especially strawberry as having a similar colour scheme to *Zobo* drink. Nevertheless, the literature is scarce on the comparative analysis between the conventional *Zobo* and the supportive roles that natural spices could play in the acclaimed health benefits of *Zobo* drink.

Spices are aromatic vegetables in the form of the root, tubers, seeds, bark of trees, or leaves which in addition to their antioxidants properties are usually added to improve the flavour of foods and drinks (Oyewale *et al*., 2020). Spices are reported to be potent sources of antioxidants, anti-inflammatory, anti-viral, and other natural immune-balancing agents (Igual et al., 2010; Saxena *et al*., 2012; Adeooye *et al*., 2019; Oyewale *et al.*, 2020). The antioxidative and immune-balancing potentials of spices, like other herbs and vegetables, are owed to their embedded bioactive compounds (polyphenols) or phytochemicals (Adeooye *et al*., 2019; Oyewale *et al*., 2020). Phytochemicals in spices and vegetables are beneficial to human health in disease prevention, even though, they are not established as essential nutrients (Egbuna and Ifemeje, 2015).

Among the natural and indigenous spices locally available in Nigeria are ginger (*Zingiber officinale*), alligator pepper (*Aframomum danielli*, "Atare"), and clove (*Syzygium aromatic,* "kanafuru"). They are used in the form of extract, paste, or dried powder to flavour many food items ranging from soups, stews, juices/drinks, traditional cuisines, or as medicine such as *agbo* or *aseje* among the Yorubas. Recently, alligator pepper has been identified as an indigenous herb used in developing medication for COVID-19 (Olarinde 2022). Hence, the addition of natural spices to the *Zobo* drink will doubtless improve its health benefit and support its

immune functioning potential. Especially, in this era of the global COVID-19 pandemic when a low or impaired immunity is the major cause of lost battles against human infections and diseases which cumulated into preventable death among all ages. Therefore, flavouring *Zobo* with spices presents naturally refreshing drink devoid of any chemical additive and regular/moderate consumption could improve the immune system of the consumers to effectively manage any sudden infections and diseases (Oboh *et al*., 2011) like COVID-19 (Olarinde, 2022).

Contrarily, the conventional and commonly hawked-around *Zobo* drinks in Nigeria are only stuffed with sweeteners and chemical-based commercial flavour essence that have no health benefit but *Zobo* made with natural spices could find better replacements that will add more health benefits to refreshing. Thus, the aim of this study is to determine the chemical and sensory properties of *Zobo* flavoured with natural spices in comparison with the conventional *Zobo* drink with the view of encouraging the use of natural spices in the preparation of *Zobo*.

2.0 Materials and Methods

2.1 Materials

The dried, red *Hibiscus sabdariffa* calyces, fresh ginger (*Zingiber officinale)* rhizomes, matured and dried alligator pepper (*Aframomum danielli)* pods, dried cloves (*Syzygium aromaticum*), commercial strawberry essence, and sugar were purchased from a local market in Ondo, Ondo State, Nigeria. Chemicals used for the analyses are of analytical grade.

2.2 Methods

2.2.1 Preparation of *Zobo* drink samples

The dried alligator pepper and clove were separately ground into powder while the fresh ginger rhizomes were washed and grated into a paste using a Warring blender (Model: HGB2WTG4). The dried, cleaned calyces of *Hibiscus sabdariffa* were washed and boiled in water for 10 min with the ginger paste which has become a household spice and method used in making *Zobo* drink in Nigeria (Adeniji, 2017). The mixture is coarsely sieved to remove the spent materials, then sugar was added and stirred to dissolve, this serves as the basal *Zobo.* An equal volume (1 litre each) of this basal *Zobo* drink while hot, was separately flavoured with alligator pepper (*Zobo*-A) and clove (*Zobo*-C) powder, stirred and covered for 30 min for the spices to infuse into the drink. Another equal volume of the basal *Zobo* drink was cooled and flavoured with the commercial strawberry essence as the usual practice in the preparation of conventional *Zobo* drink in Nigeria mainly for the impartation of aroma (Ukwo *et al*., 2019). The resulted mixture in each case was finely filtered using a muslin cloth. The drinks were cooled at room temperature, packaged in sterilized, labelled pet bottles, and refrigerated pending analyses. The recipe employed for the preparation is presented in Table 1 while the flow chart for the traditional method of preparation is depicted in Figure 1 adapted from Adeniji (2017) with modifications.

