

SELECTED FOOD ADDITIVES COMPOSITION IN BEVERAGES COMMONLY CONSUMED IN AFIKPO, SOUTH EAST, NIGERIA

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ABSTRACT

A rapid and simple UV spectrophotometric procedure were applied for the evaluation of levels of the composition of some food additives (aspartame, saccharin, potassium sorbate and sodium benzoate) in twenty (20) different beverages commercially available in south eastern part of Nigeria. The determinations were undertaken primarily to assess the compliance of composition levels of the investigated food additives with the World Health Organization (WHO) permissible levels. The results obtained from this study indicated that the composition levels of most of the studied food additives were within the maximum permissible levels. The findings of this study suggest that moderate consumption of these beverages is less likely to pose health risk associated with consumption of these additives in consumers.

Keywords

UV Spectrophotometric; Food Additives; Beverage

INTRODUCTION

Food additives are used to improve the look, textures and taste of every day foods especially beverages. Natural Sweeteners are generally sweet tasting compounds with some nutritional value consisting of either monosaccharides or disaccharides (Lau *et al.*, 2003; Tfouni, 2002). Artificial sweeteners are compounds that improve taste of foods or drinks but have little or no nutritional value (Butchko *et al.*, 2002).

Artificial Sweeteners are mostly employed in food, drinks, and pharmaceutical products being used to control calorie intake and in certain medical conditions such as diabetes and hyperglycaemia (Krebs *et al.*, 2000). The most common artificial Sweeteners are aspartame, sodium cyclamate, acesulfame-k and saccharin, which are marketed in about 90 countries of the world (David *et al.*, 2008). Artificial Sweeteners are also called low-calorie Sweeteners, sugar substitutes or non-nutritive Sweeteners (David *et al.*, 2008). They can be used to sweeten food and drinks for less calories and carbohydrate when they replace sugar (Ranney *et al.*, 2007). These Sweeteners are used by food companies to make diet drinks, baked goods, frozen desserts, candy, light yogurt, and chewing gum (Yagasakiet *al.*, 2008).

Preservatives are additives that inhibit the growth and activities of the microorganisms and help to increase shelf life of beverages for a longer time without affecting its natural characteristics (Kubo and Lee, 2008). The benzoic acid, acetic acid and sorbic acid together with propionate and sulphur dioxide are acid preservatives mostly employed in large scale in foods and soft drinks preservation (Tulamaitet *et al.*, 2005). Sorbic acid and the sorbates are the preservatives mostly used in the soft drinks industry (Pylypiw and Grether, 2000). In fruit beverages preservation, the sodium salt of the benzoic acid is frequently used, as it is more soluble than the free acid (Krebs *et al.*, 2000).

Preservatives can be used alone or together with other methods of food preservation (Tulamaitet *et al.*, 2005). Preservative may be antimicrobial preservative, which inhibit the growth of bacteria or fungi, including mold or they can be antioxidants such as oxygen absorbers, which inhibit the oxidation of food constituents (Wen *et al.*, 2007). Nonetheless, food additives have been the subject of interest among consumers, health professionals; commercial and industrial agencies alike because they are widely consumed in the diet by most segments of the population and can exert adverse health effects, especially for children and pregnant women. In fact, such food additives are subject to regulation, since excessive or inappropriate use may present food safety problems and can introduce a risk factor. For instance, a small group of people who have the hereditary disease phenylketonuria are sensitive to phenylalanine, one of the metabolites of aspartame artificial sweetener thus; all products containing aspartame must be labeled in many countries. In addition, it is well established that excessive intake and consumption of caffeine may cause many undesirable side effects. However, excessive use of some of these chemicals in food materials may cause toxicity in humans (Wakabayashi *et al.*, 1990). Some chemical food preservatives have been found to provoke urticaria, angioedema and asthma (Smith, 1999). In addition these chemical preservatives have been linked with cancer both in animal and human studies (Hoover and Milich, 1994). They have been associated with many symptoms in susceptible individuals, such as severe chest and facial pressure or overall burning sensation (Collins, 1996). The amount of these additives to be used in food and beverages are often regulated by relevant bodies. Whether beverage and food industries adhere to the permissible limits is another issue. There is therefore need for information on the levels these additives in the beverages commonly consumed as most industries tend to be profit driven rather than being concerned about the effect of excessive amount of these additives. This study set out to investigate the presence and concentration of additives in selected beverages commercially available for consumption southeast Nigeria with a view to ascertain the levels of compliance with the WHO permissible limits.

