MICROBIOLOGICAL PROFILE OF BOTTLED AND TAP DRINKING WATER IN BRGY. SAN MIGUEL, ILIGAN CITY

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Background: *The rapid rise of waterborne diseases and the growing concern about water contamination has prompted the researchers to examine the bacteriological quality of drinking water in Brgy.San Miguel, Iligan City.*

Objective: *The purpose of this study is to check the presence of non-fecal coliform, fecal E. Coli and heterotrophic bacteria in bottled and tap drinking water and determine whether they meet the criteria set by WHO.*

Methods: *Eighteen samples of bottled and tap water in 3 different sites were analyzed for total bacterial count and presence of other bacterial indicators of drinking water quality.*

Results: *An alarming 77.78% of tap water and 88.89% of bottled water had non-fecal coliform counts above the World Health Organization standards. 55.56% and 44.44% of tap and bottled water samples exceeded the criteria of zero fecal E. coli coliform per 100 ml of drinking water (WHO 2011). Basing on the WHO standards 55.56% of tap water and only 22.22% of bottled water has total heterotrophic bacterial load beyond tolerable limit making them unsafe for human consumption.*

Conclusions: *The fecal and non-fecal bacterial contamination in both tap and bottled drinking water is a serious threat to public health. Stringent tap water inspection and rigorous quality control of bottled water industry in needed in Iligan City.*

Keywords: *Tap drinking water, bottled water, contamination, coliform*

INTRODUCTION:

Waterborne diseases killed more than 3.4 million people annually, making it the leading cause of death worldwide (Berman 2009) .It is a stark terrifying reality that a child dies every 20 seconds due to water acquired illnesses. Infectious diarrhea, one of these illnesses is the second most known cause of death below five years of age which makes it more dangerous than AIDS, malaria and measles combined .(Circle of Blue 2009). 70-80% of health problems in developing countries were brought about by waterborne diseases (Jayana 2009).

As a way of protecting themselves, people opted to drinking bottled water. The study of (Chakraborti et al., 2010; Islam et al., 2006) showed that the substantial increased in bottled water consumption is due to outbreaks of diarrheal diseases caused by bacterial contamination of drinking water. The sale of bottled water has reach to more than USD 35 billion around the world (Raj, 2005) In 2011, America purchased 9.1 billion gallons of bottled water. Per capita consumption reached a new peak of 29.2 gallons from 18.2 gallons per person about a decade ago (Fishman , 2012) According to ‘ The World’s Water 2006-2007 data, per capita consumption of bottled water in the Philippines, jumped from 12.6 liters per person in 1999 to 16.4 liters per person in 2004 as calculated by the Beverage Marketing Corporation BMC (Gleick 2006).

Inspite of the belief that bottled water is clean and safe, many studies showed otherwise (Pasumbal, et. Al 2005) Kassenga 2007; Svagzdiene et al., 2010) Total coliform (Bharath et al., 2003). Fecal E. Coli (Moniruzzaman et. al., 2011) and heterotrophic bacteria (Farhadkhani 2014) was identified in mineral bottled water. The research findings of (Bartram et al.,2004; Kassenga 2007; Svagzdiene et al., 2010) revealed heterotrophic and fecal bacteria of bottled water above acceptable limit.

On the other hand, drinking tap water was associated with outbreaks in the recent years. (Fredrick 2015; Bhunia, 2009; Saha, 2009). The study of Saxena, 2015 revealed that waterborne outbreaks caused by E. Coli resulted to high prevalence of mortality around the globe. Kassenga 2007 also detected the presence of total coliform and fecal coliform organism in tap water. According to (Braeye 2015) the point source of tap water can be contaminated with pathogens. Total coliform bacteria present in drinking water treatment plant indicate a serious treatment failure (Health Canada 2006). Environmental Protection Agency explained that total coliforms naturally inhabit the soil and surface waters (rivers, lakes, etc.). Contamination of drinking water can mean possible change or breach in the integrity of water system and bacteria may have entered your drinking water.

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There are doubts about the purity of both tap and bottled water however no study has been conducted in Iligan City to assess its quality.

This study was undertaken in order to analyze the bacterial contamination of tap and bottled mineral water in Iligan city and to determine their compliance with the WHO standards.

METHODS:

Research Design:

This study follows an experimental approach specifically utilizing the Descriptive- Comparative Method .This design was chosen to attain the objectives stated in this study, mainly by comparing the microorganisms present in the water consumed by the households, both tap water and bottled water.

