PREVALENCE OF *ESCHERICHIA COLI* IN SOME PUBLIC WATER SOURCES IN GUSAU MUNICIPAL, NORTH -WESTERN NIGERIA

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ABSTRACT
This study investigated the presence of Escherichia coli from some public water sources in Gusau municipal, north- western Nigeria. This was done by determining the total coliform counts and the presence of Escherichia coli and its antibiotic susceptibility profile. A total of 180 well 60 tap and 60 packaged water samples were obtained from Gusau municipal on weekly basis over a period of seven month (August, 2006-Feb, 2007) covering part of rainy and dry seasons. Standard procedures were used for the identification of Escherichia coli and for estimating total coliform counts. All samples had coliform counts higher than the international standard recommended by the World Health Organization. Results of biochemical analysis of the samples showed that out of 63 confirmed Escherichia coli isolated, 41 (45.5%) were from well water, tap water had 14 (23.3%) while packaged water had 8(13.3%).The susceptibility profile of the isolates to nine antimicrobial agents indicated that majority of the isolates were highly susceptible to Chloramphenicol, Gentamycin, Perrfloxacin, Tarivid, Augmentin, Streptomycin, Sparfloxacin, and Ciprofloxacin, moderate susceptibility to Septrin and resistant to Amoxacilin were observed. None of the water samples met the WHO standards for drinking water and thus pose a serious health risk to its consumers and users if not properly treated.

Key words: Bacteriological quality, Escherichia coli, Prevalence, susceptibility profile, public water sources, Gusau.

INTRODUCTION
Water is the life wire of the body and infact it is the basis of life, it is a critical part of human diet, however many water sources in developing countries are unhealthy because they contain harmful physical, chemical and biological agents.

The World Health Organization (WHO) estimates that up to 80% of ill health in developing countries are water and sanitation related (Cheesbrough, 2000). High incidence of childhood diarrhea, helminthiasis, tracoma and the overall high mortality rates are associated with poor environmental sanitation (Admassu *et al.*, 2004). The contamination of water remains a problem of global concern contributing to high morbidity and mortality rates from water and food borne diseases (Olukosi *et al.*, 2008).

Escherichia coli is a common member of the intestinal microflora of both humans and warm blooded animals. It is a commensal or opportunistic pathogen implicated in acute urinary tract infections (U. T.I) and gastrointestinal tract infections (Swie-cicka *et al.*, 2003). It is a consistent and predominant facultative inhabitant of the human gastro-intestinal tract, thus its regular presence in the intestine and faeces of warm blooded animals makes the bacterium an indicator of faecal pollution (Yang *et al.*, 2004). The presence of faecal coliforms in water indicates contaminant per se, and may indicate that the sample is unsuitable for consumption, (Szakal *et al.*, 2001).

Ahmed *et al.*, (2005) reported that the use of indicator bacteria such as faecal coliforms fc and *Escherichia coli* for the assessment of faecal pollution and possible water quality deterioration in various water sources is widely accepted concept in the world. Strain of *Escherichia coli* in particular constitute an important set of water borne pathogens composed of numerous shiga toxin producing species and enterohaemorrhagic strains associated with portable and recreational water (Maynard *et al.*, 2005). Since indictor bacteria such as *Escherichia coli* are present in high abundance in the gastro intestinal tracts of most warm blooded animals. Then detection of those organism implies presence of faecal contamination (Bower *et al.*, 2005). Byamukama *et al.*, (2000) reported that indicator bacteria such as faecal coliforms and *Escherichia coli* have been used for the assessment of faecal pollution and possible water quality deterioration in various water sources.

Water contamination events often results from discharges from waste water treatment facilities, overflowing sanitary sewer systems, waste materials that find their way into domestic and industrial sewage and run off of animal faecal matters during storm events (Maynard *et al.*, 2005). Exposure to contaminated water through ingestion such as drinking water, recreation or irrigation is a significant mode of transmission of gastro intestinal tract infection (Umoh *et al.*, 2006).