MATERIALS AND METHODS

Sample Collection and Preparation

The twenty (20) beverage samples investigated in this study were categorized as: soft drinks, yoghurt/milk drinks, juice drinks, and sachet drinks commonly consumed in the south eastern part of Nigeria. The randomly purchased beverage samples were subjected to mild clean-up procedures such as filtration and degassing. Once sample bottles were open the drinks were degassed, homogenized and filtered. Quantitative analysis of this samples were done using standard spectrophotometric methods (Alghamdi *et al.*, 2005; Lau *et al.*, 1998)

DETERMINATION OF SACCHARIN

Two milliliters (2 ml) of HCl was added to 50 ml of sample in separating funnel, mixed and extracted with 50 ml of diethyl ether. The ether extract was filtered into a clean 250 ml conical flask and the solvent evaporated. Six milliliters (6 ml) of HCl and 5mL distilled water were added and evaporated on a hot water bath to about 1ml. Six milliliters (6 ml) of HCl and 5 ml distilled water were added again and evaporated to 1 ml. The solution diluted to 50 ml with distilled water. Two milliliters (2 ml) of this solution and 1mL of Nessler's reagent were added in a 25 ml volumetric flask and made up to mark with distilled water. Two milliliters (2ml) of each solution of drink samples were placed in the spectrophotometric cell after adequate dilution with distilled water and the absorbance was read at 425 nm.

DETERMINATION OF ASPARTAME

Twenty milliliter (20 ml) of beverage sample and 50mL of solvent mixture were added into a 100ml volumetric flask and shake for 30 minutes on a flask shaker and made up to mark with solvent mixture. The solution was filtered through Whatman No.1 filter paper discarding the first 20 ml filtrate and the filtrate collected in a stoppered flask. Two milliliter (2 ml) of each solution of drink samples were placed in the spectrophotometric cell after adequate dilution with distilled water and the absorbance was read at 258 nm.

DETERMINATION OF POTASSIUM SORBATE

Fifty milliliter (50 ml) of oxidant solution, 10 ml) distilled water and 3 ml beverage samples were added into a 200 ml volumetric flask. Fifty milliliters (50 ml) of oxidant solution was added again and brought to mark with distilled water. After 10-15 minutes, 2 mL of each solution of drink samples were placed in the spectrophotometric cell after adequate dilution with water and the absorbance was read at 253 nm.

DETERMINATION OF SODIUM BENZOATE

Twenty-five milliliters (25 ml) of beverage samples were poured into a 50 ml beaker and degassed by magnetically stirring until effervescence ceased. Ten milliliters (10 ml) of the degassed beverage samples, 20 ml of oxidant solution and 5mL of distilled water were added into a 200 ml volumetric flask and made up to mark with distilled water. The solution filtered with Whatman N0.1 filter paper. Two milliliters (2 ml) of each solution of drink samples were placed in the spectrophotometric cell after adequate dilution with distilled water and the absorbance was read at 425 nm.

DATA COLLECTION AND ANALYSIS

In order to conduct the necessary statistical evaluations, all quantitative measurements were repeated three times. The results were expressed as mean \pm standard deviation and difference between means obtained by one way analysis of variance ANOVA.

