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Pitout JDD, Church DL, Gregson DB, Chow BL, McCracken M, Mulvey M, Laupland KB (2007). Molecular epidemiology of CTXM-producing *Escherichia coli* in the Calgary Health Region: emergence of CTX-M-15-producing isolates. *Antimicrob. Agents Chemother.* 51: 1281-1286.

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*Full Length Research Paper*

# A prospective study of three blood-borne viral pathogens among pregnant women attending ante-natal care in Owerri, Nigeria

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A study was conducted between the months of March and August, 2010 to ascertain the rate of mono- and co-infection of three blood-borne viral pathogens namely Human immunodeficiency virus (HIV), Hepatitis C virus (HCV), and Hepatitis B virus (HCV) among 500 pregnant women aged between 21 and 40, attending antenatal clinic at a tertiary health centre in Owerri. The screening of blood samples from the sample population was carried out using immunochromatographic rapid test kits including HIV1/2 test kit (Hi Tech Diagnostic Limited, Nigeria) and Bio Tracer™ HBV and HCV Rapid card test kits (Bio Focus Limited, South Korea). Results revealed that all age groups sampled were positive for HIV, producing a total of 115 (23%) with the age group of 29 to 32 producing the highest infection rate of 32 (6.4%) while the least came from the age groups of 21 to 24 and 37 to 40, with 13 (2.6%) each. The result further revealed that only one case of HBsAg infection was recorded among the sample population within the age group of 37 to 40 with 1 (0.2%). There was no case of HCV recorded among the pregnant women neither was there a co-infection involving HIV and any of the hepatitis viruses. The monthly distribution of the viral pathogens revealed that the highest rate of HIV infection occurred in the month of March with 26 (5.2%), while the least occurred in the month of July with 13 (2.6%). The only HBV infection observed in the sample population was recorded in the month of April while none of the months under review recorded any case of HCV, HIV + HBV, or HIV + HCV coinfections among the studied pregnant women. Though no case of coinfection was recorded in this study, concurrent infection between HIV and the hepatitis viruses is a growing public health concern and enough awareness should be created to check its emergence in this part of Nigeria while the campaign against HIV monoinfection should be intensified to check its spread among the uninfected population.

**Key words:** Coinfection, pregnant women, monoinfection, Owerri, antenatal, hepatitis, human immunodeficiency virus (HIV).

## INTRODUCTION

Improved HIV treatment using the highly active antiretroviral therapy (HAART) has indeed reduced mortality of AIDS patients arising from opportunistic infections. Liver failure occasioned by chronic viral hepatitis has however, emerged as a continued threat, currently accounting for a high rate of hospitalization and death

among people living with HIV/AIDS (De Luca, 2002; Salmon-Ceron et al., 2005). Due to geographic patterns of disease and similar patterns of transmission, notably through intravenous drug abuse, blood transfusion and sexual activity, there are growing cases of coinfection of HIV with hepatitis viruses especially, hepatitis B virus (HBV) and hepatitis C virus (HCV). It is therefore not uncommon to observe coinfection involving HIV and HBV, HIV and HCV, or even all three viruses in one patient alone (Nakwagala and Kagimu, 2002; Gupta and

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Singh, 2006).

While the effect of these hepatitis viruses on HIV disease progression has remained controversial, studies have revealed that the viruses may actually increase the rate of progression to AIDS and AID-related death, impair immune reconstitution, elevate the risk of hepatotoxicity from HAART therapy and lower overall level of CD4 count (CDC, 2001), all symptoms which have been observed to account for a reduced rate of survival for coinfecting patients than for those with HIV monoinfection (Agwale et al., 2004). On the other hand, HIV infection may lead to more aggressive HBV or HCV infection and much more rapid progression to cirrhosis and end stage liver disease (De Luca, 2002; Clifford, 2008).

Coinfection of HIV with HCV is however more common than with HBV and more serious in terms of morbidity (Sherman, 2002), accounting for rates as high as 60 to 90% among intravenous drug users and persons with history of multiple blood transfusion, worldwide (Moshen et al., 2002). Owing to the grave consequences of coinfection involving HIV and the hepatitis viruses and its growing public health importance, it has become absolutely necessary to conduct a study in Owerri, South Eastern, Nigerian, to establish the presence or absence of each of the viruses alone or in combination, using pregnant women who visit a tertiary hospital in Owerri metropolis on clinic days, for their regular antenatal care.

## MATERIALS AND METHODS

### Study population

The study population included 500 pregnant women selected at random and aged between 21 and 40, who attended antenatal clinic at a tertiary health center in Owerri, South Eastern Nigeria. The purpose of the study was fully explained to them as well as the hospital management and their informed consent obtained in accordance with WHO (TDR, 2002). The study lasted for six months starting from the month of March and ending in August, 2010.

### Collection and screening of samples

5 ml blood samples were collected from the pregnant women with the aid of separate syringes and transferred to EDTA bottles, labelled and taken immediately to the laboratory for screening using immunochromatographic rapid test kits including HIV 1/2 test kit (Hi-Tech Diagnostic Limited, Nigeria), Bio Tracer™ HBV rapid card test kit (Bio Focus Limited, South Korea). They were all screened according to manufacturer's instructions. Briefly, the blood samples were transferred into sterile centrifuge tubes and centrifuged at 1,500 rpm for 15 min. The serum produced was decanted into sterile tubes from where 10 µl was transferred into the sample well using 10 µl Eppendorf pipette (Micropipettes and pipette tips). This was followed by addition of 40 to 60 µl of assay diluents. The mixture was allowed to stand on the bench for 20 min before the results were read. In HIV result, appearance of a red band only in the control reaction zone (C) indicated a negative result while in a positive case, in addition to the red band in the control reaction zone (C), another red band appeared in the test reaction zone (1 and/or 2). Similarly for HBV and HCV, the presence of one red band in the control reaction zone (C) was indicative of a negative

result while the presence of two red bands ('T' band and C 'band') within the result window indicated a positive result. For coinfection, the blood sample screened for one viral antibody was also screened for another.

## RESULTS

Table 1 represents the age distribution of the pregnant women while Table 2 represents the monthly distribution of the viral pathogens among the pregnant women. As revealed in Table 1, a total number of 115 (23%) of the pregnant women in all the age groups studied were HIV positive with the age group of 29 to 32 producing the highest number, 32 (6.4%) of cases, followed by the age group of 25 to 28 with 30 (6%), while the least was from the age groups of 21 to 24 and 37 to 40 with equal number, 13 (2.6%) of cases, each. As revealed in Table 2, the highest prevalence of HIV infection occurred in the month of March with 26 (5.2%) cases, followed by May with 24 (4.8%) while the least level of prevalence was observed in July with 13 (2.6%). The only case of HBsAg observed was recorded in the month of April with 1 (0.2%), while no month recorded any case of HCV, HIV + HBV or HIV + HCV coinfections.

## DISCUSSION

The highest rate of HIV case among these women considered to be in the prime of their reproductive and productive years (Boysen, 2003) is indeed a veritable cause for concern, but however, confirms earlier findings that HIV/AIDS in Owerri, like in other places in the world (UNAIDS, 2006) and other states in Nigeria (Obi et al., 2007) affects younger people in the prime of their life. One reason that could be adduced for this high level of prevalence could be failure to observe fidelity among married couple, a reason, Amadi et al. (2009) also advanced in a similar study on HIV prevalence in Abia State, Nigeria.

The 1 (0.2%) cases for HBsAg and none for HCV recorded in Owerri is quite commendable. It is also noteworthy to applaud the absence of HIV + HBV and HIV + HCV coinfections among the pregnant women observed in the study in Owerri especially when compared with other published results which recorded high prevalence of coinfection between HIV and the hepatitis viruses (Agwale et al., 2004; Dodig and Tavil, 2001; Jiang-Rong et al., 2007; Anuj and Arora, 2008). It could be inferred that the near absence of HBV, the complete absence of HCV, HIV + HBV and HIV + HCV coinfections were achieved probably because the pregnant women were not involved in intravenous drug abuse, had no drug addict sexual partners who could have sexually transmitted the virus to them or received blood not screened for HBV or HCV, the major routes of transmission of the hepatitis viruses (Jian-Rong et al., 2007). However in the case of HIV which is predominantly sexually transmitted

**Table 1.** Age distribution of pregnant women in a viral coinfection study in Owerri.

Age interval	Frequency	No. (%)									
		HIV		HBV		HCV		HIV+HBV		HIV+HCV	
		+ve	-ve	+ve	-ve	+ve	-ve	+ve	-ve	+ve	-ve
21-24	61(12.2)	13(2.6)	48(9.6)	-	61(12.2)	-	61(12.2)	-	61(12.2)	-	61(12.2)
25-28	111(22.2)	30(6)	81(16.2)	-	111(22.2)	-	111(22.2)	-	111(22.2)	-	111(22.2)
29-32	143(28.6)	32(6.4)	111(12.2)	-	143(28.6)	-	143(28.6)	-	143(28.6)	-	143(28.6)
33-36	90(18)	27(5.4)	63(12.6)	-	90(18)	-	90(18)	-	90(18)	-	90(18)
37-40	95 (19)	13(2.6)	82(16.4)	1	94(18.8)	-	95(19)	-	95(19)	-	95(19)
Total	500(100)	115(23)	385(67)	1(0.2)	499(99.8)	-	500(100)	-	500(100)	-	500(100)

Figures in parenthesis represent percentages. The table also shows that only 1(0.2%) pregnant woman in the age group of 37-40 was positive for HBsAg, whereas no age group produced any positive cases for HCV or co-infection of HIV with HBV or HIV with HCV.

**Table 2.** Monthly distribution of the viral pathogens among pregnant women in a viral coinfection study in Owerri.

Age interval	Frequency	No. (%)									
		HIV		HBV		HCV		HIV+HBV		HIV+HCV	
		+ve	-ve	+ve	-ve	+ve	-ve	+ve	-ve	+ve	-ve
Mar	75(15)	26(5.2)	49(9.8)	-	75(15)	-	75(15)	-	75(15)	-	75(15)
Apr	85(17)	20(4)	65(13)	1(0.2)	84(16.8)	-	85(17)	-	85(17)	-	85(17)
May	80(16)	24(4.8)	56(11.2)	-	80(16)	-	80(16)	-	80(16)	-	80(16)
Jun	90(18)	15(3)	75(15)	-	90(18)	-	90(18)	-	90(18)	-	90(18)
Jul	90(19)	13(2.6)	82(16.4)	1	94(18.8)	-	95(19)	-	95(19)	-	95(19)
Aug	80(16)	17(3.4)	63(12.6)	-	80(16)	-	80(16)	-	80(16)	-	80(16)
Total	500(100)	115(23)	385(67)	1(0.2)	499(99.8)	-	500(100)	-	500(100)	-	500(100)

Figures in parenthesis represent percentages.

(Dieterich, 2000), the comparatively high percentage of occurrence over the months confirms reports (WHO, 1987) that 90% of global HIV cases are transmitted through sexual intercourse with infected partners.

No specific reason could be given for the high or

low prevalence of HIV infection recorded in the months of March with 26 (5.2%) and July with 13 (2.6%), respectively, and only one case of HBsAg recorded in the month of April with 1 (0.2%) as observed in Table 2, since no particular month has been established to be associated with high

level of sexual promiscuity that could predispose to the sexually transmitted diseases (STDs). However, it could be assumed that the month with the least prevalence was one in which calls for, use of protective devices such as condoms during sexual intercourse and abstinence from

indiscriminate sex, were heeded.

As a result of the prevalence of HIV positive cases among pregnant women observed in Owerri, it has become compelling to call upon the Imo State Government and other health care providers in the state to renew and intensify the campaign on mutual fidelity among married couples. Equally to be intensified is the campaign on the need for intending couples and youths to abstain from pre-marital sex. The Federal and State governments should endeavour to continue to make available anti-retroviral drugs and continue to freely distribute them to HIV infected patients. Since no observation was made for HCV and only one case observed for HBsAg, it could be concluded that none of the pregnant women received un-screened blood, indulged in, or had partners who indulged in drug abuse, the major routes of infection for the two hepatitis viruses. This good moral behaviour should be sustained and encouraged. Similarly, efforts aimed at screening every blood or products prior to its transfusion should be sustained. Finally, while cases of HCV could be said to exist in other States in Nigeria like Edo (Mutimer et al., 1994), Lagos (Halim and Ajayi, 2000) and Oyo (Aliyu, 1996), the same could not be said of Imo as this study has revealed.