The role of food and water as vehicle for the transmission of this organism is well documented and represent a significant public health threat (Szakal *et al.*, 2001). This study investigates the microbiological quality of water sources in Gusau municipal.

MATERIALS AND METHODS
Study area
 Gusau, is a city in northern Nigeria, in Zamfara State. It is located on the Sokoto River in the savanna region of Nigeria. The river provides access to water supplies during the dry season. It serves as a major industrial center of northern Nigeria. Industries in the city include textile manufacturing, groundnut and tobacco processing, and cotton ginning. The city is active in mining the deposits of gold and diamonds in the surrounding countryside. Gusau is connected by roads and a railroad to other cities in the region; the city also has a regional airport. The city is part of the Hausa-Fulani cultural region of northern Nigeria. It has a substantial Muslim population and contains numerous mosques and Muslim organizations. Archaeological evidence suggests that Gusau was occupied by Old Stone Age (37,000-15,000 years ago) peoples and many quartz tools from this period have been found in the area. Gusau has a Population of (1995) 154,000 people **(**Microsoft ® Encarta ® 2009). (Encyclopædia Britannica 2009).

Samples collection
Samples from well water was collected in a sterile 250ml capacity bottle using "Guga" The bottles were filled with the water and capped carefully, labeled with sample code number and transported to the laboratory in ice cold box and analyzed within 6hrs. In the case of tap water, samples were collected in 1 liter capacity containers, labelled appropriately, coded and transported to the laboratory in ice- pack box for analysis. Packaged water samples were purchased from the sale points, placed in an insulated cold box, transported to the laboratory and analyzed within 6hrs (Cheesbrough, 2000).

Bacteriological analysis of the water samples
Enumeration of coliforms
Ten-fold serial dilutions of water samples were prepared using sterile distilled water as a diluents. From each dilution, 0.1ml was spread aseptically onto duplicate plates of Eosin methylene blue agar (Oxoid) following standard procedure, the plates were incubated (Gallen kamp England model/ IH-150) at 37^0 C for 24hrs. Typical lactose fermenting colonies 0.1-0.5mm pinkish coloured were counted and reported as cfu/ml and the value multiplied by the dilution factor to get the correct cfu/ml.

Isolation of *Escherichia coli*
Escherichia coli from various samples were isolated by pre enrichment using trypticase soy broth and high temperature methods of incubation described by LeJeune *et al.*, (2001), in which twenty 20ml of each water sample was combined with equal volume of double strength trypticase soya broth in a container and incubated for 24hrs. After the enrichment a loopful of the broth was streaked on to eosin methylene blue (EMB) agar plates (Oxoid) incubated for 24hrs. Mixed cultures were transferred on to fresh EMB to obtain pure colonies (shiny green small with dark centres which were later transferred on to nutrient agar slants for further test. (Claus,1992).

Biochemical Characterization of the Isolates
This involved carrying out the following tests: indole production, methyl red, voges-praskauer test, citrate utilization, motility test, urease production and kligler iron agar test. Isolates that gave the typical IMVic reaction and gave A/A reaction on KIA were Isolated and stored on nutrient agar slant. Gram stain reaction was also carried out to determine the Gram's behavior of the isolates.

Antibiotic sensitivity
Each of the isolates (*Escherichia coli*) were subjected to antibiotic susceptibility testing using the bauer-kirby method that has been standardized by NCCLS (WHO 2003) and evaluated by the methods of national committee for clinical laboratory standards 2002. Isolates grown overnight on Nutrient Agar were suspended in sterile normal Saline (0.9% w/v Nacl). Using a sterile wire loop until the turbidity was equivalent to 0.5 Mcfarland standard. A sterile non toxic cotton swabs dipped into the standardized innocula were used to streak the entire surface of Mueller Hinton Agar plates. Antibiotics disks were aseptically placed using a sterile forceps, and all plates incubated (Gallenkamp England model IH-150) at 37^oC for 24hrs (Mills –Robertson *et al.,* 2003).
The result was interpreted using NCCLS, (2002). guide lines, each isolate was classified as;
1. Sensitive if the observed zone of inhibition diameter was equal or greater than NCCLS sensitive diameter (mm)
2. Resistant if the observed zone of inhibition diameter was less than or equal to the NCCL resistant diameter (mm) or
3. Intermediate if the observed zone of inhibition diameter fell within the intermediate range between the NCCLS resistant and sensitive limits.