RESULTS

Table 1 shows the result obtained for the mean values of food additives compositions in canned soft drink (CSD) samples. For the sweeteners aspartame and saccharin the concentration of aspartame is significantly higher ($p < 0.05$) than that of saccharin in all the samples except in CSD-C while the concentration of preservatives potassium sorbate is significantly higher ($p < 0.005$) than that of sodium benzoate. CSD-E shows a significant ($p < 0.005$) high potassium sorbate concentration compared with

other canned drinks tested. CSD-B showed a significant ($p < 0.005$) increase in aspartame compared with other tested samples while CSD-C showed a significant ($p < 0.005$) increase in saccharin and sodium benzoate concentration compared with other samples (Table 1).

Table 1: Results for mean values of food additives compositions in Canned Soft Drink (CSD)samples.

BEVERAGES	CONCENTRATION(mg/L)			
	ASPARTAME	P.SORBATE	SACCHARIN	S.BENZOATE
CSD-A	14.55 ± 0.25	27.17±0.14	4.56±0.16	12.67±0.54
CSD-B	26.97 ±0.24	25.72±0.08	0.89±0.07	11.33±0.54
CSD-C	6.26± 0.14	21.61±0.21	9.06±0.34	14.78±0.42
CSD-D	12.72±0.43	26.33±0.14	0.39±0.08	10.89±0.69
CSD-E	22.42± 0.25	28.67±0.14	1.33±0.22	12.00±0.55

Result expressed as mean ± SD of triplicate determinations. ND= Not Detected

Table 2 shows the result obtained for the mean values of food additives compositions Yoghurt/milk drinks (YMD) samples. For the sweetener’s aspartame and saccharin, the concentration of aspartame is significantly higher ($p < 0.05$) than that of saccharin in all the samples while the concentration of preservatives potassium sorbate showed non significantly ($p > 0.05$) increase in all the samples tested compared with the concentration of sodium benzoate. YMD-C showed a significant ($p < 0.005$) increase in potassium sorbate and saccharin concentration compared with other tested milk samples, YMD-B showed a significant ($p < 0.005$) increase in aspartame concentration compared with other tested milk samples while sodium benzoate was observed to be significantly ($p < 0.005$) higher in YMD-E compared with other tested samples (Table 2).

Table 2: Results for the mean values of food additives compositions in Yoghurt MilkDrinks (YMD) samples.

BEVERAGES	CONCENTRATION (mg/L)			
	ASPARTAME	P.SORBATE	SACCHARIN	S.BENZOATE
YMD-A	147.81±0.28	24.78± 0.16	3.00± 0.36	12.78±0.69
YMD-B	219.49±0.62	25.83± 0.27	1.11±0.78	17.78±0.42
YMD-C	39.29±0.51	28.94±0.21	6.83±0.14	39.22±0.42
YMD-D	ND	26.89±0.08	1.39±0.08	12.78±0.42
YMD-E	36.26±0.29	24.00±0.27	2.78±0.08	45.44±0.42

Result expressed as mean ± SD of triplicate determinations. ND= Not Detected

Table 3 shows the result obtained for the mean values of food additives compositions insachet drinks in powdered (SDP) formsamples. For the sweeteners aspartame and saccharin there was no significant difference ($p > 0.05$) in most of the samples while the concentration of preservatives potassium sorbate is significantly higher ($p < 0.005$) than that of sodium benzoate.

Table 3: Result for mean values of food additives composition in Sachet drinks in Powdered form (SDP)samples.

BEVERAGES	CONCENTRATION (mg/L)			
	ASPARTAME	P. SORBATE	SACCHARIN	S.BENZOATE
SDP-A	12.02±0.14	27.28±0.08	6.72± 0.08	11.45±0.32
SDP-B	10.20±0.38	24.00±0.27	17.25±0.24	10.78±0.32
SDP-C	2.63±0.15	ND	3.45±3.46	12.78±0.32
SDP-D	33.3±0.25	25.5±0.08	17.37±0.08	12.11±0.57
SDP-E	3.83±0.38	25.17±0.13	6.72±0.21	3.78±0.32

Result expressed as mean ± SD of triplicate determinations. ND= Not Detected

Table 4 shows the result obtained for the mean values of food additives compositions in juicedrinks in paper bags (JDPB) samples. For the sweeteners aspartame and saccharin there was no significant difference ($p>0.05$) in most of the samples except in JDPB-B where it is significantly higher ($p<0.05$) than the maximum permissible levels. The concentration of preservatives potassium sorbate is significantly higher ($p<0.005$) than that of sodium benzoate except in JDPB-E.