Sample Collection:

Selection of household was done through Random sampling method to avoid self-selection bias. In getting the samples from each household in 3 different sites, the researchers were given 18 bottles by the Waterworks System. Two bottles were used per household (one bottle serves as a container for the raw water and the other for the bottled water). The bottles were then labeled as to which site and house number they belong and the time it was taken.

Sample Analysis:

All collected samples were analyzed for the following parameters in accordance to the standards set by WHO. The researchers analyzed the samples with the assistance of the Iligan City Waterworks System.

Bacterial profile identification:

Because it is very expensive and time consuming to test for each pathogenic organism, heterotrophic bacteria, coliform bacteria and E. coli, which originate in environmental and animal sources, serve as good pathogenic indicator organisms, are more abundant, and easier to identify than other pathogens (Wilhelm and Maluk, 1999).

In testing for the bacterial count of Fecal E. coli and Non-Fecal Coliform, Multiple Tube Fermentation Technique (MTFT) was utilize and in testing for the Heterotrophic Platelet Count, Plate Count Method (PCM) were established.

DETERMINATION OF THE PRESENCE OF NON-FECAL COLIFORM BACTERIA: MULTIPLE TUBE FERMENTATION TECHNIQUE

PRESUMPTIVE PHASE:

Lauryl tryptose Broth (Triple Strength) was used in this phase with 10ml media plus 20ml sample in 5 tubes incubated for 48 hours at 35 +/- 0.5 C. Production of an acidic reaction or gas in the tubes or bottles within 48 ± 3 h constitutes a positive presumptive reaction. The tubes with positive presumptive reaction were submitted to the confirmed phase.

CONFIRMATORY PHASE:

The inoculated brilliant green lactose bile broth tube was incubated at 35 ± 0.5°C. Formation of gas in any amount in the inverted vial of the brilliant green lactose bile broth fermentation tube at any time within 48 ± 3 h constitutes a positive confirmed phase. The Most Probable Number (MPN) value from the number of positive brilliant green lactose bile tubes was calculated.

COMPLETED PHASE:

Using aseptic technique, one LES Endo agar plate was streak from each tube of brilliant green lactose bile broth showing gas.

Plates (inverted) are incubated at 35 ± 0.5°C for 24 ± 2 h. Growth from each isolate was transferred to a single-strength lauryl tryptose broth fermentation tube .Secondary broth tubes (lauryl tryptose broth was incubated with inverted fermentation vials inserted) at 35 ± 0.5°C for 24 ± 2 h. Using drops of distilled water ,separated light emulsions of the test bacterial growth and positive and negative control cultures was prepared on the same slide. Formation of gas in the secondary tube of lauryl trypotose broth within 48 ± 3 h and demonstration of gram-negative, nonspore-forming, rod-shaped bacteria from the agar culture constitute a positive result for the completed test, demonstrating the presence of a member of the coiform group.

Most Probable Number

The MPN method by serial dilution was used to introduce one bacterium into a fermentation tube containing media for the bacteria to thrive on. The gas production or the lack of it can help determine the probable number of bacteria in the sample. Coliform

bacteria concentration in the sample was expressed as number of bacteria per 100 mL.

IDENTIFICATION OF FECAL E. COLI BACTERIA: MULTIPLE TUBE FERMENTATION TECHNIQUE

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Lauryl tryptose Broth (Triple Strength) was used in this phase with 10ml media plus 20ml sample in 5 tubes incubated for 48 hours at 35 +/- 0.5 C. Production of an acidic reaction or gas in the tubes or bottles within 48 ± 3 h constitutes a positive presumptive reaction. The tubes with positive presumptive reaction were submitted to the confirmed phase.

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DETERMINATION OF TOTAL HETEROTROPHIC BACTERIAL COUNT: PLATELET COUNT METHOD