Table 1: Recipe for the preparation of *Zobo* drinks

Ingredient	Quantity
Dried red *Hibiscus sabdariffa* calyces	250 g
Portable water	5 litres
Fresh ginger rhizomes	120 g
Granulated sugar	100 g/litre
Conventional strawberry essence	5 ml/litre (*Zobo*-S)
Alligator pepper seed powder	5 g/litre (*Zobo*-A)
Clove seed powder	5 g/litre (*Zobo*-C)

2.2.2 Determination of mineral content

The mineral elements were determined using the dry ashing procedure of official method 925.10 of AOAC (2003). A 5 ml sample in a well-cleaned porcelain crucible was evaporated on a Bunsen flame to pre-ashed. Thereafter, the sample was then subjected to dry-ashing at 550^{o}C in a Muffle furnace. The resultant grayish-white ash was dissolved in 5 ml HNO_3/HCl (1:1) while heating on a hot plate at the boiling temperature of the solution until brown fumes disappeared. To the remaining content in each crucible, 5 ml distilled water was added, and the mixture was heated until a colourless solution was obtained.

The mineral solution in each crucible was transferred into a 100 ml volumetric flask by filtering through a Whatman No. 42 filter paper and the volume made up to the mark with distilled water. The following elements; Ca, Mg, Fe, and Zn were determined using a Perkin-Elmer Atomic Absorption Spectrophotometre (AAS) model- A Analyst 200 with acetylene gas. The wavelengths of the elements were respectively 422.67, 285.21, 248.33, and 213.86 nm. Sodium (Na) and potassium (K) were determined through Flame Photometry analysis using JANEWAY PF P7.

2.2.3 Determination of vitamin C content

The amount of vitamins C in the sample was determined using the official method (967.21) of AOAC (2003). Fifty mililitres (50 ml) of each sample of *Zobo* drink was filtered into a clean 100 ml volumetric flask and 25 ml of the filtrate was pipetted into a 100 ml titration vessel with 1 ml of starch solution (0.5 % w/v) added as an indicator. The sample was titrated with iodine solution (0.005M) taken in a 100 ml graduated burette. The endpoint of the titration was identified as the first permanent trace of a dark blue-black colour due to the starch-iodine complex formation. The analysis was repeated thrice and the amount of vitamin C in the sample was calculated. Every 1 ml of 0.005M of iodine consumed is equivalent to 0.0008806 g of Vitamin C.

