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Evaluation of Physicochemical and Microbial Qualities of Bottled Water Samples Vended Around a University in the South-East, Nigeria

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Abstract

This study evaluated the quality of four (4) different bottled water samples being sold in one of the environs of Michael Okpara University of Agriculture, a University in the South East, Nigeria. Standard methods were used in assessing the physicochemical and microbiological qualities of the water samples. Results showed that coliforms and fungi were not detected in the samples, hence comply with world health organization (WHO) requirement for drinking water. However, total bacteria counts ranged from 1.54 to 2.08 log CFU/ml, deviating slightly from requirement of WHO. Results of the physicochemical parameters were within the WHO acceptable limits, with the exception of alkalinity (140.35-145.23 mg/l) and phosphorous (0.79-0.94 mg/l) which were above the permissible limits of 50mg/l and zero respectively. In conclusion, some of the physicochemical quality attributes of the bottled water samples did not conform with the WHO requirement for drinking water, making the current finding of public health significance to consumers of bottled water in the University. Strict adherence to quality control measures should be observed by manufacturers of the bottled water to help improve quality and protect public health.

Keywords

Bottled Water, Physicochemical, Microbiological, Drinking Water, Quality Control, Public Health

1. Introduction

Water is the medium for all biochemical reactions in man. It houses the largest number of living organisms when compared with other habitats and comply with certain physical, chemical and microbiological standards [1]. It has been established that good quality water produces healthier humans than one with poor quality [2]. Water quality is essentially determined by its physical, chemical as well as microbiological characteristics [3]. Such water should be palatable, oxygenated, colorless, odorless and free from harmful organisms and salts of heavy metals. Most reported cases of cholera and abdominal infections in clinical centers could be traceable to the consumption of harmful particles and microorganisms in water and beverages).

Bottled water is a term referring to water that is packaged and sold in containers or bottles, which is generally regarded as safe for usage by people [4]. Bottled water production companies are one of the fastest growing industrial sectors in developing nations of the world, particularly Nigeria.

High rate of consumption of water has resulted in the establishment of different water bottling companies, howbeit with minimal interest ensuring good quality of the product; thereby resulting in deaths of consumers due to waterborne diseases, in some cases. With the increasing demand for water, there is risk to public health if quality of the product is not given required attention by manufacturers.

Safe drinking water is fundamental to human lives. Bottled water is among the commonly vended packaged water in Michael Okpara University of Agriculture (MOUAU), a popular university in the South East of Nigeria. Due to the health challenges, such as gastrointestinal disorders and other illnesses, associated with water consumption, the present study was aimed at assessing the quality of bottled water being sold in a major environ of MOUAU), so as to help promote public health.

2. Materials and Methods

2.1. Sources of raw materials

The bottled water samples used for the study were randomly collected different retail outlets in a major environ of MOUAU, Abia State, Nigeria. One of the samples (sample A), obtained from a standard manufacturer of bottled water served as control. Asepsis and cleanliness were observed during collection to avoid possible contamination, after which the water samples were taken to the laboratory for analysis.

2.2. Physicochemical analysis

The physicochemical parameters determined in the water samples include pH, chlorides, nitrates, hardness and electrical conductivity, using standard methods described by Association of Official Analytical Chemists [5]. Analysis was carried out at the chemistry laboratory of National Root Crops Research Institute, Umudike, Umuahia, Abia State.

2.3. Microbiological analysis

2.3.1. Enumeration of total coliforms

A most probable number (MPN) test was used to enumerate the total coliforms in the packaged water samples by following the method described by Olaoye and Onilude [6]. The test was performed sequentially in three stages: presumptive, confirmed and completed tests. Double and single strength lauryl tryptose (lactose) broths in tubes were incubated with water volumes 10 ml, 1.0 ml and 0.1 ml in presumptive test. Tubes that were positive for gas production after 24 hrs incubation at 35^oC were inoculated into brilliant green lactose bile broth for confirmed test. Tubes showing positive reactions were used to calculate the most probable number (MPN) of coliforms in the water samples with reference to the statistical table described by Tillett [7]. For completed test inoculations were made on Eosin Methylene Blue (EMB) agar plates, nutrient agar slants and brilliant green lactose broths and preparation of Gram-stains were used to establish the presence of coliforms in the samples.