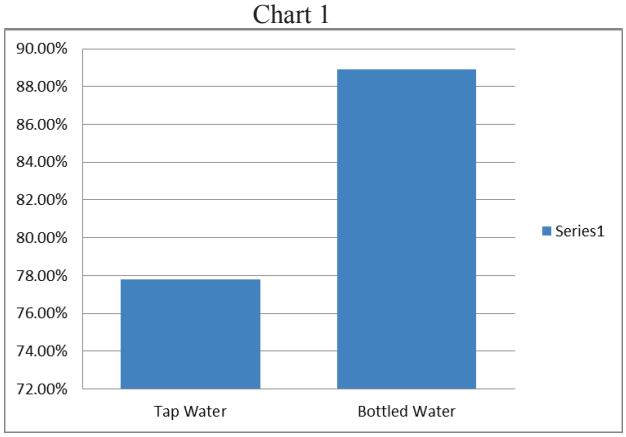
The identification of total heterotrophic bacterial count was done utilizing both the serial dilution and the pour plate technique. Serial 10 fold dilutions in sterile water were carried out and 1 ml of each dilution was aseptically placed in sterile petri-dishes in triplicates. 20 ml of molten plate count agar (Oxoid) cooled to 45°C was then added to each of the plates and mixed completely. The mixture was allowed to solidify and for 24-72 hours the plates incubated at 22°C and 37°C. The number of bacterial colonies were counted and recorded as colony-forming units per millilitre.

RESULTS AND DISCUSSION:

Drinking water can still be potable even if it is not completely free from microorganisms, provided that its number does not go beyond the range acceptable by WHO guidelines. The heterotrophic bacterial count should not cross above 50 cfu ml while Coliform and fecal e coli must be 0 cfu/ml (WHO 2011).

NON-FECAL COLIFORM TAP AND BOTTLED WATER ANALYSIS

Chart 1



Non –fecal coliform bacteria was detected in 77% of tap water and 88.89% of bottled water and making them unsafe for human consumption. These findings coincide with the study of Tsega (2013) which

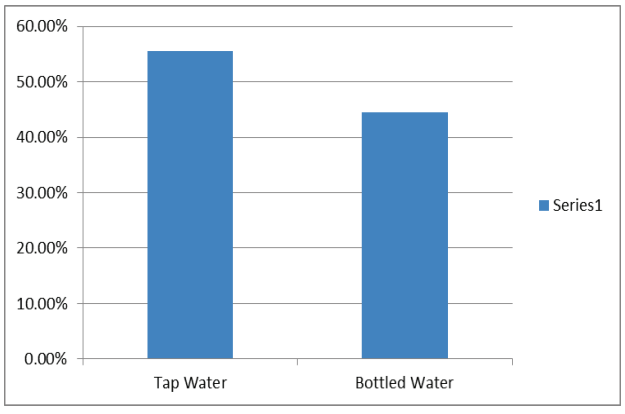
reported that tap water in rural location was heavily contaminated with coliform. In Bahir dar city, Tabor 2011 found out twenty seven (77.1%) of the household water samples had high total coliforms counts. Possible reason of coliform contamination is a crack or poor construction of the water system (Ercumen 2014). Coliform bacteria can also enter a well connected to a pipe leading to the house contaminating tap drinking water .In addition if the well cover is not airtight, mice, insects and other bacteria going into the well can infect tap water source.

Coliform contamination of bottled water was also reported in Bangladesh (Moniruzzaman 2011) Nigeria (Onweluzo 2010) Egypt (Magda 2008) and Trinidad (Bharath, et. Al. 2003) The presence of coliform indicates a need for strict monitoring of hygienic practices. Low level hygiene and poor sanitation was pointed out by tabor 2011 as causes of coliform contamination. Another contributing factor is the possibility of infection during the bottling process (WHO 2011) which is an important point of view in hygiene.

In contrast 22% of tap water and 11% bottled water conform to the WHO standards of 0 cfu/ml coliform bacterial count. A certain bottling company in Ethiopa also showed no presence of coliform in their bottled water (Biadglegne 2009).This implies that conforming to the high standards of WHO are not beyond our reach.

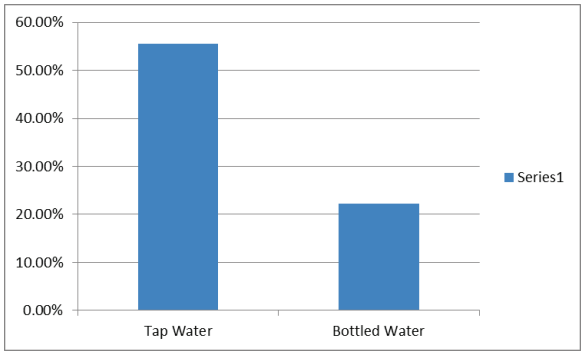
The findings of this study showed more coliform bacteria contamination in bottled than tap water negating the assumption that bottled water is safer than tap water.

