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RESEARCH ARTICLE

Studies on Physicochemical Properties, Trace Mineral and Heavy Metal Contents of Common Energy Drinks**Casimir Emmanuel Gimba, Stephen Eyije Abechi, and Nurudeen Sulaiman Abbas***

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Corresponding Author*Nurudeen Sulaiman Abbas****Abstract**

Ten brands of energy drinks were analyzed for their physicochemical properties (pH, turbidity, conductivity and total dissolved solids), essential and heavy metals contents. Results showed that the pH ranged from 4.47 ± 0.012 - 5.27 ± 0.017 . Energy drinks analyzed all fell within the FDA recommended pH range for caffeinated drinks and coffee. Turbidity ranged from 53 ± 1.732 - 592 ± 1.155 NTU. Conductivity ranged from 497 ± 0.006 - 1975 ± 1.732 S/ μ cm and total dissolved solids ranged from 243 ± 0.577 - 940 ± 0.000 mg/L. Iron, calcium, zinc and potassium were found in all the energy drinks. Cadmium was not detected in all energy drinks samples. The copper concentration of energy drinks ranged from 0.002 ± 0.0002 - 0.070 ± 0.0006 mg/L. All sampled energy drinks had concentrations below the maximum contaminant level (MCL) of copper (1.0 mg/L). Lead concentration of energy drinks ranged from 0.028 ± 0.0006 - 0.139 ± 0.0004 mg/L. These exceeded the maximum contaminant limit of lead (0.01 mg/L). Manganese was detected only in sample PH and has a concentration of 0.003 ± 0.0001 mg/L which is below the maximum contaminant level (MCL) of 0.05 mg/L.

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Introduction

Energy drinks refer to beverages that contain caffeine in combination with other presumed energy-enhancing ingredients such as taurine, herbal extracts, and B vitamins. In recent years the consumption of readily available energy drinks has increased significantly with young adults forming the largest part of the consumers. History of energy dates back to 1987 when Red Bull was introduced in Austria, it became more popular in the 1990s following its introduction to the United States. Since then the sale of this drink has increased exponentially. In 2006, the energy drink market grew by 80% (Foran *et al.*, 2011). This is because manufactures claim the drinks can boost energy levels as well as physical endurance, improve concentration and reaction speed (Van den Eynde *et al.*, 2008). The popularity of energy drinks and the growth in their consumption among adolescents and young adults have brought worries regarding general health and well being of these consumers. Adolescents and young adults are often uninformed about the content of energy drink (Rath, 2012). Although many energy drinks are promoted as being nutraceutical foods, boosting health, energy, or otherwise having sought-after benefits, there is some concern among health professionals that these beverages, and the drinking behaviors of the targeted consumers, may in fact have adverse health consequences. The most commonly reported adverse effects include insomnia, nervousness, headache, and tachycardia (Clauson *et al.*, 2008). In a recent study, heavy consumption of energy drinks was attributed to new onset seizures in four patients (Iyadurai and Chung, 2007) and hospitalization of individuals with pre-existing mental illness (Chelben *et al.*, 2008). The pH, turbidity, total dissolved solid and conductivity may be associated with the overall quality of the energy drinks. They may also point to some adverse effect of energy drinks. Monitoring pH levels in beverages for instance is important because unacceptable pH level may lead to tooth