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*Full Length Research Paper*

# Incidence of *Staphylococcus aureus*, coliforms and antibiotic resistant strains of *Escherichia coli* in rural water supplies in Calabar South Local Government Area

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An investigation on the incidence of *Staphylococcus aureus*, coliforms and antibiotic resistant *Escherichia coli* strains in both treated and untreated rural water supplies was carried out in Calabar South Local Government Area of Cross River State, Nigeria. Analysis revealed significant differences between the different water sources, locations and the months of sampling, with the stream and well water showing higher bacterial contamination compared to the tap water source ( $P < 0.05$ ). The isolation of *S. aureus*, *Bacillus* species, *Pseudomonas aeruginosa* and other bacterial pathogens present enough evidence that water from these sources are unfit for human consumption and constitute significant public health implications except subjected to further treatment. High percentages of the *E. coli* strains isolated from the water sources showed multiple resistances to most of the antibiotics commonly used by humans. Strains recovered from the stream and well water sources were most resistant and showed significantly higher minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) ( $P < 0.05$ ) than those from the tap water source. The results of this investigation therefore revealed that the bacteriological quality of both the treated tap and untreated well and stream water sources failed to meet the standards for drinking water.

**Key words:** Incidence, *Staphylococcus aureus*, coliforms, *Escherichia coli*, antibiotic resistance, water supplies.

## INTRODUCTION

The illnesses resulting from consuming faecal contaminated water are mostly treated with antibiotics, but unfortunately, there has been recent development of antimicrobial resistance by many strains of microorganisms which is now making it difficult to treat some infectious diseases (Inyang, 2009). It has long been established, that multiple drug resistances could be transferred among members of the Enterobacteriaceae, especially *Escherichia coli*; many strains of *E. coli* carry resistance factors (R-factors or plasmids) which confer resistance to antibiotics and can be transmitted among themselves and to other bacteria (Oyedeji et al., 2011). The ability of environmental bacteria to transfer antibiotic resistance genes to human pathogens can have grave

consequence for human health, most especially the children and the immunocompromised individuals who are more vulnerable to bacterial illnesses (Oyedeji et al., 2011), including limiting the number of drugs available for treatment of diseases leading to fewer treatment options for the sick (Oyedeji et al., 2011). This study therefore was designed to evaluate the incidence of *Staphylococcus aureus*, coliforms and antibiotic-resistant strains of *E. coli* in rural water supplies in Calabar South Local Government Area of Cross River State.

## MATERIALS AND METHODS

### Study areas

The study sites were rural communities randomly selected within Calabar South Local Government Area located between 4°57'N latitude and 8°19'E and covering an area of 264 km<sup>2</sup>, with the

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**Table 1.** Description of the rural water samples collected from different sources at different locations from Calabar South Local Government Area.

S/N	Location	No. of samples	Source of samples
1	Ekpo Abasi Street	30	Private tap water (treated)
2	New Airport/Jeb Street	30	Private tap water (treated)
3	Palace Road/Anantigha Street	30	Private tap water (treated)
4	Rev. Mbukpa Close	30	Well water (majority uncovered)
5	Creek Road/Ekpenyong Abasi Street	30	Well water (uncovered)
6	Jeb's area	30	Stream water
7	Anantigha abattoir water	30	Stream water (polluted with faecal materials from the slaughter)
8	Anantigha phase 2	30	Stream water

population size of 191,630 (NPC, 2006). The area is surrounded by lots of rural communities whose inhabitants engage mainly in farming and trading activities.

### Sample sources and collection

The main water sources in the rural communities were identified and sampled according to the methods described by Adejuwon et al. (2011) and Oyedeji et al. (2011). A total of 240 water samples comprising of 90 tap water samples from three locations, 60 well water samples from two locations, and 90 stream water samples from three locations were collected between the months of June to October, 2011 (Table 1). Samples from streams were collected at six different points where the communities fetch their water, thereby making direct contact with the water, while those from wells and taps were collected from six different wells and taps for each location.

### Enumeration techniques

Total heterotrophic bacteria count was prepared on standard plate count agar (Biotech Lab Ltd, UK) using pour plating technique (Oyedeji et al., 2011). Enumeration of total and faecal coliforms, *S. aureus* and *Streptococcus faecalis* were made on MacConkey agar (Biotech Lab Ltd., UK), mFC agar (Biotech Lab Ltd., UK), *S. aureus* M110 agar (Hardy Diagnostics, USA), and bile esculine agar (Biotech Lab Ltd., UK), respectively using the standard membrane filtration technique (Ojo et al., 2005; Mihdhir, 2009; Oyedeji et al., 2011). Plates were incubated at 35°C for 24, except the faecal coliform agar that was incubated at 44.5°C and thereafter, characteristic colonies indicative of these organisms were counted and expressed as colony forming unit per 100 ml of water samples.

Pure bacterial isolates were characterized and identified by standard methods (Cheesebrough, 2002; Prescott et al., 2002). Biochemical tests such as catalase, coagulase, citrate utilization, indole, methyl red, Voges-Proskauer, motility, ornithin decarboxylase production, oxidase, sugar fermentation (glucose, sucrose and lactose), gas, and H<sub>2</sub>S production on triple sugar agar (TSI) tests were employed.

### Antibiotic sensitivity screening of *E. coli*

Antibiotic sensitivity screening was carried out using multi-disc (Maxicare Lab., Nigeria) diffusion method as described by Akinyemi et al. (2005), Oyetao et al. (2007) and Duru and Mbata (2010). Precisely, 0.1 ml of the prepared *E. coli* strains in nutrient broth were poured onto the surface of dried Mueller-Hinton (MH) agar

plate spread using swab stick and allowed to dry for about 30 min at room temperature before placing the multi-disc antibiotics on the culture plates using sterile forceps. Plates were left at room temperature on the bench for 15 min to allow for diffusion of the antibiotics before incubation at 35°C for 18 to 24 h. Results were recorded by measuring the zones of inhibition and strains were recorded as resistant if the zone of inhibition was ≤ 10 mm wide around the disc, as intermediate if the zone of inhibition was ≤ 16 mm, and as sensitive if there was a clear zone of inhibition ≥ 17 mm surrounding the disc (CLSI, 2003). However, intermediate strains were considered resistant. Gram negative discs, such as ampicillin (30 µg), augmentin (30 µg), ceporex (10 µg), gentamycin (10 µg), ciprofloxacin (10 µg), nalidixic acid (30 µg), tarivid (10 µg), perflaxin (10 µg), streptomycin (30 µg), and septrin (30 µg) were used.

### Determination of minimum inhibitory and bactericidal concentration (MIC and MBC)

Determination of MIC and MBC was carried out using broth dilution method as described by Akinyemi et al. (2005) and Duru and Mbata (2010). A two-fold serial dilutions of the antimicrobial agents was done in series in test tubes to obtained different concentrations of 0.05, 0.10, 0.19, 0.39, 0.78, 1.56, 3.12, 6.25, 12.5, 25, 50.0, 100, 200, and 400 mg/ml for each of the antibiotics. After the different concentrations were obtained, sterile pipettes were used to deliver 0.2 ml of the 24 h nutrient broth cultures of the *E. coli* into each tube and were incubated at 35°C for 18 to 24 h. The least concentrations of the antibiotics that resulted in complete inhibition of the test bacteria after incubation were recorded as the MIC using turbidity as index, while the least concentrations in the MIC test, of which no growth was observed after sub-culturing a loopful onto freshly prepared nutrient agar were recorded as the MBC.

## RESULTS

### Bacterial count

All the samples collected from tap, wells, and the streams gave total heterotrophic bacterial count, total and faecal coliform counts, *S. aureus* and *S. faecalis* counts. Table 2 shows the mean counts of total heterotrophic bacteria ranged from  $2.7 \pm 4.1 \times 10^2$  cfu/ml (location 1 in June) to  $4.5 \pm 0.9 \times 10^2$  cfu/ml (location 3 in August),  $3.2 \pm 1.2 \times 10^3$  cfu/ml (location 5 in June) to  $4.8 \pm 5.2 \times 10^3$  cfu/ml (location 4 in July) and  $3.3 \pm 2.0 \times 10^4$  cfu/ml (location 7 in August) to  $5.6 \pm 1.6 \times 10^4$  cfu/ml (location 8 in June) for the tap, well and stream water samples, respectively.

The mean total and faecal coliform counts (Table 3) ranged from  $11 \pm 2.8$  cfu/100 ml (location 1 in July) to  $29 \pm 3.9$  cfu/100 ml

**Table 2.** Mean total heterotrophic bacterial counts for the water sources collected between the months June to October.

Month sample	*Sampling sources/Location							
	Tap water			Well water			Stream water	
	1	2	3	4	5	6	7	8
June	2.7 ± 4.11	2.7 ± 3.08	3.3 ± 1.61	3.6 ± 3.25	3.2 ± 1.21	3.7 ± 2.05	4.6 ± 2.89	5.6 ± 1.61
July	2.9 ± 2.75	3.0 ± 4.04	3.6 ± 2.084	4.8 ± 5.18	3.7 ± 5.69	4.0 ± 3.42	5.0 ± 3.85	3.7 ± 2.10
August	3.5 ± 3.25	3.1 ± 3.44	5 ± 0.94	4.5 ± 1.95	3.8 ± 4.76	4.5 ± 2.49	3.3 ± 2.0	3.3 ± 2.89
September	3.3 ± 3.13	3.4 ± 2.13	3.5 ± 3.0	4.4 ± 2.56	3.9 ± 3.68	4.9 ± 1.79	3.6 ± 3.10	4.0 ± 1.57
October	2.9 ± 1.97	3.5 ± 1.77	3.7 ± 1.57	4.1 ± 4.39	3.8 ± 2.31	4.6 ± 2.38	5.2 ± 4.28	4.6 ± 2.31

Data are expressed as mean ± standard error (SE) of triplicate trials. Values with different superscript across each row are significant ( $P < 0.05$ ). \*1-3 = tap water location, 4-5 = well water location, 6 – 8 = stream water location.

**Table 3.** Mean total and faecal coliform bacterial counts for the water sources collected between the months June to October.

Microbial count	Month sample	*Sampling sources/Location							
		Tap water			Well water			Stream water	
		1	2	3	4	5	6	7	8
TCBC (cfu/100 ml)	June	15 ± 3.02	26 ± 2.83	19 ± 3.55	26 ± 4.82	35 ± 2.36	35 ± 2.14	45 ± 3.13	49 ± 4.61
	July	11 ± 2.80	20 ± 4.93	29 ± 3.98	33 ± 4.96	38 ± 3.50	36 ± 2.67	51 ± 5.98	37 ± 2.12
	August	23 ± 3.85	28 ± 5.06	25 ± 1.53	29 ± 4.07	35 ± 2.16	32 ± 3.86	40 ± 5.59	40 ± 3.03
	September	18 ± 5.46	23 ± 2.75	23 ± 2.91	34 ± 1.34	32 ± 2.36	33 ± 3.99	57 ± 4.4	52 ± 2.50
	October	15 ± 3.40	16 ± 2.45	20 ± 2.92	30 ± 1.89	31 ± 3.98	35 ± 5.06	41 ± 4.64	45 ± 1.54
FCBC (cfu/100 ml)	June	6 ± 3.04	16 ± 3.79	11 ± 3.09	15 ± 2.75	19 ± 2.99	21 ± 4.92	29 ± 6.39	28 ± 2.31
	July	10 ± 3.90	13 ± 1.86	13 ± 1.59	21 ± 3.55	22 ± 2.54	27 ± 2.54	35 ± 3.12	26 ± 1.24
	August	13 ± 3.62	15 ± 2.52	18 ± 3.14	15 ± 2.77	20 ± 3.07	23 ± 4.19	28 ± 3.68	20 ± 1.13
	September	11 ± 2.29	7 ± 3.54	17 ± 3.13	24 ± 4.26	21 ± 2.29	20 ± 6.74	24 ± 3.19	28 ± 1.03
	October	9 ± 3.70	11 ± 2.94	15 ± 5.21	21 ± 2.89	22 ± 2.56	20 ± 2.51	36 ± 4.57	27 ± 1.41

Data are expressed as mean ± standard error (SE) of triplicate trials. Values with different superscript across each row are significant ( $P < 0.05$ ). \*1-3 = tap water location, 4-5 = well water location, 6 – 8 = stream water location, TCBC = total coliform bacterial counts, FCBC = faecal coliform bacterial counts.

(location 3 in July) and  $6 \pm 3.0$  cfu/100 ml (location 1 in July) to  $18 \pm 3.1$  cfu/100 ml (location 3 in August), respectively for the tap water samples,  $26 \pm 4.8$  cfu/100 ml (location 4 in June) to  $38 \pm 3.5$  cfu/100 ml (location 5 in July) and  $15 \pm 2.8$  cfu/100 ml (location 4 in June) to  $24 \pm 4.3$  cfu/100 ml (location 4 in September), respectively for the well water samples, and  $32 \pm 3.9$  cfu/100 ml (location 6 in August) to  $57 \pm 4.4$  cfu/100 ml (location 7 in

September) and  $20 \pm 2.5$  cfu/100 ml (location 6 in October) to  $36 \pm 4.6$  cfu/100 ml (location 7 in October), respectively for the stream water samples.