2.3.2. Total bacteria count

Plate count agar (PCA, Oxoid) was used for enumeration of total bacterial count (TBC). This was performed using the serial dilution agar plating method at suitable dilutions levels [8]. One millilitre each of appropriate diluted suspensions was transferred into sterile petri dishes and molten agar medium (~45^oC) was carefully poured while swirling gently. The plates were then incubated at 37^oC for 24 h, after which visible colonies were counted and results were expressed in cfu/ml. Plating of samples was done in replicates.

2.3.3. Fungi count

The enumeration for fungi, including yeasts and moulds, was performed using the methods of Olaoye and Onilude [9].

3. Statistical analysis

Data obtained in this study were reported as means of duplicate analyses. One Way analysis of variance of completely randomized design was adopted using the Statistical Package for service solution (Version 22), for comparison of between mean values. Means were separated using Duncan multiple range test and significance difference was determined at 5% level of confidence ($p < 0.05$).

4. Results and Discussion

Table 1 shows the result of the microbiological analysis of the four water samples. The total bacterial counts (TBCs) ranged from 1.54 to 2.08 log CFU/ml. Sample A (which served as control) had the lowest count (1.54) and was significantly different ($p \leq 0.05$) from samples C and D. The low TBC of sample A could be due to the stringent quality control measures adopted during production, including purification and bottling processes. However, the TBC of the water samples under investigation do not conform to the world health organization acceptable requirement (zero log CFU/ml) bacteria for drinking water. The presence of bacteria could be an indication of poor sanitary condition, and other environmental pollution or contamination [10]. The results of the TBC obtained in this study could be corroborated by that reported by Khothalang *et al*. [11] in a similar finding. According to the authors, TBCs of between 1.77 and 3.38 log CFU/ml were recorded in bottled water samples taken from Lesotho, South Africa.

Fungi and coliforms were not detected in the water samples in the present investigation, thus conforming to the WHO requirement of zero tolerance. This suggests that the bottled water samples satisfy the coliform level tolerance and could make them acceptable for human consumption. The observation could also indicate that required sanitary and hygiene measures were taken by manufacturers during production. The absence of fungi in the samples also suggests that the presumed quality control measures taken by the manufacturers may have helped in their elimination or reduction below detection level.

Presented in Table 2 are the physical properties of the water samples. There were varying levels of significant difference ($p \leq 0.05$) between the electrical conductivity, alkalinity, pH, total dissolved solids, total soluble solids and total hardness of the water samples. Pure water is not a good conductor of electric current rather a good insulator. Increase in ionic concentration enhances the electrical conductivity of water. Generally, the amount of dissolved solids in water determines the electrical conductivity. Electrical conductivity (EC) measures the ionic potential of a solution that enables it to transmit current. According to WHO standards, EC value in drinking water should not exceed 40 µS/cm. The EC of the water samples ranged from 37.19 µS/cm to 42.39 µS/cm, and hence fall within the acceptable limit of WHO, however with some recording a little higher even though may be considered insignificant. Sample A (control) was found to have the lowest EC value (37.19 µS/cm) and was significantly different ($p \leq 0.05$) from those of other samples; the low EC value of sample A could be due to likely reduced amount of dissolved solids in the water sample usually caused by the process of demineralization. This is similar to findings reported by Khothalang *et al*. [11].

The alkalinity value of the water samples ranged from 140.35 to 145.23 mg/l $CaCO_3$, which is above the permissible limit of 50 mg/l $CaCO_3$ recommended by the WHO. The values of alkalinity obtained in this study were at variance with those reported by Khothalang *et al*. [11], and this may be due to difference in treatment procedures adopted during production. Alkalinity (measured by calcium carbonate concentration) is a total measure of the substances in water that has acid neutralizing ability. Sample A was found to have the highest alkalinity content (145.23) and was significantly different ($p \leq 0.05$) from the other samples.

pH is an important parameter in evaluating the acid-base balance of water. It is also an indicator of acidic or alkaline condition of water status. WHO has recommended maximum permissible limit of pH from 6.5 to 8.5 in drinking water. The pH values of the water samples ranged from 7.11 to 7.39, and thus within acceptable range of WHO. Sample A was found to have the highest pH (7.39) and was not significantly different ($p > 0.05$) from those of other samples. The slight variation in the pH values of the water samples could be due to possible environmental conditions, which may have impact on alkalinities. pH values recorded in the present investigation is similar to those reported made by other researchers [12, 13]. The pH of drinking water has no immediate direct effects on human health, as the stomach acts as a natural buffer which stabilizes the pH. However, pH may have some indirect health effects when used in food processing, where it may bring changes in other water quality parameters, such as solubility of metals and survival of pathogens [14].