decay and weakening of immune system. The presence of heavy metals in beverages is also widely speculated. Lead toxicity causes many sign and symptoms such as abdominal pains, anaemia, anoxia, bone pain, brain damage, convulsion, dizziness, inability to concentrate Satarug and Moore, (2004). Excessive exposure to lead may also cause microcytic anemia, glycosuria, cognitive dysfunction, anorexia, metallic taste, insomnia, reticulocytosis. Target organs include the brain, bone, blood, kidneys, and thyroid gland (Satarug and Moore, 2004). Copper is needed for proteins involved in growth, nerve function and energy release (Institute of Medicine, 2001). It is vital for the formation of some important proteins. It is a critical functional component of a number of essential enzymes, known as cuproenzymes. Two copper-containing enzymes, ceruloplasmin (ferroxidase I) and ferroxidase II are involved in iron metabolism (Attieh, 1999). Copper is stored in appreciable amounts in the liver. It also has anti-oxidant properties and involved in the regulation of gene expression. The deficiency of Copper is manifested by impaired hematopoiesis, bone metabolism, disorders of digestive, cardiovascular and nervous systems (Krizek *et al.*, 1997). Manganese is both an important element and a potent neurotoxin. It accumulates in mitochondria, a major source of oxide, which can oxidize Mn^{2+} to the powerful oxidizing agent Mn^{3+} . Oxidation of important cells by Mn^{3+} has been suggested as a source of the toxic effects of manganese (Gunter *et al.*, 2004). Manganese is an actual component of manganese super oxide dismutase enzyme. It is a powerful antioxidant that searches the free radicals in human body and manages to neutralize these damaging particles and prevent any potential danger they may cause. Zinc is such a critical element in human health of which, a small deficiency is a disaster. Zinc deficiency is characterized by growth retardation, loss of appetite and impaired immune function. In more severe cases, zinc deficiency causes hair loss, diarrhea, delayed sexual maturation, impotence, hypogonadism in males, and eye and skin lesions (Ryan-Harshman and Aldoori, 2005). Iron fortification in foods has been increased to tackle the increased incidence of iron deficiency anemia, especially in the western countries (Kusumi *et al.*, (2006). Iron deficiency anemia is the most common micronutrient deficiency affecting mostly the low socioeconomic populations of developing countries (WHO). Excessive iron intake has been associated with overall increased risk of colorectal cancer (Senesse *et al.*, 2004). Only a handful of reports are available on physicochemical properties and trace metals contents of energy drinks. This study is aimed to evaluate the physicochemical properties, trace and heavy metals of energy drinks.

MATERIALS AND METHODS

Ten (10) brands of energy drinks samples were randomly purchased from the market and evaluated.

Determination of pH: The energy drinks pH was determined using a digital pH meter (JENWAY 3505).

Determination of conductivity: The electrical conductivity measurement of the energy drinks was determined using a digital TDS/conductivity meter (HACH) Sension 5.

Determination of Turbidity: The turbidity measurement of the energy drinks was determined using a digital turbidity meter (HACH DR/890 Colorimeter).

Total Dissolved Solid: The total dissolved solids measurement of the energy drinks was determined using a digital TDS/conductivity meter (HACH) Sension 5.

Acid digestion of samples:

30 ml of the sample were measured into a clean 250 ml dry Pyrex digestion flask. 25 ml concentrated aqua regia were added. The digestion flask was heated gently until frothing subsided. The samples were then heated to dryness, dissolved in 30 ml distilled water and filter with No. 42 filter paper. The solution was made up to volume in a 100 ml flask. This was used for the determination of the following elements: Cu, Zn, Pb, Mn, Ca, Cd, K and Fe by direct aspiration via atomic absorption spectrophotometer.

RESULTS AND DISCUSSIONS

pH

Table 1 shows the mean $\pm$ SD of the pH of the sampled energy drinks. The pH ranged from 4.47 ± 0.012 - 5.27 ± 0.017 . Sample HC had the least pH while sample WD had the highest. The results were higher than pH values of 2.75 – 3.66 reported by Mohammed *et al.*, (2012) for soft and energy drinks in Basrah, Iraq. They were within the pH ranged of 4.2 – 6.3 reported by Adeleke and Abiodun (2010) for local beverages in Nigeria, and had similarities with pH values of 4.2 – 6.3 reported by Obuzor and Ajaezi (2010) for malt beverages. All samples pH are acidic (i.e. their pH values are less than 7). The low pH values of these beverages may be attributed to the CO_2 gas used in the

preservation of these beverages or the presence of other acids such as citric acid, phosphoric acid, ascorbic acid, malic acid, tartaric acid used as preservatives (Bassiouny and Yang, 2005; Ashurst, 2005).