The mean counts of *S. aureus* and *S. faecalis* (Table 4) ranged from  $3 \pm 0.7$  cfu/100 ml (location 1 in August) to  $10 \pm 4.5$  cfu/100ml (location 2 in October) and  $5 \pm 2.7$  cfu/100 ml (location in October) to  $12 \pm 1.5$  cfu/100 ml (location 3 in July), respectively for the tap water samples,

$9 \pm 1.9$  cfu/100 ml (location 1 in August) to  $18 \pm 2.5$  cfu/100 ml (location 1 in July) and  $10 \pm 1.7$  cfu/100 ml (location 1 in June) to  $25 \pm 2.3$  cfu/100 ml (location 2 in July), respectively for the well water samples, and  $11 \pm 2.9$  cfu/100 ml (location 1 in October) to  $26 \pm 1.3$  cfu/100 ml (location 2 in September) and  $10 \pm 2.6$  cfu/100 ml (location 1 in August) to  $28 \pm 2.5$  cfu/100 ml (location 2 in October) for the stream water samples.

**Table 4.** Mean *S. aureus* and *S. faecalis* counts for the water sources collected between the months June to October.

Microbial count	Month sample	*Sampling Sources/Location							
		Tap water			Well water		Stream water		
		1	2	3	4	5	6	7	8
SAC (cfu/100 ml)	June	6 ± 1.21	9 ± 2.54	5 ± 2.08	15 ± 2.08	12 ± 2.49	15 ± 1.64	25 ± 1.63	14 ± 1.01
	July	8 ± 2.13	11 ± 1.38	4 ± 0.81	18 ± 2.54	15 ± 2.08	20 ± 1.37	28 ± 1.67	15 ± 1.81
	August	3 ± 0.68	3 ± 1.34	6 ± 3.21	9 ± 1.86	12 ± 2.21	12 ± 2.92	13 ± 2.92	18 ± 0.54
	September	5 ± 1.95	7 ± 3.44	9 ± 3.84	15 ± 3.21	13 ± 1.82	15 ± 1.80	16 ± 1.25	15 ± 0.05
	October	6 ± 1.77	10 ± 4.49	6 ± 3.84	13 ± 2.74	11 ± 1.38	11 ± 2.96	14 ± 2.96	15 ± 2.10
SFC (cfu/100 ml)	June	6 ± 3.48	6 ± 2.49	8 ± 1.57	10 ± 1.73	13 ± 3.30	13 ± 1.41	27 ± 3.13	26 ± 2.11
	July	8 ± 2.14	7 ± 2.36	12 ± 1.52	13 ± 3.73	25 ± 2.33	16 ± 1.59	25 ± 1.97	18 ± 1.25
	August	10 ± 2.33	8 ± 3.09	6 ± 2.47	21 ± 1.79	12 ± 2.09	10 ± 2.06	25 ± 2.56	24 ± 1.10
	September	6 ± 3.01	5 ± 2.93	7 ± 2.73	13 ± 2.23	24 ± 2.06	13 ± 3.43	21 ± 2.05	23 ± 0.0
	October	5 ± 2.73	8 ± 3.18	5 ± 2.23	23 ± 2.30	15 ± 1.89	13 ± 2.06	28 ± 2.51	24 ± 1.24

Data are expressed as mean ± SE of triplicate trials. Values with different superscript across each row are significant ( $P < 0.05$ ). \*1-3 = tap water location, 4-5 = well water location, 6-8 = stream water location, SAC = *Staphylococcus aureus* count, SFC = *Streptococcus faecalis* counts.

Table 5 presents a summary of the morphological and biochemical characteristics of the bacteria isolated from the rural water samples from the different sources between the months of June to October.

#### Incidence of antibiotic-resistant *E. coli*

The overall resistance observed was most frequently observed to ampicillin, augmentin, ceporex, gentamycin, nalidixic acid, tarivid, and perflaxin (Table 6). Isolates that exhibited resistance to at least three antibiotics were regarded as multiple antibiotic-resistant strains (Table 7). The result shows that 6 (15.4%), 19 (37.3%), and 39 (53.4%) strains from tap, well, and stream water samples, respectively were resistant to three or more antibiotics. Strains isolated from stream and well water samples gave highest MIC and MBC as compared to the tap water samples (Table 8 and 9).

#### DISCUSSION

The results of the investigation revealed that the bacteriological quality of both the treated tap water samples and the untreated well and stream water samples collected from the different locations failed to meet the standard for drinking water, although significant ( $P < 0.05$ ) differences existed between the water sources, with the stream and well water samples consistently showing higher bacterial contamination as compared to the tap water samples. The values were higher than the recommended standard for total heterotrophic bacterial counts in drinking water. Other studies had earlier reported such high bacterial loads in treated and untreated water supplies (Inyang, 2009; Oyedeji et al., 2011).

The presence of coliform in a high proportion of water samples is a good indicator of water contamination. Water meant for human

consumption should be free of coliform (NIS, 2007; WHO, 2007). A high proportion of the rural water samples analysed in this study were positive for total and faecal coliforms. Stream and well water samples showed significantly ( $P < 0.05$ ) higher total and faecal coliforms as compared to the tap water samples. The World Health Organisation (2007) recommends zero counts of faecal coliform bacteria in any 100 ml of drinking water.

The high counts obtained therefore suggest the unsuitability of these water sources for consumption purposes. The high faecal coliform bacteria counts obtained in the stream water samples could be attributed to the faecal materials consistently disposed into the stream from the abattoir house. The differences in the levels of contamination of the well water studied reflect the usually washed before used. In a similar study, Oyedeji et al. (2011) reported that the

**Table 5.** Morphological and biochemical characteristics of isolates.

Isolate No.	Microscopy		Catalase	Coagulase	Citrate	Motility	Indole	Ornithin	MR	VP	Oxidase	Glucose	Lactose	Sucrose	Gas	H <sub>2</sub> S	Probable organism
	Gram's reaction	Shape															
1	-	Short rods	NT	NT	-	+	+	+	+	-	-	+	+	+	+	+	<i>E. coli</i>
2	+	Cocci in clusters	+	+	+	-	-	+	+	-	-	+	-	+	-	-	<i>S. aureus</i>
3	+	Cocci in chains	-	-	+	-	-	+	+	-	-	+	-	-	+	-	<i>S. faecalis</i>
4	-	Short rods	NT	NT	+	+	-	+	+	-	+	-	+	+	+	-	<i>Enterobacter aerogenes</i>
5	-	Short rods	NT	NT	+	-	-	-	-	+	-	+	+	+	+	-	<i>Klebsiella spp.</i>
6	-	Short rods	NT	NT	+	+	-	+	+	-	-	+	+	+	+	+	<i>Salmonella typhi</i>
7	-	Short rods	NT	NT	+	+	+	-	-	+	+	+	-	-	+	-	<i>Pseudomonas aeruginosa</i>
8	-	Short rods	NT	NT	+	+	-	+	-	+	-	+	-	+	+	-	<i>Serratia marcescens</i>
9	-	Short rods	NT	NT	-	+	+	-	+	-	-	+	-	+	+	+	<i>Proteus species</i>
10	-	Short rods	NT	NT	-	-	-	+	+	-	-	+	+	-	+	-	<i>Shigella species</i>
11	+	Long rods	+	NT	+	+	-	-	+	-	-	+	+	-	+	-	<i>Bacillus species</i>
12	-	Short rods	NT	NT	+	+	-	-	+	-	+	+	-	-	-	+	<i>Chromobacterium violaceum</i>
13	-	Short rods	NT	NT	+	+	-	+	+	-	-	+	+	+	+	+	<i>Citrobacter species</i>
14	+	Cocci (singly)	+	-	+	-	-	+	+	-	-	+	-	-	+	-	<i>Micrococcus species</i>

NT = Not tested, MR = Methyl red, VP = Voges-Proskauer, + = Positive test, - = Negative test.

**Table 6.** Frequency and percentage resistance of *E. coli* strains isolated in the rural water sources to test antibiotics.

*Antibiotic (µg/disc)	Tap water (n = 39)		Well water (n = 51)		Stream water (n = 73)	
	Frequency	Percentage	Frequency	Percentage	Frequency	Percentage
AMP (30)	8	20.5	17	33.3	58	79.5
AUG (30)	1	2.6	6	11.8	40	55.0
CEP (10)	6	15.4	16	31.4	35	47.9
CN (10)	7	17.9	16	31.4	31	42.4
CPX (10)	0	0.00	0	0.00	0	0.00
NA (30)	4	10.3	14	27.5	43	59.0
OFX (10)	4	10.3	9	17.7	28	38.4
PEF (10)	4	10.3	15	29.4	25	34.2
S (30)	0	0.00	0	0.00	13	4.0
SXT (30)	0	0.00	0	0.00	0	0.00

\*AMP = Ampicillin, AUG = Augmentin, CEP = Ceporex, CN = Gentamycin, CPX = Ciprofloxacin, NA = Nalidixic acid, OFX = Tarivid, PEF = Perflaxin, S = Streptomycin, SXT = Septrin.

indiscriminate use of buckets for other purposes apart from drawing of water from wells alone could also be a potential source of

contamination as these may have had contact with human faecal matter. They also reported that rain water can also pick harmful bacteria and

other pollutants on the land surface and if this water pools are nearsanitary and hygienic qualities of the locations which they are sited



**Table 7.** Frequency and percentages of multiple-antibiotic resistance among *E. coli* strains.

*Number of antibiotic	Tap water (n = 39)		Well water (n = 51)		Stream water (n = 73)	
	Frequency	Percentage	Frequency	Percentage	Frequency	Percentage
1	9	23.1	8	15.7	25	34.2
2	3	7.7	6	11.8	4	5.5
3	5	12.8	8	15.7	12	16.4
4	1	2.6	8	15.7	14	19.1
5	0	0.00	1	1.9	7	10.0
6	0	0.00	2	3.9	2	3.0
7	0	0.00	0	0.00	4	5.4
% not resistant:	21	53.8	18	35.3	5	7.1
% resistant:	18	46.2	33	64.7	68	93.2
% resistant to $\geq 1$ :	12	30.8	14	27.5	29	40.0
% resistant to $\geq 3$ :	6	15.4	19	37.3	39	53.4

\*No of antibiotics resistant.

**Table 8.** Antibiotic resistance patterns among the *E. coli* strains.

<sup>a</sup> Number of antibiotic	<sup>b</sup> Resistance pattern
1	AMP; AUG; CEP; CN; NA; OFX; PEF
2	AMP/CEP; AMP/PEF; CN/NA; AMP/CN; CN/OFX; NA/CEP
3	AMP/CN/CEP; AMP/CE/PEF; AMP/CN/OFX; CN/NA/CEP; CN/NA/OFX; AUG/CN/OF; AMP/CN/PEF; CN/NA/PEF; AMP/OFX/CEP; AMP/AUG/NA; AMP/NA/CEP
4	AMP/CN/OFX/PEF; AMP/OFX/CEP/PEF; AMP/CN/NA/PEF; AMP/AUG/NA/PEF; AMP/NA/CEP/PEF; AMP/CN/OFX/CEP; AUG/CN/OFX/PEF; CN/NA/CEP/PEF; AMP/NA/OFX/CEP
5	AMP/CN/NA/CEP/PEF; AMP/AUG/OFX/CEP/PEF; AMP/CN/NA/OFX/PEF; AMP/AUG/CN/S/NA
6	AMP/AUG/NA/OFX/CEP/PEF; AMP/AUG/CN/NA/OFX/PEF; AMP/AUG/CN/OFX/CEP/PEF
7	AMP/AUG/CN/NA/OFX/CEP/PEF

<sup>a</sup>No of antibiotics resistant to. <sup>b</sup>Resistance pattern constructed from the antibiogram; antibiotic codes as defined under methodology. AMP = Ampicillin, AUG = Augmentin, CEP = Ceporex, CPX = Ciprofloxacin, OFX = Tarivid, PEF = Perflaxin, S = Streptomycin, SXT = Septrin, CN = Gentamycin, NA = Nalidixic acid.

(Oyedemi et al., 2011). Majority of the well water studied were without protective covers and buckets used in taking water from all the wells from all locations were left carelessly on the ground after fetching water and were not the wells they can seeps down and pose potential health problems to those using the water from the wells.