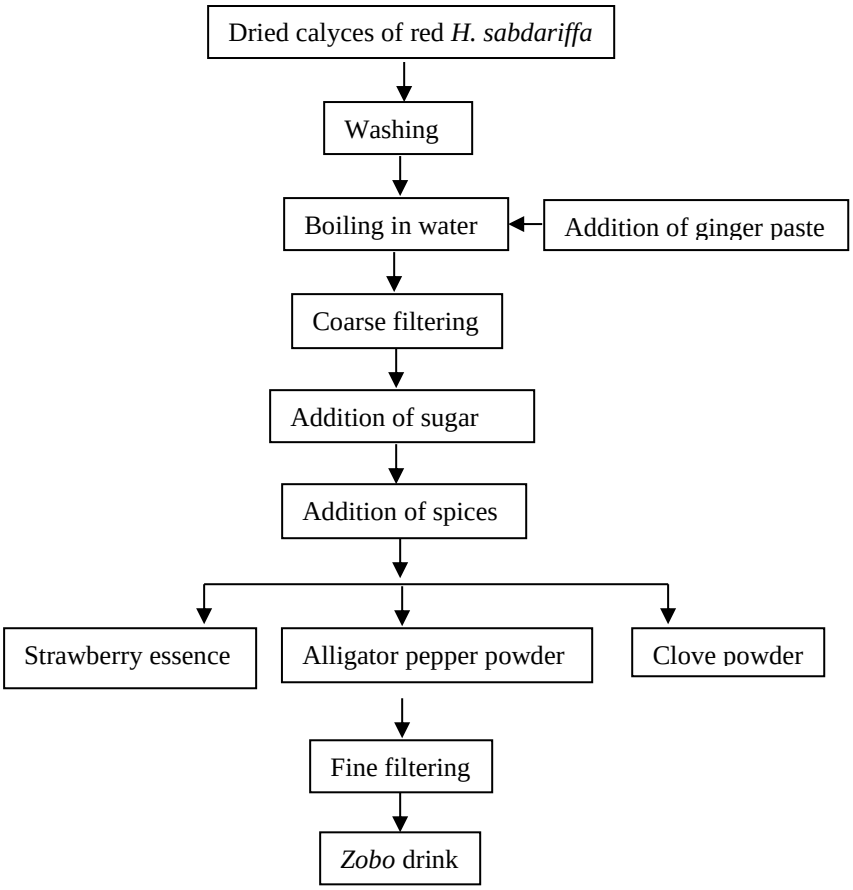

Figure. 1: Flow chart for the preparation of *Zobo* drink

2.2.4 Determination of β- carotene content

The β- carotene **(**Pro-vitamin A) was determined using the method adapted from (IVACG, 1992). The samples were mixed with volatile organic solvent (chloroform) and filtered. The absorbance of the filtrate was read with a UV- spectrophotometer at 328 nm.

2.2.5 Determination of pH

The pH of each drink was determined with a pH meter (JANEWAY Instrument, Model 3510, UK). Fifty millilitre (50 ml) of each of the *Zobo* drink samples was thoroughly mixed for 5 min before the bulb end of the pH meter was dipped for measurement. The pH meter was standardised with a buffer solution at pH 7 before being used to ascertain the sensitivity. The method of (Osundahunsi, 2003) was adopted with little modification in the quantity of the sample and the equipment used.

2.2.6 Determination of titratable acidity

The total titratable acidity (TTA) was determined by the official *method* (942.15) of AOAC (2000); 10 ml of the sample was thoroughly mixed for 5 min before being titrated with 0.1N NaOH using phenolphthalein solution as an indicator. The TTA was expressed as %lactic acid (w/v), 1ml 0.1N 0.009 g lactic acid.

2.2.7 Identification and quantitative analysis of tannins

Ten milli litres (10 ml) of bromine water was added to the 0.5 g aqueous extract; discolouration of bromine water showed the presence of tannins (Gul *et al*. 2017) which was further quantified by the titrimetric method according to the official methods 30.018; 30.019 of AOAC (1984) as adopted by Marin *et al*. (2009) with modification. The modification was that no extraction was necessary again since the *Zobo* drink samples were aqueous extracts forms of the materials used but only filtered through a Whatman No. 1 filter paper.

2.2.8 Determination of phytate

The method described by Bello (2013) was adopted with modification of no more extraction in the determination of phytate content. Fifty millilitres (50 ml) of the sample was diluted with 100 ml distilled water and filtered into a 250 ml beaker. Then, 10 ml of 0.3% ammonium thiocyanate solution was added as an indicator and titrated with standard iron (III) chloride solution which contained 0.00195 g iron per ml. The endpoint was observed to be yellow which persisted for 5 min and phytate content was calculated as %phytic acid (g/100 g).$\%\text{Phytic acid} = \frac{(\text{Titre value x } 0.0019 \text{ x } 100)}{2}$

All the analyses in sections (2.2.2 – 2.2.8) were performed at the Multidisciplinary Central Research Laboratory (MCRL), University of Ibadan, Ibadan, Oyo State, Nigeria.