These acids inhibit the growth of microorganisms such as bacteria, mould and fungi which may contaminate the beverages. Drinking acidic beverages over a long period can erode tooth enamel and predispose the consumer to dental disease (Marshall *et al.*, 2003; Bassiouny and Yang, 2005).

The recommended pH for caffeinated drinks and coffee is ranged 4.7 to 6.0 (FDA, 2003). The pH of the analyzed energy drinks is in the range recommended by FDA for caffeinated drinks and coffee.

As shown in Table 1, mean pH are labeled a-i, which gives the order of increasing acidity, and represents the ANOVA analysis for the mean pH values of the sampled energy drinks. All pH of sampled energy drinks were significantly different from each other except for samples SO and EV, which were not significantly different from each other at $p < 0.05$ confidence limit.

Turbidity

The mean $\pm$ SD of the turbidity readings of the sampled energy drinks is shown in Table 1. The turbidity of energy drinks ranged from 53 ± 1.732 – 592 ± 1.155 NTU. These were lower than results reported by Obuzor and Ajaezi (2010) for malt drinks which had turbidity values above detection limits (>1000 NTU). Sample KM is least turbid while sample LB had the highest turbidity. Turbidity is the measure of the degree to which water loses its transparency due to presence of suspended particles. The more total suspended solids in the water, the murkier it seems and the higher the turbidity (Maurice, 2010). Turbidity is considered as a good measure of the quality of water. The suspended particles help the attachment of heavy metals and other toxic organic compounds which may pose negative health effects to the consumers.

Table 1: pH, Turbidity, Total dissolved Solids (TDS) and Conductivity of Energy drinks.

Samples	pH	Turbidity (NTU)	TDS (mg/L)	Conductivity (S/ μ cm)
SO	4.63 ± 0.012^c	126 ± 1.732^f	483 ± 0.577^c	1025 ± 1.732^d
BU	5.18 ± 0.023^h	68 ± 0.577^b	914 ± 2.309^h	1881 ± 0.577^l
HC	4.47 ± 0.012^a	84 ± 2.209^e	550 ± 0.577^d	1151 ± 0.577^e
LB	4.54 ± 0.012^b	592 ± 1.155^h	243 ± 0.577^a	497 ± 1.155^a
EV	4.60 ± 0.006^c	450 ± 0.577^g	477 ± 1.732^b	999 ± 1.732^c
PH	4.83 ± 0.017^e	$75 \pm 1.732^{c,d}$	762 ± 0.577^f	1577 ± 1.732^g
XL	5.04 ± 0.023^g	77 ± 1.155^d	676 ± 1.732^e	1410 ± 2.887^f
JW	4.99 ± 0.006^f	84 ± 2.309^e	849 ± 0.577^g	1761 ± 1.555^h
WD	5.27 ± 0.017^i	$71 \pm 0.577^{b,c}$	940 ± 0.000^i	1935 ± 1.555^j
KM	4.69 ± 0.006^d	53 ± 1.732^a	241 ± 0.577^a	516 ± 1.555^b

Results are expressed as Mean $\pm$ SD. a-i: Increasing order of TDS. a-j increasing order of conductivity. a-i: increasing order of pH. a-h: Increasing order of Turbidity.

Values with the same letters superscript in the column, by treatment, are not significantly different while those with different letters are significantly different at $p < 0.05$ from each other.