The high total and faecal coliform bacteria obtained in the treated tap water samples in this destruction, in addition to providing functional study are not surprising and may be a reflection of several factors. It has been reported that coliform can be found both in chlorinated and unchlorinated water and that their total elimination

from water would require the knowledge of their population in such water and determining the quantity of chlorine needed for their complete chlorinators (Inyang, 2009). However, tap water are usually stored in storage devices such as tanks and reservoirs after harvesting and therefore, having unsanitary storage devices is

**Table 9.** MIC and MBC of the different antibiotic against strains of *E. coli* isolated from well water samples.

Antibiotic	Concentration (mg/ml)												MIC range (mg/ml)	MBC range (mg/ml)
	400	200	100	50.0	25.0	12.5	6.25	3.13	1.56	0.78	0.39	0.10		
AMP	-	-	-	$\beta_1$	$\beta_2$	$\beta_3$	$\beta_4$	+	+	+	+	+	6.25 - 50	25 - 100
AUG	-	-	-	$\beta_1$	$\beta_2$	$\beta_3$	+	+	+	+	+	+	12.5 - 50	25 - 100
CEP	-	-	$\beta_1$	$\beta_2$	$\beta_3$	$\beta_4$	+	+	+	+	+	+	12.5 - 100	50 - 200
CPX	-	-	-	$\beta_1$	$\beta_2$	$\beta_3$	+	+	+	+	+	+	6.25 - 50	25 - 100
OFX	-	-	$\beta_1$	$\beta_2$	$\beta_3$	$\beta_4$	+	+	+	+	+	+	12.5 - 100	50 - 200
PEF	-	-	$\beta_1$	$\beta_2$	$\beta_3$	$\beta_4$	+	+	+	+	+	+	12.5 - 100	25 - 200
S	-	-	-	$\beta_1$	$\beta_2$	$\beta_3$	$\beta_4$	$\beta_5$	+	+	+	+	3.13 - 50	12.5 - 100
SXT	-	-	-	+	$\beta_1$	$\beta_2$	$\beta_3$	$\beta_4$	+	+	+	+	3.13 - 50	12.5 - 50

AMP = Ampicillin, AUG = Augmentin, CEP = Ceporex, CPX = Ciprofloxacin, OFX = Tarivid, PEF = Perflaxin, S = Streptomycin, SXT = Septrin.  $\beta$  with different subscripts indicate MICs of each drug against test bacteria (many strains shared the same MIC and MBC). - = No turbidity, + = visible turbidity

known to contribute to substantial reduction in water quality (Welch et al., 2000).

Members of the genus *Staphylococci*, mostly *S. aureus* is considered as an indicator of hygienic status employed in the field of production or distribution of drinking water (Mihdhir, 2009). Majority of the water samples from all the sources were positive for *S. aureus* and *S. faecalis*, with significantly higher counts in the stream water samples, followed by the well water samples than the tap water samples. There are many reasons for potential concern when *S. aureus* is present in drinking water supplies; *S. aureus* is a pathogen and survives longer than coliforms in water (Antai, 1987) and are implicated in waterborne diseases. The high bacterial counts obtained in this study were also recorded by other workers (Fong et al., 2007; Popoola et al., 2007; Mihdhir, 2009; Oyedeji et al., 2011).

The presence of enteric bacterial pathogens in water sources may spell health hazards, such as diarrhoeal diseases, which accounts for a substantial degree of morbidity and mortality in adults and children (Obi et al., 2004). The situation is further complicated if these etiologic

agents are antibiotic resistant strains (Olaoluwa et al., 2010). In this study, high incidence of *E. coli* strains resistant to commonly used antibiotics by humans is recorded. Higher incidence of multi-resistant strains were recorded in the stream and well water sources than the tap water source. Antibiotic resistance in bacteria is a serious problem facing our society today and one of the reasons responsible for this is overuse of antibiotics by humans (Oyedeji et al., 2011).

### Conclusion

The results of this investigation revealed that the bacteriological quality of both the treated tap water samples and the untreated well and stream water sources failed to meet the standard for drinking water. Strains of *E. coli* isolated from the stream and well water sources showed greater multiple antibiotic resistance as compared to the tap water source. It is therefore recommended that water from these sources be treated either by boiling or chlorinating before drinking, while the concern governmental agencies should channel effort towards improving or providing safe drinking

water supplies to the areas. Thirdly, the indiscriminate use of antibiotics in therapy should be avoided to prevent the development of more antibiotic resistant bacterial strains. Further studies on this subject to include other rural water sources and communities are suggested.

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*Full Length Research Paper*

# Breast cancer knowledge and screening practices among women in selected rural communities of Nigeria

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Globally, the devastation that befalls women diagnosed of breast cancer remains inestimable. Early detection remains a major effective approach that should be employed to combat the disease. The issue of concern, however, is whether women in the rural underserved areas are aware of these early detection methods. This study was designed to assess rural women's awareness and knowledge of breast cancer and their screening practices. The study was a descriptive cross-sectional study utilizing self developed structured questionnaire. Reliability of the instrument was determined and alpha correlation values range between 0.81 and 0.95 for the different components of the questionnaire. Data was processed using descriptive analysis and associations tested with Chi-square at 5% level of significance. The results of the findings showed that 52.7% of the women had adequate knowledge about breast cancer risk factors and symptoms. Regarding the women's awareness about breast cancer screening methods, 52.8 and 51.7% of women have heard about Breast Self-Examination (BSE) and Clinical Breast Examination (CBE) respectively. However, few numbers of respondents (3.9%) were aware about mammography and these are people who did it for the purpose of diagnosing breast problem. Majority of the women (72.8%) did not practice BSE which is the most readily available screening method. Considering the lack of sophisticated diagnostic technology such as mammography for breast screening in the rural areas, BSE provided a unique opportunity for the women to be breast aware and to identify breast problems which may constitute danger to their health in the future. Health care professionals, most especially those at the primary health care level, should enhance the women's skills to use this simple procedure effectively to promote their health.

**Key words:** Community health nurses, breast awareness, breast examination, knowledge, practice.

## INTRODUCTION

The rising global incidence of malignant diseases as documented by World Health Organization (WHO, 1996) is an issue of serious concern, particularly in the developing countries where the increase seems to be more preponderant. In Nigeria for example, over 100,000 people develop cancer annually with majority of patients arriving medical centres at a late stage, thus resulting in a high mortality rate (Sasco, 2001). Over the years, people had the belief that breast cancer is an older woman's disease, therefore, the primary focus has been on prevention, detection and treatment of breast cancer for

Women who are 50 and older (Kinnon, 2003). But in the African-American community, the disease can strike well at a younger age and this prompted physicians and cancer advocate groups to recommend that women should get baseline mammograms at age 40. The American Cancer Society (2012) also recommends that women, starting from age 20 should be educated on the benefits and limitations of performing a monthly Breast Self- Examination (BSE).

Breast screening refers to tests and examinations used to detect a disease such as cancer in people who do not have any symptoms. Since the degree of success in treating breast cancer is influenced primarily by the stage at which intervention is introduced, secondary prevention (early detection) is the mainstay (Morrison, 1996).

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Changes in the breasts can be detected early by screening methods such as breast self examination (BSE) which is also known as breast awareness, clinical breast examination (CBE) and mammography screening. An ideal screening test would be simple, inexpensive, and effective. Breast self-examination fulfils the first two criteria, but previous results of two randomised trials conducted in Russia and China suggest that it would not be effective in reducing mortality from breast cancer (Semiglazov et al., 1993; Thomas et al., 1997). Despite the varying controversies about the use of BSE, including its sensitivity, specificity in detecting breast cancer as highlighted by Allen et al. (2010), BSE still remains the most readily available methods of screening to rural women especially in most part of the low resource countries where sophisticated diagnostic screening methods are not easily accessible in term of affordability and availability. Therefore, it is still important for women to be breast aware and also to be able to do this simple procedure efficiently to detect any abnormality in their breasts.

Clinical breast examination is also relatively simple and inexpensive, but its effectiveness in reducing mortality from breast cancer has not been directly tested in a randomized trial. Mammography is complex and expensive, but may pick up tumors long before they can be detected in any other way, thus enhancing better prognosis than those whose cancer is detected in some other ways (Aldridge, 2005). However, early diagnosis has been found to improve survival chances irrespective of the method that is used. Early diagnosis is only possible by educating women on the importance of examining their breast regularly to identify any abnormality, which could later lead to significant health problems. Although mass screening programmes for breast cancer were welcome for health promotion purposes, they are not always attended by those women mostly in need (Faithful, 1994). Despite the fact that the benefits of screening for breast cancer are well documented, many women, particularly poor, medically underserved, and ethnic minority, do not participate in the screening programmes. Past literatures have linked poor participation in screening to barriers such as high cost, lack of awareness of the need for screening and fear of breast cancer (Adam, 2000). Poor participation in breast self examination has also been linked to lack of confidence in performing the procedure (McMillan, 1990; Salazar, 1994; Budden, 1995; Javandi et al., 2002; Ahuja and Chakrabarti, 2010). In the developed countries, the National Breast Screening Programme currently provides routine mammograms every three years for women between 50 and 64 years. For women who are too young or too old to be included in the screening programme, breast awareness has been of great importance to help in the discovery of any early changes in the breast tissues. Women age 20 and older have been advised to receive clinical breast examination every 3 years; women age 40

and older to receive clinical breast examinations every year.

This study was influenced by researchers' interaction with clinic attendees of the General Outpatient Department of a tertiary Institution in the same state where the current study was done. Many women were referred to this institution on diagnosis of breast disease, which was usually confirmed to be advanced carcinoma of the breast. From consultation with these women, many of them were not really aware of the exact problem they have. Majority thought it was a mere infection of the breast that could be easily cured with drugs. It was also discovered that majority of the women came from the rural parts of the state and other rural areas in the Southwestern states of Nigeria. These generated a lot of questions as to whether these women are aware about breast cancer, the risk factors and screening methods. This study was therefore designed to determine the knowledge of women in the rural communities about risk factors and symptoms associated with breast cancer, awareness level about the screening methods and their screening practices. The study is part of a larger study that looks into women's knowledge and health beliefs about breast cancer and screening practices in Egbeda Local Government Area of Oyo State.

## METHODOLOGY

### Study design

This study adopted a descriptive cross sectional design using four rural local communities in Egbeda Local Government Area (LGA) of Oyo State, Nigeria.

### Study location

Egbeda Local Government Area is a semi-rural LGA and about 60% of the LGA is rural. This percentage is beginning to change with the rapid development in the erstwhile rural parts of the LGA. The local government consists of 11 political wards with projected estimated population of 242, 297, while women of childbearing age in the local government was 53,305.

### Sampling of study location and subjects

The study utilized a multistage sampling method for selection of location and study samples. The LGA selected for the study was purposively selected among the 11 LGAs in Ibadan, Oyo State. Selection was because the LGA consists of core rural areas that are ideal for the study among other LGAs. Three wards were randomly selected for the study out of the eleven political wards and six primary health care facilities (PHCF) were randomly selected from the list of eighteen PHCF in the selected wards by simple balloting, with two PHCF selected from each ward. The average monthly attendance of women attending the clinics in these health facilities were 150, 70, 60, 170, 90 and 60, respectively. Proportional method was used to select 30% of the women from each clinic to form a sample size of 180. The study utilized convenience sampling to identify women who participated in the study and these were women within the age range of 20 and 60



attending the clinic at the time of visitation of interviewers and who gave their consent to participate.

#### Data collection

Interviewer-administered technique was employed. Data was collected using self-developed questionnaire by the researchers after extensive review of literature for a period of four weeks. The instrument explored respondent's level of knowledge on the symptoms and risk factors of breast cancer and their screening practices. Some questions were asked sequentially on the participant's knowledge and scores were attached to each of the question answered. Seven different questions were asked and a total score of 21 obtainable. The women's knowledge were graded as good and poor. The knowledge is poor when a woman scores below the average score of 11 that is between 0-10 and good when she scores between 11 to 21. The instrument also sought information on respondent's awareness of breast cancer screening methods and their practices. Questionnaire was translated into local language to facilitate understanding and uniformity of information by the interviewers. Six research assistants were recruited and trained from Egbeda Community Women Action Group (a voluntary community based organization in the Local Government Area that works with the PHCF) to administer the questionnaire. The women consisted majorly of retired and active health care practitioners. Reliability of the different components of the instrument was done through test-retest method with alpha correlation of 0.81 to 0.95.

#### Data analysis

The raw data from the field was screened for inconsistencies and duly edited. Data was processed using Statistical Package for Social Sciences. Analysis was done using descriptive statistics and inferential statistics. The results obtained were plotted on frequency distribution. Cross tabulation was used to examine relationships between variables. Associations were tested using Chi-square test.

#### Ethical consideration

Permission to carry out the study was sought from the medical officer in charge of the local government area and each clinic before the commencement of the study. Participation in the study was made voluntary. Informed consent was obtained and participants were assured of the confidentiality of their responses.