2.2.9 Sensory evaluation

The scoring method was used to determine the preferential test on the samples of *Zobo* drink. Twenty semi-trained panelists were used for the evaluation based on familiarity and experience in drinking *Zobo*. The panelists were seated wide apart in a well-illuminated food product development laboratory of the Department of Food Science and Technology, Wesley University Ondo to analyse the drinks. The samples were coded before presenting to the panelists in disposable tasting cups and portable water to rinse mouth in between each tasting was provided. The 5-points hedonic scale was used in the rating with 5=like very much; 3=neither like nor dislike and 1=dislike very much. The evaluation was conducted on the sensory attributes of colour, consistency, aroma, taste, and overall acceptability according to the method described by Otolowo and Olapade (2018).

2.2.11 Statistical analysis

The data obtained were subjected to various statistical analyses which include simple descriptive mean, standard deviation, and analysis of variance (ANOVA) while Duncan's multiple range test (DMRT) was used to separate the means using SPSS 20.0 Software Inc. USA. Significance was taken at $p<0.05$.

3.0 Results and Discussion

3.1 Minerals and vitamins analyses of *Zobo* drink

The results of mineral and vitamin analyses were depicted in Table 2. The findings showed that the mineral contents in mg/L were in ranges of (165.88-173.28 calcium), (105.40-113.80 magnesium), (540.00-578.00 potassium), (41.68-48.16 sodium), (4.70-48.16 iron), and (7.14-13.34 zinc).

Table 2: Mineral and vitamin constituents of *Zobo* drinks

Sample	Calcium (mg/L)	Magnesium (mg/L)	Potassium (mg/L)	Sodium (mg/L)	Iron (mg/L)	Zinc (mg/L)	Na/K	Vitamin C (mg/100 g)	β-carotene (mg/100 g)
Zobo-S	173.28^{a}±0.00	105.40^{b}±0.00	522.00^{c}±0.00	41.68^{c}±0.00	4.75^{b}±0.00	7.14^{c}±0.00	0.080	0.29^{a}±0.01	0.09^{a}±0.00
Zobo-A	171.40^{b} ±0.00	113.80a±0.00	578.00^{a}±0.00	48.16^{a}±0.00	4.70^{c}±0.00	8.33^{b}±0.00	0.083	0.26^{b}±0.01	0.05^{c}±0.00
Zobo-C	165.88^{c} ±0.00	105.40^{b}±0.00	540.00^{b}±0.00	44.24^{b}±0.00	6.94^{a}±0.00	13.34^{a}±0.00	0.082	0.26^{b}±0.00	0.07^{b}±0.00
Standard (lenntech , 2016)	210-1300 mg/day	30-420 mg/day	400-4700 mg/day	1000-1500 mg/day	0.27-18 mg/day	08-14 mg/day	0.082 <1.0	0.26^{b}±0.00	0.07^{b}±0.00

Values are means of triplicate determinations±standard deviation; means with different superscripts along the same column are significantly ($p<0.05$) different

Key**:** *Zobo*-S = *Zobo* drink flavoured with commercial strawberry essence; *Zobo*-A = *Zobo* drink flavoured with alligator pepper; *Zobo*-C = *Zobo* drink flavoured with clove

The *Zobo* made with spices had most of the upper limits for the mineral elements and the obtained values were found to be adequate in respect of the RDA values stated by Ukwo *et al*. (2019).

Calcium is essential for the development and maintenance of strong bones and teeth and as a co-factor in some enzyme catalysis (Achi *et al*., 2017). Magnesium helps in maintaining normal nerve and muscle function, supports a healthy immune system, keeps the heartbeat steady, and also helps bones remain strong (Witkowski *et al*., 2011). Potassium (K) was observed to be the most abundant in the entire samples which agreed with the findings of Ukwo *et al*. (2019); K is important in maintaining a balanced body fluid with sodium to prevent cases of hypertension (Otolowo and Olapade, 2018) and it was highest (578 mg/l) in *Zobo*-A supporting the anti-hypertensive health benefit of Zobo made spices. Moreover, in the entire samples, the occurrence of sodium is in good ratios with potassium (Na/K <1) to maintain electrolytes' balance in the cells and the obtained range of values (0.080-0.083) further supports the anti-hypertensive potential of Z*obo* drink. Iron in the *Zobo* is essential for hemoglobin/blood formation and supports the immune system (Weiss, 2002). Zinc is a trace element important for body immunity; it is an antiviral mineral, critical for the development and function of immune cells, and thus, reported to be medicinal (Achi *et al*., 2017). Zinc was highest in *Zobo*-C (13.34 mg/L) followed by *Zobo*-A (8.33 mg/l) while *Zobo*-S had the least value (7.14 mg/l). The higher content of Zn in *Zobo* prepared with spices will play a better therapeutic role especially, in immune strengthening in correlation with the report of Olarinde (2022). The value obtained in *Zobo*-C for Zn (13.34 mg/L) is in consonance with the values reported by Nunes *et al*. (2017) for extracts of some Portugal herbs (*Geranium purpureum*-11.63 mg/kg and *Mentha suaveolens*- 16.36 mg/kg).