Water has the ability to dissolve a wide range of inorganic and organic minerals or salts such as potassium, calcium, sodium, bicarbonates, chlorides, magnesium and sulfates among others. These minerals may bring about undesirable taste and appearance of water. Occurrence of high total dissolved solids (TDS) in water indicates that the water could be highly mineralized, that is containing high level of minerals. Therefore, TDS is a very important parameter when water is being considered for drinking. Desirable limit for TDS in drinking water is 500 mg/l and maximum limit is 1000 mg/l. The TDS in the water samples in the present study ranged from 192.37 to 196.49 mg/l, which is within the standard recommended by the world health organization. Water sample A was found to have the highest TDS (196.49), and significantly different ($p \leq 0.05$) from the rest of the samples. The low TDS value of the other samples could be attributed to possible process of demineralization due to environmental conditions or different treatments given to the water samples during production.

Water hardness depends on anions such as bicarbonate, sulphate and chloride and major cations, such as calcium and magnesium, which are all below the permissible limits. Water has been classified as soft (0-75 N/mm^2), moderately hard (75-150 N/mm^2), hard (150-300 N/mm^2) and very hard (>300 N/mm^2) [15]. From the result in Table 2, the total hardness ranged from 120.13 to 125.15 N/mm^2, indicating that the water samples are moderately hard, and fall within the WHO requirement (200 N/mm^2). Sample D was found to have the highest hardness (125.15 N/mm^2) and was significantly different ($p \leq 0.05$) from the rest of the samples. The higher level of hardness of sample D could be due to likely high contents of calcium, magnesium and carbonate ions, which are the main causes of hardness in water.

Table 3 shows the result of chemical analysis of the water samples. There was significant difference ($p \leq 0.05$) in the chloride, nitrate, phosphorus and fluoride contents. Fe was not detected in the water samples, which is in compliance with WHO requirement of below 0.3 mg/l. The chloride value ranged from 25.31 to 32.02 mg/l, which also satisfy the

WHO standard of below 250 mg/l in drinking water. However, the value of chloride was found to be higher than those reported by Khothalang *et al*. [11]. The researchers recorded chloride contents of 0.02 to 0.08 mg/l in bottled water samples from South Africa. Sample D was found to have the highest chloride content and was significantly different ($p \leq 0.05$) from the other samples. The higher chloride value of sample D could be due to the high content of chlorine in the water, probably influenced by the chlorination process. The chlorine may have reacted with the calcium and magnesium ions to form chlorides.

Nitrate is one of the most important factor in water that may cause a condition known as blue baby syndrome in infants. The sources of nitrate are nitrogen cycle, industrial waste, nitrogenous fertilizers etc. WHO maximum permissible limit of nitrate is 5 mg/l in drinking water. The nitrate content of the water samples ranged from 0.27 to 0.34 mg/l, which is within the permissible limit of WHO. Sample A was found to have the lowest nitrate content (0.27), and there was no significant difference ($p > 0.05$) between the nitrate contents of the water samples. Hence, the quantities of nitrates in the water samples could be considered acceptable for drinking.