Data analysis
Data obtained was analysed using frequency, and two way ANOVA

RESULTS
none of the water samples had count less than 2 cfu/ml, thus, the limits of the WHO drinking guidelines was surpassed, well water had the highest coliform count in the dry seasons than during the wet season, in contrast to tap and packaged water that had high coliform count during the wet seasons.
However result of two ways ANOVA revealed that there was significant difference (P < 0.05) between the mean total coliform counts for the water samples analysed, but no significant variation was observed across the seasons as seen in Table 1.
Table 2 shows the frequency of isolation of *Escherichia coli* in the water samples during wet and dry seasons. It was observed that the isolates were more prevalent in well water and tap water sources during the wet seasons. Out of the 41 isolates from well water, 25 were isolated in wet season while 16 were isolated during the dry season, and of the 14 isolates from tap water, 08 were isolated during wet season while 06 were isolated in dry season, and out of 08 *Escherichia coli* isolated in package water 05 and 03 were isolated in wet and dry season respectively.

The antibiotics susceptibility profile of the isolates showed that, the highest resistance was shown to Amoxacilin with 30 isolates representing 50% of the isolates resistance to the antibiotic. The highest susceptibility was shown to gentamycin and perfloxacin with 100% (60) and 91.6% (55) isolates susceptible to these antibiotics respectively, similarly, 88.3% (53), 83.3% (50), 80% (48), 76.6% (46) and 75% (45) susceptibility was recorded for Chloramphenicaol, Tarivid, Augmentin, Streptomycin and Sparfloxacin respectively. Moderate susceptibility was shown to septrin with 35% (21) isolates sensitive to it as presented in Table 3.

Table 1: Mean seasonal coliform counts from different water sources.

Water Sources	Season	
	WET (Mean± S.E)	DRY (Mean ± S.E)
Well n=30	$9.5 \times 10^5 \pm 1.4 \times 10^4$	$1.4 \times 10^6 \pm 4.2 \times 10^5$
Tap n=10	$8.6 \times 10^3 \pm 5.2 \times 10^2$	$3.3 \times 10^1 \pm 2.4 \times 10^1$
Packaged n=10	$5.6 \times 10^3 \pm 1.7 \times 10^3$	$2.6 \times 10^3 \pm 5.4 \times 10^2$

n=location of water source , S.E= standard error

Table 2: Frequency of isolation of *Escherichia coli* in wet and dry seasons

Water sources	No. of samples analyzed	Wet	Dry	Total
Well	180	25	16	41
Tap	60	08	06	14
Packaged	60	05	03	08

Table 3: Antibiotic Susceptibilities of *Escherichia coli* isolates.

S/N	Antibiotics	Disc conc. [µg]	Susceptibility Number [%] Resistant	number [%] Intermediate	number [%] Susceptible
1.	Septrin	30	10 [16.6]	21 [35]	29 [48.3]
2.	Chloramp	30	7 [11.6]	0 [0]	53 [88.3]
3.	Sparfloxacin	10	12 [20]	3 [5]	45 [75]
4.	Ciprofloxacin	10	17 [28.3]	3 [5]	40 [66.6]
5.	Amoxicillin	30	30 [50]	5 [8.3]	25 [41.6]
6.	Augmentin	30	12 [20]	0 [0]	48 [80]
7.	Gentamicin	10	0 [0]	0 [0]	60 [100]
8.	Perfloxacin	30	0 [0]	5 [8.3]	55 [91.6]
9.	Tarivid	10	8 [13.3]	2 [3.3]	50 [83.3]
10	Sterptomycin	30	14 [23.3]	0 [0]	46 [76.6]