Table 4: Results for mean values of food additives composition in Juice Drinks in Paper Bags (JDPB) samples.

BEVERAGES	CONCENTRATION (mg/L)			
	ASPARTAME	P.SORBATE	SACCHARIN	S.BENZOATE
JDPB-A	6.67±0.25	135.95±0.16	11.44±0.21	14.44±0.42
JDPB-B	ND	26.78±1.15	124.67±0.24	13.56±0.42
JDPB-C	7.27±0.25	27.68±0.12	8.67±0.14	11.78±0.42
JDPB-D	10.1±0.14	23.95±0.48	11.5±0.14	12.56±0.42
JDPB-E	4.04±0.14	4.72±0.34	1.29±0.08	11.89±1.29

Result expressed as mean ± SD of triplicate determinations. ND= Not Detected

Table 5. Permissible limits of the additives in the different classes of beverages (Source; FAO, 2016)

Additives	Permissible Levels	
	Carbonated Drinks	Non-carbonated Drinks
P. Sorbate	1000mg/kg	500mg/kg
Aspartame	600mg/kg	600mg/kg
Benzoate	250mg/kg	250mg/kg
Saccharin	300mg/kg	300mg/kg

DISCUSSION

The use of additives in food industry has been increased with the advancement in the production technologies. These chemicals are principally used for the protection of the foods from microorganisms. Some of these chemicals are used as additives to improve the texture, taste, favor and color of the foods and beverages. However, excessive use of some of these chemicals in food materials may cause toxicity in humans (Wakabayashi *et al.*, 1990).

This study analyzed the levels of selected food additives composition of twenty beverages commonly consumed in south-eastern part of Nigeria. The result in Table 1 shows aspartame to be highest in CSD-B but lowest in CSD-C which has the highest concentration of saccharin whereas CSD-B has the lowest concentration of saccharin. For the preservatives, the concentration of potassium sorbate appears to be generally higher than the concentration of sodium benzoate. CSD-E and CSD-C have the highest concentration of potassium sorbate and sodium benzoate respectively whereas CSD-C and CSD-D have the lowest concentration of potassium sorbate and sodium benzoate respectively. Reaction of sorbates is rare, but have included reported of urticaria and contact dermatitis (Kinderterer and Hatton, 1990). Ahmad *et al.* (2005) reported the composition levels of potassium sorbate in several soft drinks ranged from 20.4 ± 0.1 mg/l to 29.13 ± 0.03 mg/l in Riyandh City. Prodolliet and Bruelhart, (2003) has also reported the maximum level of potassium sobate in soft drinks is 30.11 ± 0.13 mg/l. The composition levels recorded here are in agreement with earlier reports of food additives in beverages (Ahmad *et al.*, 2005; Daniel *et al.*, 2008). The World Health Organization has set the maximum permitted level of potassium sorbate in beverages to be 300 mg/l. All the analyzed beverage samples are far below this recommend level by the World Health Organization (WHO). According to Sabah and Scriba, (2008), the compositions of aspartame in beverages were mostly higher than other food additives. Weihranch and Diehl, (2004) reported that the high composition of aspartame in beverage is due to its ability to extend flavor making them seem sweeter and given them a more full-bodied taste.