### RESULTS

There were 180 women recruited for the study on their knowledge and health beliefs about breast cancer and screening practices. The average age of the women was 37.13 years, with standard deviation of 11.94. Their minimum and maximum ages were 20 and 60 years, respectively, while a good proportion of women's age was between 20 to 21 years (31.7%). Equal proportion of women (8.3%) was found between age 50 to 59 years and 60 to 69 years (8.3%). A little above thirty percent (30.6%) of the women had tertiary education followed by those with secondary education (29.4%). Equal proportion (20.0%) of women conferred that they had no formal education and primary education. Majority (78.9%) of the women were married, while 14.4% were single,

3.9% were separated, while 1.1 and 1.7% were widows and divorcees respectively (Table 1).

Table 2 shows the awareness of respondents about the screening methods and their practices. A little above half of the women (52.8%; n=95) declared that they have heard about breast self-examination, 51.6% (49) of these women performed BSE out of which 20% (36) of them performed it for the purpose of detecting breast cancer early. Meanwhile, few women (7.2%; n = 13) performed BSE during pregnancy and not for breast cancer purposes. The major reason for not performing BSE by the respondents was lack of self confidence to do it. Majority of the women (47.2%; n = 95) who have heard about BSE got the information from the health professionals. This was followed by the media which constituted 4.5% of the total population. The last and very negligible source of information was family and friends. Out of the 49 (27.2%) women who practiced BSE, 21 (11.7%) of them did it on monthly basis, followed by women who only practiced it during pregnancy period (7.2%; n=13). The rest would either do it everyday, twice a month, or every six month. Only one respondent did it on a yearly basis. Majority of the women (61.1%; n= 110) who were not practicing BSE would like to start doing it, while few of the women (11.7%; n= 21) did not see any reason for examining their breasts on a regular basis. According to them, every human being must experience what he/she had been destined to experience and therefore, there was no reason to worry themselves over what they did not have control over.

Clinical breast examination (CBE) was known to 51.7% (n=93) of the women and 31.7% (n=57) of them confirmed that they have had breast examination by health professionals. While 19.4% (n=35) had it for the purpose of detecting breast cancer, others had it during pregnancy as part of general examination. CBE was mostly performed by the medical doctor in 32 (17.8%) and followed by nurses in 25 (13.9%). Majority of the respondents (41.1%; n= 74) were of the opinion that women should visit hospital to have their breast examined by health professionals on a yearly basis. This was followed by those who felt that women should examine their breast on a monthly basis (10.6%; n=19). However, 38.9% (70) of the women did not know the frequency of performing CBE. Others were those who felt that women should go for breast examination with health care providers once in two years (6.7%; n= 12), twice a year (2.2%; n= 4) or once in a week (0.6%; n= 1). The study also showed clearly that majority (77.8%; n=140) of the women have not heard about mammography as a screening test for breast cancer. Forty women (22.2%) said they had heard about the screening method out of which 3.9% (n=7) of them confessed that they had the screening for the purpose of knowing whether they have breast cancer. Most women (18.3%; n= 33) got information on mammography from the hospital and this was followed by the mass media which constituted 3.3%

**Table 1.** Demographic characteristics of respondents.

<b>Characteristics</b>	<b>Frequency (n = 180)</b>	<b>Percent (%)</b>
Age (years)		
20-29	57	31.7
30-39	51	28.3
40-49	42	23.3
50-59	15	8.3
60-69	15	8.3
Level of education		
None	36	20.0
Primary	36	20.0
Secondary	53	29.4
Tertiary	55	30.6
Marital status		
Single	26	14.4
Married	142	78.9
Separated	7	3.9
Divorced	2	1.1
Widowed	3	1.7
Religion		
Christianity	113	62.8
Islam	65	36.1
Traditional religion	2	1.1

**Table 2.** Respondents' awareness of breast cancer screening methods and practice.

<b>Breast cancer screening methods awareness and practice</b>	<b>Frequency (N = 180)</b>	<b>(%)</b>
<b>Breast self examination</b>		
Heard about breast self examination		
Yes	95	52.8
No	85	47.2
Performing breast self- examination		
Yes	49	27.2
No	46	25.6
Not applicable	85	47.2
BSE for detecting breast cancer		
Yes	36	20.0
No	13	7.2
Not applicable	131	72.8
<b>Mammography</b>		
Heard about mammography		
Yes	40	22.2
No	140	77.8
Had a mammogram		
Yes	7	3.9

**Table 2.** Contd.

No	33	18.3
Not applicable	140	77.8
Mammogram for detecting breast cancer		
Yes	7	3.9
Not applicable	173	96.1
<b>Clinical breast examination</b>		
Heard about clinical breast examination		
Yes	93	51.7
No	87	46.1
Had breast examination by health professional		
Yes	57	31.7
No	36	20.0
Not applicable	87	48.3
CBE for detecting breast cancer		
Yes	35	19.4
No	22	12.2
Not applicable	123	68.3

**Table 3.** Respondents' knowledge of breast cancer symptoms and risk factors.

Level of Knowledge /range of score	Frequency (N = 180)	Percentage (%)
Poor knowledge (0 - 10)	78	43.3
Good knowledge(11 - 21)	102	56.7

(n=6) of the overall population. A respondent got information on mammography from the mother. Examining the participants' opinion about the age for the first mammogram, findings showed age range 15-24 as being considered the best age for the first mammogram for women (34.4%; n=66). Respondents were of the opinion that mammogram should be performed by women who are still sexually active especially women in the reproductive age range.

Respondent's level of knowledge about the risk factors and symptoms of breast cancer is provided in Table 3. Less than fifty percent (43.3%; n=78) of the study population had poor knowledge of breast cancer and its risk factors. The symptoms and risk factors of breast cancer were known by more than half (56.7%; n=102) of the respondents. This means that women with good knowledge of breast cancer dominated the study (56.7%). They were women who scored above the average of total score while those who scored below the average were women with poor knowledge (43.3%). The demographic variables such as age, marital status and religion had no significant association with the women's screening practices. However, a significant association

was found between the educational status of women and their screening practices ( $p=0.000$  for BSE; 0.01 for Mammography and 0.000 for CBE). Meaning that the higher the educational status of women, the more the likelihood that they are going to participate in screening test for breast cancer.

## DISCUSSION

The knowledge demonstrated by the participants ranged from little to adequate knowledge. The level of knowledge reported among the women was in contrast with studies conducted in Turkey where majority of the women has little knowledge (Cetingoz et al., 2002) and among US minority where the knowledge of the women is limited (Ko et al., 2003). Also, the result was in contrast with findings by Oluwatosin and Oladepo (2006) that knowledge of breast cancer risk factors among rural women was poor. The knowledge of the women in this study supported findings by other researchers that knowledge of breast cancer varies among women (Milaat, 2000; Odusanya, 2001). The practice of women regarding BSE

corroborated the studies by Hill et al. (1988), Sadler et al. (2001) and Ahuja and Chakrabarti (2010) that many women do not practise BSE. This was contrary to previous studies in suburban and urban communities in Nigeria where it was reported that higher percentages of population practise BSE. However, they do not practice BSE according to the classic technique (Ajayi and Adebamowo, 1999; Jebbin and Adotey, 2004).

Exploring the intention of the women to start practicing regular screening showed that some of the women (11.7%) still do not see any reason for screening. This attitude towards screening could be linked with the Filipino-American beliefs of "*bahala na*" that one needs not worry about unpleasant circumstances because such events are beyond the individual control (Wilson and Billones, 1994). From this study, the main reason for not performing BSE as stated by the respondents was majorly lack of self confidence. This supported findings by McMillan (1990), Salazar (1994), Budden (1995), Javandi et al. (2002) and Ahuja and Chakrabarti (2010). The women's commonest source of information on BSE was through health care professionals. The same trend was observable for other screening methods. These findings were in line with studies by Ko et al. (2003) among the US minority groups where health care professionals remain the commonest source of information. Other studies by Ajayi and Adebamowo (1999), Friedman et al. (1999) and Jebbin and Adotey (2004) are equally in agreement. However, our result disagreed with findings by Ahuja and Chakrabarti (2010) where majority get information from family and friends.

Furthermore, the findings from the study that majority of women have not heard about mammography as a screening modality for breast cancer was in line with study by Adebamowo and Ajayi (2000) that mammography is not known to many women in Nigeria and Oluwatosin and Oladepo (2006) where none of the respondents studied acknowledged mammography as an early detection measure. The findings from this study further showed that mammography was not known by women and it was not a readily available method of screening in the rural areas. Even though, mammography method of breast screening is not easily accessible to the rural dwellers, there is need for them to be well informed on this as the need for it may arise in the future. They can be prepared for such tests, thereby reducing the perceived barriers that may be associated. In this study, only the level of education had been significantly linked with respondents screening practices. The findings also revealed that the higher the level of education, the more the likelihood that a woman would practice BSE and easily utilize other screening methods if the need arises. This corroborated the studies cited by Maxwell et al. (2000, 2001). For example, the level of education of a woman is likely to influence a woman's adherence to mammography screening. Also, a woman with higher level of education is likely to report promptly to the

hospital for proper examination if any symptoms suggestive of cancer are seen or felt. The finding was however in contrast to study by Sensiba and Stewart (1995) that educated women often forget to practise BSE. Past literatures and the present study have confirmed that health care providers remain the major source of information to the people on breast issues. The community health nurse (CHN) is rightly placed to encourage and teach preventive behavior to rural women since women in the rural setting patronize primary health care centers more than other levels of health care. The CHN should combine approaches in encouraging women to develop self confidence and be compliant with screening especially BSE and to do it regularly and correctly through:

- (1) Education/Information: Providing regular information to women on breast cancer and screening method through the clinics and community awareness and sensitization programmes. Community structures as designed within the primary health care system can also be utilized by community health nurses as a medium for dissemination of information on breast cancer and screening practices. Such structures include Community Development Committee (CDC), Village Development Committee (VDC) and Traditional Birth Attendants (TBA). With these, women will be empowered to identify deviation from normal in their breast and report promptly to the health centre.
- (2) Demonstration: Women can effectively use BSE to detect abnormality in their breast if they develop self efficacy in doing it. The technique of BSE should not only be taught, but should be demonstrated using the model of the breast. Women should be allowed to practise it on the model and feedback provided. This will enhance their self confidence and increase their compliance with BSE practice.
- (3) Reinforcement: Women need a reminder to encourage them to perform BSE regularly. This may be developed in relation to each person's schedule. The reminder may be associated with something done as a routine monthly. For example women's performance of BSE may be linked with their monthly 'market' or church programmes that is of importance to them. The particular time should however be left to their discretion.
- (4) Advocacy: The community health nurse can also play advocacy role between the government and the people. Breast cancer is a disease with fatality and the importance of addressing the well-being of women needs to be advocated for. This can encourage a sponsored periodic community sensitization and awareness activities by the local health authority on the general health of the women including breast awareness. Media programme, including jingles which are presented in language that can be easily understood by people could also be used to provide necessary information. Women identified through this means with benign growth in the

breast or with early stage of cancer could be supported by government or other interested stakeholders for prompt treatment.

This study focused more on breast awareness and breast self examination because it is the most easily and readily accessible method to rural dwellers. However, other screening methods were explored such that when the women need such services; the barrier to accessing them could be minimized based on prior information and knowledge of the usefulness of such screening measures. This research is descriptive in nature and therefore relied on self-reported behaviour, which could have been sensitive to demand and presentation concerns. The main limitation of this study was the small sample size and its geographical restriction to one Local government area in Nigeria. Therefore, caution should be taken in generalizing the findings of this study. In addition to the aforementioned limitation, there had not been any specific data on existing cases of breast cancer in the Local Government Area which would have been more appropriately employed in calculating the proportion of the population to be sampled. There may be need for intervention studies to further ascertain the usefulness of BSE in Nigeria and other low resource countries. Such studies should be designed to explore greater number of women in the rural areas for better representation.

## Conclusion

A lot of research has been done to promote treatments and prognosis of breast cancer. However, early detection still remains the best antidote. One easy and accessible way of detecting any abnormality in the breast is through breast awareness, which also includes BSE. All women need to be encouraged to be responsible for their own health and well-being by utilizing this simple procedure. They need to be encouraged to perform BSE regularly and earnestly report any abnormality to the health care providers. Community health nurses are rightly positioned to advocate for this behaviour and by so doing, express their commitment to community vis-à-vis reduction in the mortality associated with breast cancer.