DISCUSSION

Several studies have reported that surface and groundwater contamination by faecal pathogens generally occur through surface run-off, leaching and direct faecal deposition into the water bodies via several livestock production activities like confined animal feedlot, free range system, abattoir wastes, land spreading of manure e.t.c (Kuczynska *et al.*,2005;Timpe and Fulhage,1995).The contamination of water bodies/source is majorly from either a point source which is a single identifiable source of pollution or a non-point source which is the major threat to ground water pollution (Bouwer *et al.*, 1999). such contamination brings the threat of infection for people who use the water for drinking, bathing or watering fruits and vegetable. In Nigeria some of the major sources of water are open and shallow wells, streams and even ponds. Also animals/livestock are reared within compounds and households of owner and are allowed to roam freely in search for food hence they consequently serve as sources of faecal contamination of water sources. (Tabukum *et al.,* 1996).

Coliform bacteria have been widely used as indicator of the microbiological quality of surface and ground waters (Ahmed *et al*, 2005). thus the presence of coliforms is an index of bacteriological quality of water, the isolation of coliforms especially *Escherichia coli,* from the water sources is attributable to contamination by material of human and animal origin and this is of health significance as these organism have generally been reported as causative agent of gastroenteritis in humans. From the coliform count results, none of the water simples met the WHO standard for drinking water which states that the coliform count in drinking water both piped and unpiped should be zero/100ml. All the water sampled had very high counts and this indicates that they are unfit for human consumption. Equally the high coliform count observed is indicative of the likely presence of other pathogenic organism in the water sample analyzed. Result of two way ANOVA revealed significant difference between the water source analysed, (P= 0.0386) no significant difference was observed across the seasons (P = O.4370) the significant difference observed between wall tap and packaged water may be ascribed to the fact that tap water are supposed to be treated before use in contrast to well water that is highly exposed to contamination, the lack of significant difference across the season may be due to similarity in geographical condition and sources of contamination. However, the mean coliform counts of the water samples were high in each case and this represents a risk in the consumption of the water (Table 2).

The frequency of isolation of *Escherichia coli* was high during the wet season because of pollution of the well waters which is in agreement with the findings of Kisteman *et al*. (2002) who demonstrated that substantial shares of the total microbial load in water courses and drinking water reservoirs in Germany resulted from rainfall and extreme events.

The high total *Escherichia coli* count in the wet season is in agreement with a similar study by Lejeune *et al*. (2001) who reported higher *Escherichia coli* counts during the summer, high counts in spring and least in winters meant for livestock drinking. In the same manner, this organism has been isolated in tap and packaged water, which is also attributable to contamination of the water.

The tap water, which ought to be treated must have been contaminated as a result of non treatment or poor water treatment or as a result of leaky water pipes, which are buried in municipal drainage systems thus getting contaminated. Conversely, the packaged water might have been contaminated during packaging, these bacteria represent potential threat to human Health causing common diseases such as acute gastroenteritis, urinary tract infections and nosocomial infections. The effects arising from contact with such water vary depending on the volume of the water ingested by an individual and the individual's immune status, with the children and elderly being the most susceptible (Mazari-hiriart *et al*., 2005).

Conclusion/ Recommendations

The water used for public consumption is unsafe for drinking, because none of the waters sampled met the international standards for drinking water. The presence of indicator bacteria in the waters calls for reassessment of water treatment methods particularly for tap and packaged water. As for well water, the siting of all the wells, their construction

and condition was poor, thus there is the need for construction of the wells far away from municipal sewage and drainage systems, provision of protective covering can also minimize the possibility of contamination especially during rainfall, and storm events.There is the need to develop cheap and effective small-scale water disinfection methods for making well water safe for drinking, thus the appropriateness of water storage, solar water disinfection, boiling and chlorination as points of use (small-scale) methods for making well water safe for drinking.

Hand washing before processing and packaging water particularly of packaged water can lead to significant reduction in water related gastroenteritis which is in agreement with the findings that hand writing with soap can lead to a marked reduction in diarrheao without improving water quality even among malnourished children who are at increased risk of death from diarrheao. However access to safe water remains a prerequisite for maintaining hygiene and to further reducing diarrheao disease, 88% of which is attributed to unsafe water supply, inadequate sanitation and hygiene.

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