The occurrence of mineral contents showed the imparted improvement by the spices against the chemical-based strawberry essence and the obtained values are comparable to the report of Marin *et al*. (2009) for various Cerrado's fruits.

The vitamins C and β-carotene (precursor of vitamin A) contents of *Zobo* are also presented in Table 2. The range of values; 0.26-0.29 vitamins C and 0.05-0.09 mg/100 g β-carotene occurred within a comparable range among the samples and moderately adequate in the entire samples; the ranges are also comparable with the range of values reported by Marin *et al*. (2009) for the aqueous extract of *Ficus capensis* leaves (0.26 and 0.061 mg/100 g for vitamin C and vitamin A, respectively) observed to be medicinal. Both vitamins C and A, play important roles in human health. Vitamin C is a strong antioxidant that can strengthen the immune system for the body's natural defenses against free radicals (Kathleen, 2015) and facilitates the absorption of dietary iron from the intestine (Button, 2004). *Zobo* drink was documented to be rich in vitamin C (Adeniji, 2017) and the presence of ascorbic acid (vitamin C) was reported in different varieties of *Hibiscus sabdariffa* by Babalola *et al*. (2001). β-carotene is converted to vitamin A by the body hence, called a precursor of Vitamin A. Vitamin A is also essential for immune function, eye health, cell recognition, reproduction, and fetal development (Clagett-Dame and DeLuca, 2002). Therefore, *Zobo* drink with moderate amounts of vitamins C and A as observed in the present work will add to the nutrient intake and be a potent health drink for the development of adequate immunity.

With exception of calcium and iron, *Zobo* made with spices were higher in mineral contents than the conventional *Zobo* drink (*Zobo*-S) and were at a comparable range in the occurrence of the vitamins. Thus, *Zobo*-A and *Zobo*-C have good nutritive value in terms of suitable contents of the determined micronutrients with antioxidant properties that are necessary to maintain a balanced immune system in combating infections for a stable health condition (Adeooye *et al*., 2019). Consequently, *Zobo* prepared with natural spices as used in this study stands to be an additional source of micronutrients with better immune-strengthening potentials that will support healthy living than the conventional *Zobo*.

3.2 The pH, lactic acids (TTA), and phytochemical analyses of *Zobo* drinks

The chemical composition of *Zobo* drinks is presented in Table 3. The results indicated that the pH of the samples ranged from (2.47-2.56); samples *Zobo*-A and *Zobo*-C had the same value (2.56) which was significantly ($p<0.05$) higher than the value (2.47) obtained in *Zobo*-S, implying a decrease in the acidity of *Zobo*-A and *Zobo*-C which might be as a result of certain components in the spices. The obtained pH values were comparable though lower than the range of values (3.94-7.67; 4.46-5.56) reported by Adesokan *et al*. (2013) and Badejo *et al*. (2014) for *Zobo* made with ginger and galic; and Tigernut beverage blends (in which *Hibiscus sabdariffa* extract is one of the components), respectively. The disparities might probably be as a result of differences in the recipes and methods of preparation.