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*Full Length Research Paper*

# **A pilot study of iodine and anthropometric status of primary school children in Obukpa, a rural Nigerian community**

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**This study was designed to assess the iodine and the anthropometric status of primary school children in a rural Nigerian community (Obukpa). Stratified random sampling technique was used in selecting the respondents for the study. The sample size was 330 children aged 6 to 12<sup>+</sup> years. The anthropometric measurements of the children comprising 170 boys and 160 girls were obtained using standard procedures. The urine of a sub-sample of 36 children were subjected to laboratory analysis for iodine using “method H”. Data generated from this study were analyzed using Statistical Package for the Social Sciences (SPSS) version 12, means, standard deviation, student t-test, analysis of variance (ANOVA) and correlation coefficient (r) values. Anthropometric measurements of the children showed that 25% were stunted. More boys (13.7%) were stunted than girls (11.3%). Stunted children correlated negatively with wasted children significantly ( $P < 0.05$ ). Urinary iodine showed that 58.3% of the children had  $<20 \mu\text{g/L}$  indicating severe iodine deficiency. Iodine levels in urine correlated with stunting and underweight.**

**Key words:** Urinary iodine status, anthropometric status, primary school children.

## **INTRODUCTION**

**Iodine Deficiency and Physical Development:** The physiological effects of iodine are realized by the thyroid hormone in human body. Thyroid hormone is one of the most important hormones in the human body. Hence the physiological effects of iodine are: Firstly, it can maintain the metabolism of energy in the body ([www.lookchem.com](http://www.lookchem.com)). It breaks down the substances in human body, so as to provide the energy required for basic life activities. What's more, it can promote the production of heat, so as to maintain the normal temperature of human body. Iodine deficiency will decrease the synthesis of thyroid hormone, which may cause damage on the basic life activities and decline physical ability. Secondly, iodine can promote physical development. Iodine can regulate the development of bones and muscles for children at the growth and developmental stage. Iodine deficiency can lead to physical retardation, muscle weakness and other symptoms. Thirdly, iodine can

promote the development of brain. For the fetuses and infants, the development of their brain is closely related with iodine. As a result, iodine deficiency will greatly affect brain development, and even may lead to mental retardation, while this disorder is essentially irreversible.

Foods that contain a large number of iodine: In general, foods which contain great amounts of iodine are mostly seafood, such as kelp, seaweed, dried scallop, mussel, sea cucumber, jellyfish, lobster, and so on. Seaweed contains the highest content of iodine (one kilogram of fresh kelp contains more than 2000 micrograms of iodine); and followed by marine fish and shellfish. Apart from seafood, other foods such as egg, milk and meat also contain a large number of iodine. Plant food contains the lowest amount of iodine in all the foods.

In addition, iodized salt contains a really small amount of iodine, especially the purified salt. Every one kilogram of sea salt contains about 20 micrograms of iodine. If one person takes in 10 grams of salt every day, he can only take in 2 micrograms of iodine. This can hardly meet the need for the prevention of the disorders caused by iodine deficiency ([www.lookchem.com](http://www.lookchem.com)). In many developing countries, however, children hardly grow to their full

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potential as a result of many environmental factors such as malnutrition and infections (Van de Poel et al., 2008). Children from poor or less privileged families in those countries are the most affected due to food insecurity, inadequate facilities, infection and poor general environmental sanitations.

The causes of child undernutrition are complex, multidimensional, and interrelated, ranging from factors as fundamental as political instability and slow economic growth to those as specific in their manifestation as respiratory infection and diarrhea disease (ACC/SCN, 2000). Lack of progress to combat malnutrition is damaging to children and nations. For every visibly undernourished child, there are several more battling a hidden nutritional crisis. Many are seriously deficient in essential vitamins and minerals such as Iodine, Vitamin A and Iron (Clements, 2006). Poor nutrition remains a global epidemic contributing to more than half of all children's deaths, about 5.6 million per year. UNICEF's charts and progress reports towards the first UN Millennium Development Goal are aimed to eradicate extreme poverty and hunger by 2015. Achieving this goal means halving the proportion of children who are underweight for their age, the most visible sign of malnutrition (Clements, 2006).

Most of the Iodine status monitoring with anthropometric studies was done outside Nsukka has boundary with Kogi state that is endemic of goiter. Thus, there is the inadequate documentation of the Iodine status and anthropometric status of Nsukka school age children. These groups of children are also susceptible to Iodine deficiency disorders.

The general objective of this study was to assess the Iodine status and the anthropometric status of primary school children in rural community- Obukpa in Nigeria.

## MATERIALS AND METHODS

The study was carried out in Obukpa in Nsukka Local Government Area of Enugu State, Nigeria. The subjects consisted of primary school children aged 6 to >12 years.

### Approach to the local community

A letter from the University introduced the researcher to the Chief of the community. Subsequently, the Chief gave approval that was presented to the Headmaster of each school; permission for the study was obtained.

### Design and sampling procedure

3 out of 7 primary schools in the community were randomly selected for this study.

### Sample size and sample size calculation

This was calculated using the formula ([www.herkimershideaway.org/writings/sizesam.htm](http://www.herkimershideaway.org/writings/sizesam.htm)) as follows:

$$N = \frac{4P(1 - P)}{W^2}$$

where N = total number of children required, P = proportion of the subjects assumed to have subnormal iodine urine excretion and nutritional status. P = 28%.

Maziya-Dixon et al. (2004) reported in International Institute of Tropical Agriculture (IITA) that 27.5% of the children suffered various degrees of Iodine deficiency. W = required precision level or probability level for the study. W = 0.05(5%). In order to make up for drop outs, this figure was approximated to 330. The sub-sample was obtained using 10% of the total sample, which were used in determining the casual urinary iodine excretion

$$[10 \times 330] / 100 = 33$$

The 33 were marked up to 66 in case of any drop out.

### Sampling procedure

A multistage sampling procedure was used in the study. The names of all the primary schools in Obukpa Community were collected from the Local Government Area Education Authority. Three schools were randomly selected from the list of schools in the community. The sampling frame comprised all school children from elementary 1 to 5. The source of sample frame was obtained from the school register. This was done by stratified systematic random sampling and simple random sampling. The three schools constituted the three strata. The total sample size was 330 children.

### Data collection

Two research assistants from the community were trained on collection of urine and taking of anthropometric measurements. General anthropometric measurements were made on all the units in the sample size to determine their nutritional status.

Anthropometric measurements included height and weight. The heights of the children were taken using a portable height gauge calibrated in centimeters (cm). The children were measured without shoes on a flat floor. A good standing posture was maintained before measurement. The heels were pulled together touching the base of the wall. The head was erect and hands hung at their sides in natural manner. The child's line of sight was level with the ground for accuracy. The measurements were taken to the nearest 0.1 cm. after the head piece was placed on the head of each subject; the reading was taken to the nearest 0.1cm.

### Weight

The weights of the children were taken using a Salter beam balance scale this was calibrated in kilograms and pounds. The scale was adjusted to zero before each measurement. The children were weighed with minimum clothing. The reading was taken to the nearest 0.1 kg. Ages of the children were derived from the school registers the school records of pupils' age were derived from birth certificates on admission.

Anthropometric status was derived by computing weight-for-age, height-for-age and weight-for-height using the z-score classification system. The anthropometric value was compared with World Health Organization (WHO, 2006) reference standard.

### Urinary Iodine collection

Casual urine samples were collected from the subjects, in sterilized

clean containers marked, numbered and distributed to the children on the day of collection. The samples were transported in an ice pack to the laboratory and were analyzed immediately, with the help of a biochemist, using "method H", as recommended by IDD Newsletter 1999. Urinary iodine levels were determined using small samples of urine (250 to 500  $\mu$ l) digested with ammonium persulfate at 90 to 110°F, adding arsenious acid and uric ammonium sulphate. The decrease in yellow colour over a fixed time period was measured by a spectrophotometer and plotted against a curve constructed from standards with known amounts of iodine, ran in the same assay. Replicates of 3 samples were analyzed to check the accuracy and reproducibility of the method

#### Data analysis

The data were coded and keyed into the computer, then analyzed using the Statistical Package for the Social Sciences (SPSS) version 12 computer software. Descriptive statistics such as frequencies, percentages, mean, standard deviation as well as correlation coefficient ( $r$ ) were used to appraise the validity and reliability of the dependent variables such as weight-for-age, height-for-age, weight-for-height, and causal urinary iodine levels. Analysis of variance (ANOVA) was used to compare means such as weight-for-age, height-for-age, weight-for-height, and causal urinary iodine levels. The Student t-test was used to compare the males and females' urinary iodine mean levels.

## RESULTS

Figure 1 shows the prevalence of (13.0%) underweight of the surveyed respondents, using weight-for-age, while 25.0% were stunted using height-for-age and 5.3% were wasted using weight-for-height.

Figures 2 and 3 compared the mean weight-for-age and height-for-age of the males and females with their NCHS/WHO reference 50<sup>th</sup> percentile. Figure 2 shows that the entire age-groups of the males fell below their NCHS/WHO reference 50<sup>th</sup> percentile. There were fluctuations in the weight as the ages increased but from 10 years on, there was a steady increase. Figure 2 shows that the females in the age-groups also fell below their NCHS/WHO reference 50<sup>th</sup> percentile, but had a steady appreciation in weight as the ages increased. All the males in the age-groups in Figure 3 had their heights below their NCHS/WHO reference 50<sup>th</sup> percentile. Like the males, the heights of the female age-groups fell below their NCHS/WHO references except age-groups 6 to 7 years, 11 months.

Anthropometric status classification according to sex in Table 1 showed that more females (3.0%) than males (2.3%) were wasted, while more males (13.7%) than females (11.3%) were stunted and more males (6.7%) than females (6.3%) were underweight.

Table 2 displaying anthropometric status classification according to age-group, revealed that more of age-group (6 to 9 years, 11 months) (3.3%) than age-group (10 to 13 years, 11 months) (2.0%) were wasted. Weight-for-age showed that more (9.3%) of (10 to 13 years, 11 months) were underweight than 3.7% of age-group 6 to 9 years, 11 months. Height-for-age revealed that more 15.4% of 10 to 13 years, 11 months were stunted than

9.8% of 6 to 9 years, 11 months.

#### Iodine status of the children in the studied communities

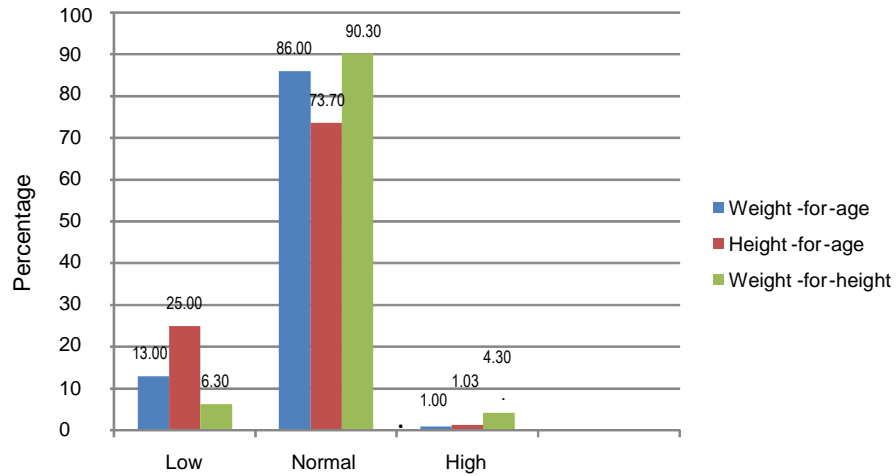
Figure 4 revealed that majority (58.33%) of the children had <20  $\mu$ g/L iodine in urine which denotes severe iodine deficiency. About 13.89% had 50 to 99  $\mu$ g/L (mild iodine deficiency), and 11.11% had >300  $\mu$ g/L (possible excess). On the other hand, 8.33, 5.56 and 2.78% had 100 to 199  $\mu$ g/L (optimal), 20 to 49  $\mu$ g/L (moderate iodine deficiency) and 200 to 299  $\mu$ g/L (more than adequate), respectively. Table 2 shows that more boys (33.33%) than girls had <20  $\mu$ g/L (severe iodine deficiency level) and more girls (13.89%) than boy (0%) had 50 to 99  $\mu$ g/L (mild iodine deficiency). More girls (5.56%) than boys (2.78%) had 100 to 199  $\mu$ g/L (optimal) iodine in urine. However, only girls (2.78%) had 200 to 299  $\mu$ g/L (more than adequate) and more boys (8.33%) than girls (2.78%) had >300  $\mu$ g/L (possible excess) iodine.

Table 3 shows that the correlation coefficient ( $r$ ) between urinary iodine level of the pupils and the underweight and stunted had positive values ( $r = 0.240$ ) and ( $r = 0.314$ ), respectively and the wasted had negative value ( $r = -0.179$ ). The relationship was stronger among stunted children and urinary iodine level ( $r = 0.314$ ). However, it was generally weak with other nutritional parameters. Stunted children had the highest urinary iodine correlation coefficient ( $r = 0.314$ ) level. This relationship could be regarded as below average within the degree range of 0 to 1. Table 3 also shows a correlation coefficient ( $r = 0.351$ ) between underweight and stunting as being significant ( $P < 0.05$ ). There was a negative relationship ( $r = -0.398$ ) between wasted and stunted children and it was significant ( $P < 0.05$ ).