Table 2: Cadmium, Copper, Zinc, Lead and Manganese concentrations of energy drinks

Samples	Cd(mg/L)	Cu (mg/L)	Zn(mg/L)	Pb (mg/L)	Mn (mg/L)
PH	ND	ND	0.062 ± 0.0005	ND	0.003 ± 0.0001
XL	ND	0.002 ± 0.0002	0.049 ± 0.0009	0.054 ± 0.0010	ND
EV	ND	0.007 ± 0.0002	0.085 ± 0.0002	0.028 ± 0.0006	ND
BU	ND	0.012 ± 0.0001	0.045 ± 0.0001	0.068 ± 0.0002	ND
JW	ND	0.019 ± 0.0002	0.053 ± 0.0005	ND	ND
HC	ND	0.018 ± 0.0001	0.052 ± 0.0006	0.050 ± 0.0000	ND
WD	ND	0.021 ± 0.0001	0.050 ± 0.0002	0.078 ± 0.0005	ND
LB	ND	0.024 ± 0.0003	0.049 ± 0.0003	ND	ND
KM	ND	0.022 ± 0.0002	0.057 ± 0.0001	0.043 ± 0.0003	ND
SO	ND	0.070 ± 0.0006	13.887 ± 0.0037	0.139 ± 0.0004	ND

Results are expressed as MeanSD. ND: Not Detected

Table 3: Iron, Calcium and Potassium Concentrations of energy drinks.

Samples	Fe (mg/L)	Ca (mg/L)	K (mg/L)
PH	0.480±.0008	13.143±.0021	3.50
XL	0.418±.0002	5.386±.0005	3.50
EV	0.539±.0005	7.402±.0020	6.50
BU	0.294±.0005	2.831±.0011	2.00
JW	0.412±.0003	2.763±.0009	16.00
HC	0.326±.0006	8.531±.0013	6.50
WD	0.386±.0002	8.290±.0012	5.50
LB	0.586±.0010	4.707±.0008	110.00
KM	0.353±.0007	3.546±.0004	2.00
SO	1.961±.0003	5.117±.0001	25.50

As shown in Table 1, the mean turbidity of energy drinks are labeled a-h, which gives the order of increasing turbidity of the drinks and represents the ANOVA analysis for the mean turbidity values of the sampled energy drinks. There was no significant difference in the mean turbidity values of samples WD and BU, samples WD and PH, and samples PH and XL. The mean turbidity values of all other samples were significantly different at $p < 0.05$ confidence limit.

Total dissolved solids (TDS)

The total dissolved solids (TDS) ranged from 243 ± 0.577 – 940 ± 0.000 mg/L as shown in Table 1. These values were within the range of 327.37 – 1480 mg/L reported by Obuzor and Ajaezi (2010) for malt drinks. Sample WD had the highest TDS while sample LB had the least TDS. Beverages with high values of TDS are likely to contain metals (essential and toxic) at high concentrations which may cause adverse health effects when consumed.

As shown in Table 1, the mean TDS values are labeled a-i, which gives the order of increasing TDS of the analyzed samples and represents the ANOVA analysis for the TDS values of energy drink samples. There was no significant difference between the mean turbidity values of samples LB and KM. the mean turbidity values of all other samples were significantly different from each other at $p < 0.05$ confidence limit.

Conductivity

Conductivity is the ability of electricity to pass through water using the impurities contained in the water as conductors. When water has a lot of impurities, it is more conductive, however, if water is pure, it is less conductive unless it is polarized (Maurice, 2010). Hence, energy drinks conduct electricity because it contains ions and it follows that energy drink with the highest concentration of ions will conduct the most. The conductivity of energy drinks ranged from 497 ± 0.006 – 1935 ± 1.555 S/ μ cm as shown in Table 1. These values were within the range of 2.93 – 1999 S/ μ cm reported by Obuzor and Ajaezi (2010) for malt drinks. Sample LB is least conductive while sample WD has the highest conductivity. The mean conductivity of all sampled energy drinks was within the permissible limit for conductivity in beverages.

As shown in Table 4.1, the mean conductivity is labeled a-j, which gives the order of increasing conductivity of the samples and represents the ANOVA analysis for conductivity values of energy drink samples. The mean conductivity of all sampled energy drinks was significantly different from each other at $p < 0.05$ confidence limit.