The total titratable acidity (TTA) in terms of the %lactic acid content of *Zobo*-S was significantly ($p<0.05$) higher (1.06 g/100 ml) than those of *Zobo*-A (0.93 g/100 ml) and *Zobo*-C (0.94 g/100 ml) which were not significantly different from each other. This correlates with the order in which the pH occurred in the samples; *Zobo*-S with a higher TTA had the lower pH value. The lower TTA in the *Zobo* made with spices implies a reduced 'acid taste' in accordance to the report of Badejo *et al*. (2014) with a comparable range of values (0.88-1.53 g/100 ml). However, the acidic nature of the drinks will be advantageous in preventing the thriving of some pathogenic bacteria in the gastrointestinal tract (Ayoade *et al*., 2012) without a negative impact on the host. This is because the drink is usually not taken on an empty stomach but normally taken during mealtime or with a light snack as noted also by Ukwo *et al*. (2019).

The phytochemicals of importance that were considered, majorly because of their biological and health-related activities include tannins and phytates (Olubunmi *et al*., 2019; Adeleke, and Babalola, 2020). The values obtained for tannins (ranged, 0.31-0.44 mg/100 g) and phytates (0.09 mg/100 g) were comparable with the safe values (0.49 and 1.10 mg/100 g, respectively) reported by Ekpa and Sani, 2018) for pawpaw. The addition of clove positively improved the tannin content in *Zobo*-C (0.44 mg/100 g) while no variation was observed for the phytate contents. The presence of phytochemicals including tannin and phenols were reported by Builder *et al*. (2010) in a formulated extract of *Hibiscus sabdariffa*. It was also reported that tannins have shown potential antiviral, antibacterial, and anti-parasitic effects; and certain tannins are able to inhibit HIV replication selectivity and are useful as a diuretic. Likewise, phytates (phytic acid) were reported potent in reducing blood glucose and possess health benefits to diabetic patients (Chukwuebuka and Chinenye, 2015).

Although, tannins and phytates (tannic acid; phytic acid) as found in plant materials when in excess of the safe level was said to be antinutritive or possess a chelating effect on some essential mineral elements such as Ca, Mg, Fe, and Zn; however, when occurred in moderate amount as in the present work, they are well known for their therapeutic properties rather than could impair the said minerals' absorption (Umaru *et al*., 2007). Moreover, the obtained values in the present work were below the level (4-9 mg/100 g) at which phytate could exhibit the chelating effect, though, the toxic level for tannin is said to be obscure (Ekpa and Sani, 2018).

In addition to antioxidants activity, phytochemicals in human diets help stimulate the immune system and are potent against some viruses and bacteria *In-vitro*; phenolic compounds were reported to have antiviral activity against several viruses (Chukwuebuka and Chinenye, 2015). Specifically, as revealed by the present study, the contents of the minerals, vitamins, and phytochemicals compounds of the water-extracts of red *Zobo* calyxes' must have been improved by those present in the added spices and stands a good contributor to the overall health-claim benefits of *Zobo* which corroborates the reports of Juliani *et al*.2009 and Zhen *et al*. 2016. It was also reported that the dietary constituents contributing to the protective effects of plant materials (as used in the preparation of *Zobo* drinks) are plant secondary metabolites in the form of phytochemicals, minerals, and vitamins (Okereke *et al*., 2015).

Thus, pleasurable, moderate, and regular consumption of hygienically prepared *Zobo* flavoured with natural spices alongside other healthy diets will provide strong immunity the body needs to fight an outbreak of infection and diseases as recently witnessed by the COVID-19 pandemic. This is supported by resent research findings that identified Alligator pepper (one of the spices used in favouring *Zobo* in the present work) as a treatment option for COVID-19 (Olarinde, 2022). Hence, the *H. sabdariffa* and the natural spices used are economically important for adequate maintenance of proper metabolic processes. This is in consonance with the observation made by Okereke *et al*. (2015), and the call made by Akinwande (2021) to the government of Nigeria to engage in the cultivation of spices and other neglected crops for their sustainability and as a panacea to ravaging food insecurity in the nation. Additionally, in this era of the COVID-19 pandemic, building a strong or balanced immune system through diet will be a natural defensive mechanism to survive the infection especially with foods and drink prepared with natural spices. This is in consonance with the report of Olarinde (2022) and probably may be unconnected with some COVID-19 infection victims in Nigeria that survived the infection through the trial-immune-strengthening treatment while most of the death toll were said to be victims of underlining sicknesses that must have deeply impaired the immune system of the deceased (PTF, 2020).