FECAL E. COLI TAP WATER AND BOTTLED WATER ANALYSIS



The presence of E. coli in drinking water is a confirmation of fecal contamination. Their occurrence suggests a strong likelihood that animal or human wastes are infiltrating the water system. The table above shows that 55.56% and 44.44% of tap and bottled water samples exceeded the criteria of zero fecal E. coli coliform per 100 ml of drinking water (WHO 2011). The percentage of bottled water samples in this study is higher than previous study of kasengga (2007), Bharath (2003) indicating a more serious problem. This requires immediate attention for public safety. The study of Saxena (2015) identified E. coli from water sources as the culprit behind water-related outbreaks causing high prevalence of mortality worldwide. A study in Pakistan found out that most mineral water samples were contaminated with E. coli (Taj & Baqai, 2007).These findings suggest that tap and bottled water contaminated by E.coli endangers consumers especially the immunocompromised, hospitalized patients infants and elderly (Leclerc et. Al, 2002).The occurrence of coliform and E coli in tap and bottled water samples not only indicates occurrence of potential presence of other harmful organisms but also raises concern about the integrity and effectiveness of water processing and production system.

HETEROTROPHIC PLATE COUNT TAP AND BOTTLED WATER ANALYSIS



Heterotrophic Plate Count measures the overall bacteriological quality of drinking water. Basing on the WHO standards 55.56% of tap water and only 22.22% of bottled water has total heterotrophic bacterial load beyond tolerable limit. In contrast, Farhadkhani (2014) study showed that heterotrophic contamination of bottled water is significantly higher than tap water. Kasenga (2007) detected 92% of heterotrophic bacteria in bottled water samples which is far higher compared to this study which revealed only 22%.

Generally heterotrophic bacteria are considered to be harmless part of the environment and is also found in soil, on the skin, and in considerable numbers in food products such as vegetables, fruits, meats, etc.(Reynolds, 2005)

The presence of this non-pathogenic heterotrophs even in high numbers does not necessarily pose health risk but is significant to immunocompromised patients, people with diabetes, cancer, etc.(Bartram 2003).Considerable numbers of heterotrophic bacteria in bottled water is an indication of poor practices during the processing of drinking water (Magda 2008).According to National Primary Drinking Water Regulations established by the U.S. EPA low heterotrophic bacteria count in the drinking water is related to a good maintenance of the distribution and treatment management.

On the other hand, high heterotrophic count can be a breeding ground for dangerous bacteria, like E. Coli, causing foul-tasting water, leading to corrosion or slime growth in pipes.

Table 4:TEST DIFFERENCE OF TAP AND BOTTLED WATER BACTERIAL LOAD

	Wilcoxon W value	P value	Remarks
Fecal E. Coli	9	0.460	NS
Non Fecal	8.67	0.494	NS
Heterotrophic	8.67	0.489	NS

As a protective measure, bottled water was advertised as a better option especially for immune-compromised people, and infants. (Warburton et al., 1992).Bottled water was perceived to be safer and purer than tap water but this study shows no significant difference in their fecal, non fecal and heterotrophic bacterial load. Various studies reported that drinking bottled water is not always safe(Zeenat, et al., 2009, Venieri et al, 2006) According to Leclerc et al 2002, contaminated bottled water poses great danger to hospitalized patients, elderly and infants.

CONCLUSIONS:

Majority of the tap and bottled water samples in this study contain bacteria at an alarming number. These findings underscore the importance of regulatory agencies in Iligan City. The assumption that bottled water is always safer needs to be corrected. Vigilance is also required in drinking tap water. Increased risk for gastroenteritis was significantly associated with drinking tap water (Braeye 2015). In 2014,more than 200 people from 20 villages in Iligan have been hospitalized due to diarrhea and, reportedly caused by contaminated water due to a busted pipeline. (Philstar 2014)An effective alternative is boiling the drinking water. It can deactivate or kill all classes of pathogens in water including chemical-resistant protozoan cysts, bacterial spores, even small viruses that cannot be removed by microfiltration (Block 2001). The research findings of (Feachem et al 1983) showed that 55 C of heated water can inactivate or kill waterborne organism like most pathogenic bacteria, protozoa and helminthes.

The data presented here about water quality was supported by results of the study of Rosenberg 2003 which showed that poor quality control of drinking water is increasing. This is also consistent with the recent data of Farhadkhani (2014) which reported deterioration of drinking water quality. These findings highlight the importance of strict and continuous surveillance of tap and bottled water system to produce drinking water compliant to the standard parameters.

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