The results for the mean values of additives compositions in yoghurt/milk drink are shown in Table 2. The concentration of aspartame appears to be significantly ($p < 0.05$) higher compared with the concentration of saccharin in the yoghurt/milk drink. YMD-A and YMD-C have the highest concentration of aspartame and saccharin respectively whereas vita milk and YMD-B have the lowest concentration of aspartame and saccharin respectively. However, aspartame was not detected in YMD-D. For the preservatives, the concentration of potassium sorbate appears to be generally higher than the concentration of sodium benzoate. YMD-C and YMD-E have the highest concentration of potassium sorbate and sodium benzoate respectively whereas YMD-E has the lowest concentration of potassium sorbate; YMD-A and YMD-D have the lowest concentration of sodium benzoate. Benzoates have been used mainly in marinated fish, fruit-base fillings, jam, salad, cream, soft drinks and beer; have been reported to provoke urticaria, angioedema and asthma (Gustafsson *et al.*, 2003).

The results for the mean values of food additives composition in sachet drinks available in powdered form are shown in Table 3. The result shown that for the sweeteners, SDP-D has the highest concentration of both aspartame and saccharin whereas SDP-C has the lowest concentration of both aspartame and saccharin. For the preservatives, SDP-A and SDP-C have the highest concentration of potassium sorbate and sodium benzoate respectively whereas SDP-C and SDP-E have the lowest concentration of potassium sorbate and sodium benzoate respectively. However, potassium sorbate was not detected in SDP-C.

The result for the mean values of food additives composition in juice drinks in paper bags are shown in Table 4. The concentration of saccharin appears to be generally higher than the concentration of aspartame. JDPB-D and JDPB-B have the highest concentration of aspartame and saccharin respectively whereas JDPB-B and JDPB-C have the lowest concentration of aspartame and saccharin respectively. However, aspartame was not detected in JDPB-B. Maja and Zorka (2011) reported that, the concentration of saccharin in various beverages ranged from 35.11 mg/l to 134.59 mg/l. According to the US Food and Drug Administration (FDA), the maximum permissible level of saccharin in beverages is 80 mg/l (Maja and Zorka, 2011). The concentration of saccharin in JDPB-B found to be higher than the WHO permissible levels is however worrisome for the preservatives, the concentration of potassium sorbate appears to be generally higher than the concentration of sodium benzoate. JDPB-A has the highest concentration of both potassium sorbate and sodium benzoate whereas JDPB-D and JDPB-C have the lowest concentration of potassium sorbate and sodium benzoate.

Most of the analyzed beverage samples except JDPB-B juice showed significant ($p < 0.005$) decrease when compared to the recommend levels by the World Health Organization (WHO).

CONCLUSION

The use of preservatives in food industry has been increased with the advancement in the production technologies. However, excessive use of some of these chemicals in food materials may cause toxicity in humans. The findings of this study suggest that, the concentrations of these additives in the various beverages analyzed are mostly within regulatory bodies' permissible levels and hence consumption of these beverages are not likely to pose health risk associated with moderate consumption of these additives in consumers.

REFERENCES

- Ahmad, H. A., Alghamdi, A. F. and Ahadulrahman, A.A. (2005). Determination of Content Levels of some Food Additives in Beverages Consumed in Riyadh City. *Food Chem Control* **13(2)**, 117 - 123.
- Alghamdi, A. H. Alghamdi A. F and Alwarthan A. A. (2005). Determination of Content Levels of Some Food Additives in Beverages Consumed in Riyadh City. *J. King Saud Univ. Science*, **18(2)**, 99 – 109.
- Butchko, H., Stargel, W.W. and Comer, C.P. (2002). Aspartame; Review of Safety. *Regulatory Toxicology and Pharmacology* **32(2)**, 51 - 93.
- Collins, W. C. (1996). Intolerance of Food Additives. *Curr. Allergy Asthma Rep.* **51**, 315 – 316.
- Daniel, M. (2008). Reactions to Food Additives and Preservatives. *Food Australia*, **45 (8)**, 374-380.
- David, J. A, David, P. P, Sott, A.H, Alan, R.K, Indra, P and Walters, D.E. (2008). Commercial, Synthetic Non Nutritive Sweeteners, *Angewandte Chemie International Edition*, **37 (13)**, 1802-1817.
- Guastafsson, E., Edlund, M. and Hagberg, M. (2003). Effect of Food Additives. *Health Beliefs*. **34 (5)**, 565 – 570.
- Hoover, D., Milich, R. (1994). Food Additive. *Journal of Abnormal Child Psychology*. **22 (15)**, 501 - 515
- Kinderlerer, J. L., Hatton, P. (1990). Fungal metabolites of sorbic acid. *Food Addit. Contam.* **7 (5)**, 657 – 669.