Table 3: The pH, lactic acid (TTA), and phytochemical contents of *Zobo* drinks

Samples	pH	TTA (g/100 ml)	Tannin (mg/100 g)	Phytate (mg/100 g)
Zobo-S	2.47^{b}±0.01	1.06^{a}±0.02	0.34^{b}±0.02	0.09^{a}±0.01
Zobo-A	2.56^{a}±0.01	0.93^{b}±0.01	0.31^{c}±0.02	0.09^{a}±0.01
Zobo-C	2.56^{a}±0.01	0.94^{b}±0.01	0.44^{a}±0.03	0.09^{a}±0.01

Values are means of triplicate determinations ± standard deviation, means with different superscripts along the same column are significantly different. Significant taken at $p<0.05$

Key: TTA = Total titratable acidity; *Zobo*-S = *Zobo* drink flavoured with commercial strawberry essence; *Zobo*-A = *Zobo* drink flavoured with alligator pepper; *Zobo*-C = *Zobo* drink flavoured with clove.

3.4 Sensory evaluation of *Zobo* drinks

The mean scores for the sensory attributes of *Zobo* drinks are presented in Table 4. The results indicated that the drinks were well accepted which is in line with the reports of Ogiehor and Nwafor (2004) and Oboh *et al*. (2011). However, in the present work significant ($p<0.05$) differences exist in the sensory attributes between the samples with *Zobo*-A being the most preferred in colour (4.26), taste (3.80), and overall-acceptability (4.26) while *Zobo*-C had the best consistency (4.11) and aroma (4.21) with the five-point hedonic scale used. This implies that natural spices produced a more acceptable sensory quality in the drinks than the chemical-based strawberry essence in *Zobo*-S. This is in agreement with the previous findings that spices have a desirable influence on the overall organoleptic quality when added to food (FAO, 2005; Ogiehor *et al*., 2008; Adesokan *et al*., 2013). The findings in the present study also conform to that of Oluwalana *et al*. (2013) where a significant difference was discovered on the sensory properties of sorghum sheath (*Sorghum bicolor*; "poporo") beverage treated with *Aframomum meleguata*. Accordingly, the addition of natural spices improved the general sensory acceptability of the *Zobo* drink.

Table 4: Sensory evaluation of *Zobo* drinks

Samples	Colour	Consistency	Aroma	Taste	Overall acceptability
Zobo-S	4.25^{a}±0.01	3.96^{b}±0.01	4.06^{a}±0.01	2.96^{c}±0.01	3.51^{c}±0.01
Zobo-A	4.26^{a}±0.01	3.95^{b}±0.01	3.93^{a}±1.15	3.80^{a}±0.01	4.26^{a}±0.01
Zobo-C	3.66^{b}±0.01	4.11^{a}±0.01	4.21^{a}±0.01	3.41^{b}±0.01	3.96^{b}±0.01

Values are means±standard deviation, means with different superscripts along the same column are significantly different. Significant was taken at $p<0.05$

Key**:** *Zobo*-S = *Zobo* drink flavoured with commercial strawberry essence; *Zobo*-A = *Zobo* drink flavoured with alligator pepper; *Zobo*-C = *Zobo* drink flavoured with clove

4.0 Conclusion

The findings of this study showed that natural spices used could strengthen the acclaimed health benefits of *Zobo* better than the commercial flavour essence. Thus, the use of natural spices at both household and commercial preparation levels should be encouraged among the local producers of *Zobo* drinks. If proper packaging and shelf-life study is carried out, industrial-scale production will also be a possibility which, in turn, will improve the cultivation of *Hibiscus sabdariffa* and the spice crops for economic sustainability. Furthermore, culinary use of natural spices for healthy diets should be encouraged through government advocacies for natural body defense that will lessen the expensive treatment measures against a sudden outbreak of infections and diseases as recently witnessed by the COVID-19 pandemic. Nevertheless, in the light of the present global COVID-19 pandemic, the advisories by all the appropriate health authorities should still be strictly observed.

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