Heavy metals

Heavy metals are commonly defined as those having a specific density of more than 5 g/cm³ (Duffus, 2002). The main threats to human health from heavy metals are associated with exposure to lead, cadmium, mercury and arsenic. Adverse health effects of heavy metals have been known for a long time. However, exposure to heavy metals continues and is on the increase. Table 2 shows the mean $\pm$ SD for the concentration of heavy metals in the sampled energy drinks.

Lead

Lead concentration of energy drinks ranged from 0.028 ± 0.0006 - 0.139 ± 0.0004 mg/L with sample EV having the least and sample SO having the highest concentration. Lead was not detected in samples PH, JW and LB as shown in Table 2. The lead concentration of energy drinks was lower than lead concentrations of $0.020 - 0.46$ mg/L reported by Krejpcio (2005). Onianwa *et al.*, (1999) reported lead levels of 0.04 ± 0.01 ppm in carbonated soft drinks 0.06 ± 0.08 ppm in fruit juice Nigeria. Maduabuchi *et al.*, (2006) also reported lead levels of $0.002 - 0.0073$ mg/L in canned drinks and 0.092 mg/l in non-canned drinks. These were lower compared to the values determined in energy drinks. The maximum contaminant level of lead is 0.01 mg/L (WHO 1993). Lead detected in samples was above the maximum contaminant level (MCL).

Copper

The copper concentration of energy drinks ranged from 0.002 ± 0.0002 - 0.070 ± 0.0006 mg/L. Sample XL had the least concentration while sample SO had the highest concentration of copper. Copper was not detected in sample PH as shown in Table 2. The concentration of energy drinks was low compared to the values $0.047 - 1.840$ mg/L reported by Krejpcio *et al.* (2005) for fruit juice samples in Poland. The concentrations was however higher than values $0.01 - 0.02$ mg reported by Obuzor and Ajaezi (2010) in malt beverages, and values $0.0004 - 0.001$ mg/kg, reported by (MAFF, 1998) determined in non-alcoholic beverage. The maximum contaminant level (MCL) of copper is 1.0 mg/L (WHO 1993). All sampled energy drinks had concentrations below the MCL of copper.

Manganese

Manganese was detected only in samples PH as shown in table 2 above and has a concentration of 0.003 ± 0.0001 mg/L. The value was low compared to $0.001 - 0.730$ mg/L for canned and $0.001 - 0.209$ mg/L for non-canned beverages reported by Maduabuchi *et al.*, (2006) but close to 0.01 mg reported by Obuzor and Ajaezi (2010) and $0.01 - 0.021$ reported by Mohammed *et al.*, (2012). The maximum contaminant limit (MCL) of manganese is 0.05 mg/L (US EPA). The concentration of manganese detected in sample PH was below the MCL. Deficiency in manganese leads to various health problems, which may include bone malformation, eye and hearing problems, high cholesterol levels, hypertension, infertility, weakness, heart disorders, memory loss, muscle contraction, tremors, seizures (Institute of Medicine, 2001). It could also result in decreased learning ability in school-aged children and increase the propensity for violence in adult (Finley, 2004).

Zinc

The concentration zinc was with in range of 0.045 ± 0.0001 - 13.887 ± 0.0037 mg/L. Sample SO had the highest concentration of 13.887 ± 0.0037 mg/L. These values were similar to those reported by Bingol *et al.*, (2010) and Krejpcio *et al.* (2005) except for sample SO. The maximum contaminant limit for zinc is 5.0 mg/L (WHO 1993). The zinc concentration of all sampled energy drinks were below the MCL of zinc. Only sample SO had a concentration higher than the MCL of Zinc. Zinc is involved in numerous aspects of cellular metabolism. It is required for the catalytic activity of approximately 100 enzymes and it plays a role in immune function, protein synthesis, wound healing, DNA synthesis, and cell division (Institute of Medicine, 2001; Prasad, 1995). A daily intake of zinc is required to maintain a steady state because the body has no specialized zinc storage system (Ryan-Harshman and Aldoori, 2005; Institute of Medicine, 2001).