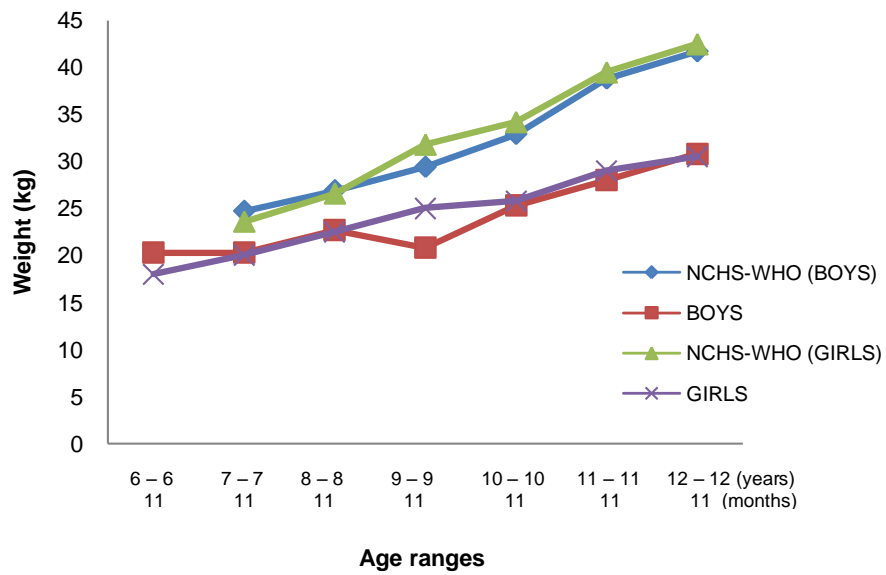
ANOVA test in Table 4 shows that Owerre-Obukpa CPS had the maximum (94.0  $\mu$ g / L  $\pm$  7134) mean urinary iodine level followed by Ajuona CPS (63.0  $\mu$ g / L  $\pm$  70.05) and Amarua/Amagu (55.0  $\mu$ g / L  $\pm$  48.28) and there was no significant ( $P > 0.05$ ) differences. The student t-test comparing urinary iodine level of the males and females, shows that the females had the highest (80.0  $\pm$  63.12) urinary iodine level than the males (54.0  $\pm$  58.40) and there was no significant ( $P > 0.05$ ) difference

## DISCUSSION

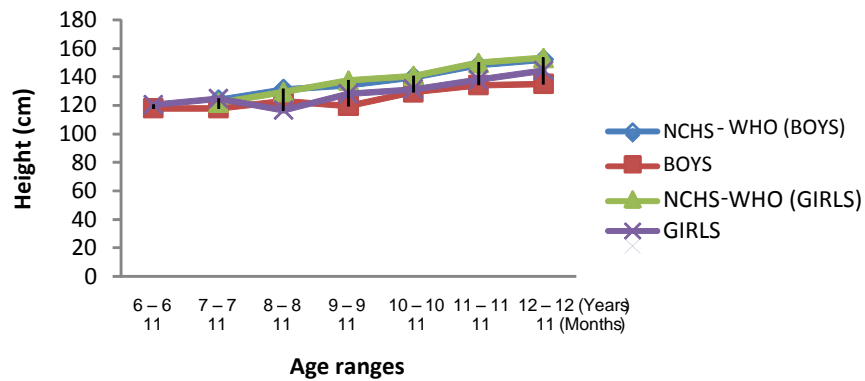
The 25.0% stunting, 13.0% underweight and 6.3% wasting observed in this survey agreed with those of Grantham-McGregor et al. (1989) who observed that globally stunting is much more prevalent. The FGN/UNICEF (1994) study observed more stunting (36 to 52%) and underweight (27 to 45%) than wasting (5 to 12%). On the other hand, the magnitude of stunting and wasting observed in the present study was lower than that of Nnanyelugo et al. (1990). They observed 50.0% stunting



**Figure 1.** Prevalence of underweight, stunting and wasting of school children in Obukpa community.



**Figure 2.** Comparing mean weight-for-age of males, females with NCH/WHO reference group.



**Figure 3.** Comparing mean height-for-age of males and females with NCH/WHO reference group.

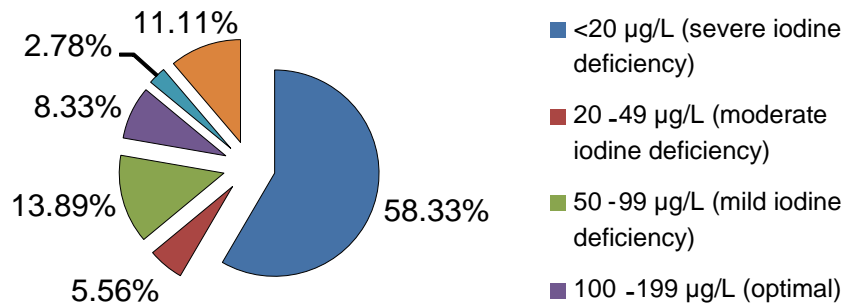


Figure 4. Iodine status of the school children.

Table 1. Distribution of underweight, stunting and wasting among males and females in Obukpa community.

Variable	N (%)	
	Boys	Girls
<b>Weight-for-age</b>		
<-2SD	20 (6.7)	19 (6.3)
-2 to +2SD	131 (43.7)	127 (42.3)
>+2SD	3 (1.0)	-(-)
Total	154 (51.3)	146 (48.7)
<b>Height-for-age</b>		
< -2SD	41 (13.7)	34 (11.3)
-2 to +2SD	111 (37.0)	110 (36.7)
>+2SD	2 (0.7)	2 (0.7)
Total	154 (51.3)	146 (48.7)
<b>Weight-for-height</b>		
<-2SD	7 (2.3)	9 (3.0)
-2 to +2SD	139 (46.3)	132 (44.0)
>+2SD	8 (2.7)	5 (1.7)
Total	154 (51.3)	146 (48.7)

< -2SD, Low; -2 to +SD, normal; >+2SD, above normal; W/A, weight-for-age; H/A, height-for-age; W/H, weight-for-height. Z-score of < -2.0SD from NCHS-WHO (1976) median value was used as cut-off. Low weight-for-age, underweight; low height-for-age, stunting; low weight-for-height, wasting.

Table 2. Iodine status of the children according to sex.

Iodine level in urine (µg/L)	Status	Boys		Girls		Total	
		N	%	N	%	N	%
<20	Severe Iodine deficiency	12	33.33	9	25	21	58.33
20 - 49	Moderate Iodine deficiency	1	2.78	1	2.78	2	5.56
50 - 99	Mild Iodine deficiency	-	-	5	13.89	5	13.89
100 - 199	More than adequate	1	2.78	2	5.56	3	8.34
200 - 299	Optimal	-	-	1	2.78	1	2.78
>300	Possible excess	3	8.33	1	2.78	4	11.11
Total		17	47.22	19	52.79	36	100.0

**Table 3.** Correlation coefficient (r) values expressing the relationship between the iodine level in the urine and malnutrition status. N= 33.

Variable	Underweight	Stunted	Wasted	Iodine level in urine
Underweight	1	.351 (*)	-.110	0.240
Stunted	0.351	1	-0.398 (*)	-0.314
Wasted	-0.10	-0.398 (*)	1	-0.179
Iodine level in urine	0.240	0.314	-0.179	1

\*Correlation is significant at 0.05 level (tailed).

**Table 4.** Mean ( $\pm$ SD) urinary iodine level as influenced by schools and sex of respondents.

School	Minimum	Maximum	Mean	Standard deviation
Ajuona CPS	14.9	163.40	63.0	$\pm$ 70.05
Amagu/ Amarua CPS	14.9	133.70	55.0	$\pm$ 48.28
Owerre-Obukpa CPS	14.9	208.0	94.0	$\pm$ 71.34
LSD <sub>(0.05)</sub>			NS	
<b>Sex</b>				
Male	14.90	163.40	54.0	$\pm$ 58.40
Female	14.90	208.00	80.0	$\pm$ 63.12
LSD <sub>(0.05)</sub>			NS	

and 70.0% wasting. Onofioke (1998) reported 43.0% underweight, 33.0% stunting and 18.6% wasting. The explanation for the differences might be that the previous studies were conducted in the rural farmland communities who depended heavily on cassava consumption. Obukpa is a semi-urban who consumed a lot of mixed cereals, legumes and vegetable dishes. These foods are rich in nutrients and essential amino acids which are more energy dense than cassava. The community is very close to the University of Nigeria Teaching Hospital, coupled with, possible improved maternal literacy. However, Olivieri et al. (2007) reported that if substantially more than 5% of an identified child population has height-for-age that is less than the 50<sup>th</sup> percentile on the reference curve, then the population is said to have a higher-than-expected prevalence of stunting, and malnutrition is generally the first cause considered.

Much is still being desired for children of developing countries to catch-up with their WHO reference age-group. The 58.33% severe iodine deficiency (<20  $\mu$ g/L), 5.56% moderate iodine deficiency (20 to 49  $\mu$ g/L) and 13.89% mild iodine deficiency (50 to 99  $\mu$ g/L) were observed in this survey. Kennedy et al. (2003) also noted that the true prevalence of iodine deficiency is even more widespread than the number of those affected with goiter which seems to indicate, however, that there were no global estimates for prevalence of low urinary iodine, which is the best sub-clinical indicator. The study agreed with Kennedy et al. (2003) that sub-clinical iodine deficiency

was detected by measuring urinary iodine which was an indication of low iodine intake and/or utilization. The low urinary iodine level reported in this study may be an indication of poor utilization of iodine consumed. Goitrogens are able to disrupt normal thyroid function by inhibiting the body's ability to use iodine, block the process by which iodine becomes the thyroid hormones thyroxine (T4) and triiodothyronine (T3), inhibit the actual secretion of thyroid hormone, and disrupt the peripheral conversion of T4 to T3 (thyroid.about.com ). Akunyili (2007) rightly reported that Nigeria currently ranks high on global and regional report cards. In 2005, the goitre rate was 6.2%, down from 20% in 1993. Median urinary iodine excretion rate has consistently been over 130  $\mu$ g/dl since 1999. Improvements in urinary iodine excretion and goitre rates have been substantive. In addition, the 10 point criteria for USI certification have consistently been met, which paved way for Nigeria's certification as the first country in Africa to achieve USI compliance in 2005 by the Network for Sustained Elimination of Iodine Deficiency. There is need therefore to look into the states foods that are consumed and their preparations

Low urinary iodine level of the stunted children was observed in this study. Stunting may be the effect of low iodine utilization, although there may be other predisposing factors, such as infections and poor nutrition that could give rise to stunting. Sofra et al. (1998) defined iodine as an essential mineral required by the body that directly affects thyroid gland secretions, which themselves to a great extent controls heart actions, nerve

response to stimuli and rate of body growth and metabolism. Therefore, estimates for prevalence of low urinary iodine, is the best sub-clinical indicator.

The non-significant ( $P > 0.05$ ) differences observed in the urinary iodine level between the community primary schools and sexes may be attributed to the small sub-samples size, which was a result of non-funding of the study.

## CONCLUSION AND RECOMMENDATIONS

The study indicated a higher prevalence of stunting than underweight. The existence of stunting, underweight and wasting in the school children in the semi-rural community may have serious implications in the general wellbeing of the children and their achievement and attainment later as adults. There was evidence of the intake/utilization of inadequate iodine as evidenced in the urinary iodine output. Important findings of this study are information on the relationship between growth and iodine deficiency and significant information on the nutritional status of Obukpa rural school children which can be extrapolated to other rural communities in iodine endemic areas for the purpose of intervention programmes, such as eradication of micronutrient deficiency.

The study revealed that more attention should be given to school children in developing countries like Nigeria. A large sample size should be studied in this community; also, urgent intervention programmes are recommended.