Table 1. Counts (log CFU/ml) of bacteria, fungi and coliforms in the water samples

Samples / Parameters	TBC	TFC	TCC
A	$1.54^{c} \pm 0.13$	-	-
B	$1.58^{b} \pm 0.07$	-	-
C	$2.08^{a} \pm 0.04$	-	-
D	$1.61^{b} \pm 0.02$	-	-

Counts are expressed as logarithm of colony forming units per milliliter of water (log CFU/ml)
Values are means of the replicate determinations. Means with different superscripts within same column are significantly different ($p \leq 0.05$).
A, B, C, D are samples of bottled water collected from an environ of the University;
TBC, total bacteria count; TFC, total fungi count; TCC, total coliform count

Table 2. Results for physical analysis of the water samples

Sample / Parameters	Electrical conductivity, EC (µS/cm)	Alkalinity (mg/l $CaCO_3$)	pH	Total dissolved solids, TDS (mg/l)	Total soluble solids, TSS (mg/l)	Hardness (N/mm^2)
A	$37.19^{d} \pm 3.21$	$145.23^{a} \pm 0.21$	$7.39^{a} \pm 0.24$	$196.49^{a} \pm 0.021$	$145.02^{a} \pm 4.28$	$120.10^{d} \pm 0.035$
B	$42.39^{a} \pm 0.32$	$140.35^{d} \pm 0.14$	$7.11^{c} \pm 0.16$	$143.56^{c} \pm 0.021$	$124.35^{b} \pm 1.33$	$124.35^{b} \pm 0.028$
C	$124.35^{b} \pm 1.28$	$143.26^{b} \pm 0.17$	$7.25^{b} \pm 0.66$	$195.67^{b} \pm 0.021$	$144.81^{b} \pm 4.23$	$121.02^{c} \pm 0.028$
D	$40.25^{b} \pm 2.21$	$142.44^{c} \pm 0.19$	$7.22^{b} \pm 0.71$	$194.54^{c} \pm 0.028$	$140.73^{d} \pm 2.41$	$125.15^{a} \pm 0.021$
LSD	0.35	0.34	0.33	0.53	0.43	0.37

Values are means of the replicate determinations. Means with different superscripts within same column are significantly different ($p \leq 0.05$)
A, control; B, C, D are samples of bottled water collected from an environ of the University;
LSD, least significant difference

Table 3. Result for the chemical analysis of the water samples

Samples / Parameters	Fe	Chloride	Nitrate	Phosphorus	Copper	Fluoride
			mg/l			
A	-	$25.31^{d} \pm 0.31$	$0.27^{b} \pm 0.14$	$0.79^{b} \pm 0.02$	-	$0.07^{b} \pm 0.02$
B	-	$29.24^{c} \pm 0.28$	$0.325^{a} \pm 0.01$	$0.94^{a} \pm 0.04$	-	$0.15^{a} \pm 0.01$
C	-	$30.56^{b} \pm 1.73$	$0.315^{ab} \pm 0.02$	$0.875^{a} \pm 0.07$	-	$0.08^{ab} \pm 0.01$
D	-	$32.02^{d} \pm 2.34$	$0.34^{a} \pm 0.03$	$0.88^{a} \pm 0.02$	-	$0.11^{ab} \pm 0.04$
LSD	-	0.52	0.42	0. 49	-	0.39

Values are means of the replicate determinations. Means with different superscripts within same column are significantly different ($p \leq 0.05$)
A, control; B, C, D are samples of bottled water collected from an environ of the University;
LSD, least significant difference

The fluoride value ranged from 0.15 to 0.07 mg/l. Sample A was found to have the lowest fluoride content (0.07) and was not significantly different ($p>0.05$) from samples C (0.08) and D (0.11). The optimal concentration of fluoride for dental health in drinking water is generally between 0.5 and 1.5 mg/l [16]. This may largely be influenced by the volume of water consumption by individuals, as well as intake and exposure from other sources. The fluoride contents of the water samples in the present study were within the acceptable range.

Sample B had the highest phosphorus content (0.94 mg/l) and was not significantly different from others. The phosphorus contents of the water samples did not comply with WHO standards, which requires zero content of the element. Presence of phosphorus in water is considered an indication of contamination, and may pose a serious health problems to human. There was no detection of copper in all the water samples analyzed.

5. Conclusion

This study revealed that the physicochemical properties of the bottled water samples were within the range specified by the world health organization, with the exception of alkalinity. The values of Fe, chloride, nitrate and fluoride were within permissible limits. Microbial analysis did not reveal the presence of fungi and coliforms, thus conforming with WHO requirement. However, total bacteria counts of some of the water samples did not conform with permissible limits, thus raising public health concerns.

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