Cadmium

Cadmium was not detected in all energy drinks as shown in Table 2. Cadmium intake in relatively high amount can be detrimental to human health. Over a long period of intake, cadmium may accumulate in the kidney and liver, and because of its long biological half-life, may lead to kidney damage (Maduabuchi *et al.*, 2006).

Iron

The iron level of energy drinks ranged from 0.294 ± 0.0005 - 1.961 ± 0.0003 mg/L with sample SO having the highest value and sample BU having the least as shown in Table 3. The values were lower compared to $0.020 - 2.090$ mg/L for non-canned and $0.020 - 2.460$ mg/L for canned beverages reported by Maduabuchi *et al.*, (2006) but higher than values $0.11 - 0.28$ mg/L reported by Obuzor and Ajaezi (2010). The maximum contaminant level (MCL) or iron is 0.03 mg/L (WHO, 1993). All the sampled energy drinks had iron concentration higher than the MCL. Iron is important in many biological processes because it is an ideal oxygen carrier and because it can function as a protein-bound redox element. Iron deficiency is common worldwide and in infants can cause severe neurological deficit (Prasad, 1995).

Calcium

Calcium concentration of energy drinks ranged between 2.763 ± 0.0009 – 13.143 ± 0.0011 mg/L, with sample JW having the least and sample PH having the highest value. The calcium concentration of energy drinks were low compared to 0.28 – 262 mg/L reported by Obuzor and Ajaezi (2010). Calcium is needed for the formation and maintenance of bones, the development of teeth and healthy gums. It is necessary for blood clotting, stabilizes many body functions and is thought to assist in preventing bowel cancer (Attieh, 1999). It has a natural calming and tranquilizing effect and is necessary for maintaining a regular heartbeat and the transmission of nerve impulses. The required amount include: 1,000 mg/day for people aged 19 - 50 years and 1,200 mg per day for people over the age of 51 years. The maximum level of calcium is 2.5 g/day (Ryan-Harshman and Aldoori, 2005). Rickets, tetany, and osteoporosis can result its deficiency. Hypertension and colon cancer may relate to chronic low intake.

Potassium

Potassium concentrations of the energy drinks were relatively low compared to the recommended daily intake (RDI) of potassium. It ranged from 2.0 to 110 mg/L as shown in Table 3. The RDI of potassium ranged between 1600-5000 mg/day. Potassium is the major intracellular ion, intimately related to sodium movement out of the cell via Na/K ATPase. As such, it participates in maintaining normal membrane potential in cells. Potassium is also a major factor in osmotic fluid dynamics in the body. Renal control mechanisms keep blood levels within a narrow range. Normal or increased potassium antagonizes uptake/retention. Potassium deficiency results to irregular/rapid heart beat, High Blood Pressure (HBP), kidney disease, infrequent menstrual cycles, ovarian cysts, muscle spasms/cramps, joint pains, weakened immune system and hyperglycemia. Excess potassium results to hypotension, reactive hypoglycemia, and increased risk for cancer. Toxicity is possible via supplements at doses near 18g or in kidney dysfunction.

Conclusion

Iron, calcium, zinc and potassium were found in all the energy drinks. These are essential elements needed for the general well being of the body. Although their values were low compared to the acceptable total intakes, their presence contributes to the daily iron, calcium, zinc and potassium sources needed in the body. Cadmium was not detected in all energy drinks. It is a non-essential metal and produces only significant adverse health effects. The lead concentration of energy drinks were above the MCL of 0.01 mg/L while copper and manganese concentration of energy drinks were below the MCL of 1.0 mg/L and 0.05 mg/L respectively. The physical chemical properties such as pH, total dissolved solids, turbidity and conductivity were also investigated. These were in conformity with the standards set by regulatory bodies such as FDA and WHO.

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