- Krebs, H.A, Wiggins, D, Stubbs, M, Sols, A. and Bedoya, F. (2000). Studies on the Mechanism of the Antifungal Action of Benzoate. *Biochem. J.* **214(3)**, 651-563.
- Kubo, I. and Lee, S. H. (2008). Potentiation of Antifungal activity of sorbic acid. *J. Agric Food chem.* **46**, 4052-4055.
- Joint FAO/WHO Codex Alimentarius commission. (2016). General standard for Food Additives. Rome: World Health Organization: Food and Agriculture Organization of the United Nations
- Lau, O. W. Luke, S. F. and Chan, W. M. (1988). Spectrophotometric Determination of Aspartame in soft Drinks with Ninhydrin as Reagent. *Analyst* **113**, 765-768
- Maja, S. and Zorka, K. (2011). Determination of Artificial Sweeteners in beverages and special Nutritional products using High performance Liquid chromatography *Arch Hig Rada Toksikol* **62**, 169 - 173.
- Newsome, R. In: Altschul, A.M. (Ed.), *Low-Calorie Foods Handbook*. New York: Marcel Dekker,1993, p. 139.
- Prodoliet, J. and Bruehlhart, M. (2003). Determination of Aspartame and its Major Decomposition Products in Foods. *J. AOAC Int.* **76(2)**, 275-282.
- Pylypiw, H.M. and Grether, M.T. (2000). Rapid high-Performance Liquid Chromatography Method for the Analysis of Sodium benzoate and Potassium Sorbate in Foods. *J. Chromatogr. A* **883(2)**, 299 - 304.
- Ranney, R.E, Opermanu, J.A, Muldoon, E. and McMahon, F.G. (2007). Comparative Metabolism of Aspartame in Experimental Animals and Humans. *J. Toxicol Environ Health* **2(2)**, 441 - 451.
- Sabah, S. and Scriba, G. K. E. (2008). Determination of Aspartame and its Degradation by Capillary Electrophoresis *J. Pharma. Biomed. Anal* **16**, 1089-1096.
- Smith, J. M. (1999). Adverse reaction to food and drug additives. *EuropeanJournalClin. Nutri.* **45** (20), 17 – 21.
- Tfouni, S. A. V. and Toledo, M. C. F. (2002). Determination of benzoic and sorbic Acids in Brazilia Food. *Food control* **13**, 117 - 123.
- Tulamait, A, Laghi, E, Mikrut, K, Carey, R. B. and Budinger, G. R (2005). Potassium Sorbate reduces gastric colonization in patients receiving mechanical vitalization *J.Crit care* **20(3)**, 281 - 287.
- Wakabayashi, K. (1990). Identification of food mutagens, In: Mendeisohn ML. Albertini R.J (eds.), In Mutation and the Environment. Part E: Environmental Genotoxicity, risk and modulation. Wiley-Liss, New York. Pp. 107-116.
- Wehranch, M. F. and Diehl, V. (2004). Artificial Sweeteners-do they bear a carcinogenic risk? *Annals of Oncology* **15(10)**, 1460 - 1465.
- Wen, Y., Wang, Y. and Feng, Y.Q. (2007). A simply and rapid method for simultaneous determination of benzoic and sorbic acids in Food using in-tube Solid phase micro exaction coupled with high-performance liquid Chromatography. *Anal Bioanal Chem.* **388(8)**, 1779 - 1787.
- Yagasaki, M, and Hashimoto, S. (2008). Synthesis and application of dipeptides, current status and perspectives. *Applied Microbiology and Biotechnology* **81(1)**, 13 - 22.