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*Full Length Research Paper*

# Prevalence of malaria and soil-transmitted helminth infections and their association with undernutrition in schoolchildren residing in Mfou health district in Cameroon

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**Malaria and soil-transmitted helminths are common parasitic diseases found in school children in sub-Saharan Africa. We determined the prevalence and intensities of these infections in apparently healthy schoolchildren living in Mfou health district, where malaria and intestinal infections are among the first causes of morbidity. In a cross-sectional study involving 503 schoolchildren, anthropometric parameters were measured. Finger-prick blood and fresh stool samples were collected for malaria parasites determination, assessment of anaemia and detection of helminths' eggs. Logistic regression analysis was used to investigate the association between these infections and other factors. Overall, 40.6 and 29.6% of children harboured malaria parasites and worms respectively. Prevalences of mild, moderate and severe undernutrition were respectively 22.2, 2.3 and 0.5% for underweight, and 21.9, 7.6 and 2.0% for stunting. In logistic regression analysis, anaemia (OR=2.64, 95% CI: 1.71-4.07) and infection with *Ascaris lumbricoides* (OR=1.72, 95% CI: 1.01-2.91) were significantly associated with malaria infection. Infection with *Trichuris trichiura* was significantly associated with increased risks of underweight (OR=2.11, 95% CI: 1.11-4.01). Moreover, rural schoolchildren showed increased chances of carrying worms, compared to their urban counterparts (OR=2.60, 95% CI: 1.75-3.86). Malaria prevention and school-based deworming activities should be re-enforced in Mfou health district to reduce the burden of these infections in children.**

**Key words:** Malaria, soil-transmitted helminths, undernutrition, schoolchildren, Mfou, Cameroon.

## INTRODUCTION

Malaria infection and soil-transmitted helminths (STHs) are among the most prevalent endemic parasitic diseases in sub-saharan Africa, where both diseases have similar geographical distribution and co-infections are common (Snow et al., 2005; Mwangi et al., 2006;

Brooker et al., 2007; Brooker, 2010). In Cameroon, both infections are prevalent and responsible for increased morbidities and associated consequences in vulnerable populations, including young children, pregnant women and school-age children (Quakyi et al., 2000; Brooker et al., 2000; Kimbi et al., 2005a,b; Tchuem and N'Goran, 2009; Leke et al., 2010). Schoolchildren living in an environment with inadequate sanitation (Ziegelbauer et al., 2012), usually in deprived communities in rural areas, are likely to be infected with at least one of the three main

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STHs (*Ascaris lumbricoides*, *Trichuris trichiura* and hookworms [*Ancylostoma duodenale* and *Necator americanus*]), as well as other helminths species (Tchuem et al., 2003; Bethony et al., 2006; Mupfasoni et al., 2009; Ayalew et al., 2011). Infections by intestinal helminths have been shown to result in abdominal pain, diarrhea, anaemia, malnutrition, ulcers, intellectual retardation, cognitive and educational deficits, intestinal obstruction and, in severe chronic and untreated infections, could even lead to death (Crampton and Nesheim, 2002; Hotez et al., 2008; Hall et al., 2008; Brooker et al., 2010). In the context of continuous exposure to *Anopheles* mosquitoes, these conditions are further exacerbated with asymptomatic malaria parasites, often resulting in co-infections with *Plasmodium*, and thus, putting these children at enhanced risk of clinical disease (Mwangi et al., 2006; Nkwo-Akenji et al., 2006; Achidi et al., 2008).

Current preventive strategies adopted by the Ministry of Public Health for malaria in Cameroon are based on the use of insecticide-treated nets in households and intermittent preventive treatment with sulphadoxine-pyrimethamine for pregnant women (Ministry of health/National Malaria Control Programme, 2011). On the other hand, activities aiming at reducing the burden of helminths infections in children are based on annual distribution of anthelmintics in primary schools (Tchuem and N'Goran, 2009), as well as other deworming activities which are integrated to immunization in pre-school children (1-5 years) within the community.

Regular update on the prevalence and intensities of these infections are important in order to assess the impact of these control interventions on the burden of the infections, especially in children. However, there is currently a scarcity of such information in Mfou health district where malaria and intestinal infections are ranked respectively among the first and third cause of morbidities within this rural district. Therefore, our study was designed to determine the prevalences of malaria and STHs and their association with undernutrition in primary schoolchildren residing in this study locality.

## MATERIALS AND METHODS

### Study area

The study was conducted in 4 primary schools of Mfou health district, a forest area located in the Mefou and Afamba division, in the centre region of Cameroon. Two of the schools were situated in the district headquarter (Mfou urban), an emerging semi-urban area located at ~25 km of Yaoundé, the Capital of Cameroon; while the two others were found in the rural area (Mfou rural). The population of the district was over 71 000 inhabitants, who were mainly farmers (District Health Management Team, 2011). Most households in Mfou urban and nearly all in the rural area have no access to potable water and appropriate sanitation, but the socio-economic conditions are relatively improved in Mfou urban. Mfou is hyperendemic for malaria; transmission occurs all year round, with peaks during the rainy season and transition to the dry season.

Malaria morbidity ranked the first position among the top ten causes of morbidity in the district, while intestinal infections ranked the third position (District Health Management Team, 2011).

### Study population

The study involved apparently healthy primary schoolchildren of both sexes aged between 3 and 16 years. This population also included children from the nursery section of one of the rural primary schools. Each participant was clinically examined for febrile symptoms (fever) or any other clinical conditions (headache, abdominal discomforts, etc.). Sick children were referred to Mfou district hospital for appropriate management.

### Anthropometric measurements and assessment of undernutrition

The age, sex, height and weight of children were measured to determine their anthropometric indices. Age of each child was obtained from school records, while weight was measured to the nearest 100 g using an electronic personal scale (Geepas®). Height was measured to the nearest 1 mm, using a height measuring board (Innotech International, 7-9 avenue F.V. Raspail-94110-France). Weight-for-age Z-score (WAZ) and height-for-age Z-score (HAZ) were calculated to assess underweight and stunting status respectively, as indicators of undernutrition. WAZ is easily available and can capture both stunting, associated with long-term undernutrition and wasting, a manifestation of recent and acute undernutrition (Caulfield et al., 2004). These calculations took into account children' weight and height, as well as the median weight and height of healthy children of the same age and sex (WHO, 1983). WAZ and HAZ scores were classified as mild (-1.01 to -2.00 Standard Deviation [SD]); moderate (-2.01 to -3SD) or severe (<-3SD) undernutrition, and normal (WAZ≥-1SD) (Caulfield et al., 2004; Crookston et al., 2010; Mfonkeu et al., 2010).

### Sample collection and laboratory analyses

Finger prick blood samples were collected directly onto a clean glass slide to prepare thick and thin blood films. Heparinised micro capillary blood was collected at the same time for assessment of anaemia. Additionally, fresh stool samples were collected from each child, in a clean plastic container. The samples were immediately transported in the laboratory for processing and analysis.

For malaria parasite detection, thin blood films were first fixed in methanol, stained with 50% May-Grunwald, and both thin and thick films were stained with 10% Giemsa stain. Slides were examined under a microscope (x100 objective). A slide was considered negative if malaria parasites were not detected after examination of 200-oil immersion fields of thick smear. For positive samples, the number of parasites per 200 white blood cells (WBCs) was counted and the parasite density estimated based on an average of 8000 WBCs/μl of blood. All slides were read independently by two microscopists, and when discrepancy occurred, the slide was read by a third person. *Plasmodium* species was determined in thin films. Children with malaria parasites were treated with a fixed combination of Artemether-Lumefantrine for three consecutive days, as recommended as one of the first line treatment for uncomplicated malaria in Cameroon.

For assessment of anaemia, heparinised micro capillary tubes containing fresh blood were centrifuged at 15,000 rpm for 5 min (IEC Micro-MB centrifuge). The packed cell volume or hematocrit level was determined using a micro hematocrit reader. Anaemia was defined as hematocrit level <33% (Quakyi et al., 2000).

Fresh stool samples were immediately processed using the Kato-

Katz thick smear method. Two slides were prepared per child, kept refrigerated overnight and egg-counts were performed within 15-24 h after slides preparation. The number of eggs counted in 41.7 mg of faeces was extrapolated as eggs per gram (epg) of faeces. Infected children were treated with a standard oral dose of Mebendazole, 2x100 mg per day for three consecutive days.

### Statistical analysis

All statistical analyses were performed using SPSS version 18.0 and threshold for statistical significance was set at  $p < 0.05$ . Overall descriptive statistics were carried out to select variables for consideration in multivariate regression models, using malaria infection or helminths status as dependent variable. Independent variables that were significant at  $p < 0.10$  were included in the models.

### Ethical considerations

The study was approved by the National Ethics Committee of Cameroon. Participation was dependent on a written consent given by the parents/guardian of the children.

## RESULTS

### Description of the study population

A total of 503 children participated in the study which included 269 (53.5%) males and 234 (46.5%) females. About 55.5% resided in the urban area of Mfou, while 44.5% lived in the rural area. Age ranged from 3 to 16 years, with a mean of  $8.4 \pm 2.7$  years. Children from urban Mfou were significantly younger (mean age =  $7.9 \pm 2.0$  years;  $p = 0.001$ ) than their rural counterparts (mean age =  $9.1 \pm 3.3$  years). A total of 40 children (8.0%) were feverish (axillary temperature  $> 37.5^\circ\text{C}$ ) at the time of the survey and the prevalence of fever was significantly elevated in children residing in Mfou urban (22.2%;  $p = 0.024$ ), compared to those from Mfou rural (17.8%).

### Prevalence of malnutrition and anaemia in the study children

Table 1 shows the prevalence of the different categories of undernutrition according to WAZ and HAZ scores' classification, as well as in relation to the sex of children. Overall, mild undernutrition ( $-1.01 \leq Z\text{-score} \leq -2\text{SD}$ ) was more prevalent in these children, while the prevalences of moderate ( $-2.01 \leq Z\text{-scores} \leq -3\text{SD}$ ) and severe ( $Z\text{-scores} < -3\text{SD}$ ) undernutrition were relatively low. Children residing in the rural area had significant increased chances of being underweight compared to their urban counterparts (OR=1.63, 95% CI: 1.02-2.61;  $p = 0.040$ ). Likewise, male schoolchildren relative to females, showed increased chances of being underweight (OR=1.66, 95% CI: 1.04-2.66;  $p = 0.033$ ). There was a significant age-related increase in the prevalence of underweight ( $p = 0.041$ ).

Similarly, rural schoolchildren had more than two-fold increased chances of being stunted than those living in urban Mfou (OR=2.33, 95% CI: 1.59-3.42;  $p = 0.000$ ). Also, male schoolchildren showed increased chances of being stunted compared to females (OR=1.59, 95% CI: 1.09-2.34;  $p = 0.016$ ). There was no significant age-related change in the prevalence of stunting ( $p = 0.210$ ). The overall prevalence of anaemia was 33.5% (168 children) and there was no significant difference between the proportion of anaemic children residing in the rural area (33%,  $n = 74$ ) and that from urban Mfou (33.6%,  $n = 94$ ). Likewise, no significant difference was observed in the prevalence of anaemia between male and female schoolchildren. However, there was a significant age-related decrease ( $p = 0.000$ ) in the prevalence of anaemia; ranging from 66.7 to 37.7% and 17.3%, respectively in under 5, 5-9 and 10-16 years old children. Additionally, children with mild to severe underweight showed increased prevalence of anaemia (43.3%) compared to healthy ones (36.7%), but the difference was not statistically significant ( $p = 0.246$ ). Likewise, those with mild to severe stunting were also more likely to be anaemic (36.9%) compared to healthy ones (31.9%). The difference also did not reach statistical significance ( $p = 0.156$ ).

### Prevalence and intensities of malaria and soil-transmitted helminths in children

Overall, 204 children (40.6%) were infected with malaria parasites, among whom, 182 (89.2%) were asymptomatic carriers and 22 (10.8%) were symptomatic cases (axillary temperature  $> 37.5^\circ\text{C}$ ). *Plasmodium falciparum* was the predominant species (96.6%) with parasite density ranging from 0-792,000 parasites/ $\mu\text{l}$  of blood. There was no significant difference in the prevalence and geometric mean parasite density in relation to sex of children and the area of residence; however, an age-related decrease in the geometric mean parasite density was observed (Table 2).

A total of 149 children (29.6%) were infected with STHs. They included: 113 cases of *A. lumbricoides* (75.8%), 79 cases of *T. trichiura* (53%) and 45 cases of co-infections with both species (30.2%). No infection with hookworm was detected. The prevalence of STHs was significantly higher in rural schoolchildren (40.6%;  $p = 0.000$ ) compared to those living in Mfou urban (20.8%). However, geometric mean density of *T. trichiura* was significantly elevated in children from urban Mfou (mean=307 epg;  $p = 0.039$ ) compared to rural school children (mean=130 epg). Children infected with *T. trichiura* had more than two fold increased chances of being underweight compared to uninfected ones (OR=2.33, 95% CI: 1.25-4.36;  $p = 0.008$ ). There was no significant association between the prevalence of anaemia and presence of STHs. However, children co-infected with malaria parasites and STHs showed increased

**Table 1.** Prevalence of malnutrition and anaemia in relation to sex of children.

Characteristic	Male		Female		Total population		P- value
	n	Prevalence (%)	n	Prevalence (%)	n	Prevalence (%)	
<b>Underweight category</b>							
Mild ( $-1.01 \leq WAZ < -2$ )	50	24.6	36	19.6	86	22.2	0.015
Moderate ( $-2 < WAZ < -3$ )	9	4.4	0	0.0	9	2.3	
Severe ( $-3 < WAZ$ )	1	0.5	1	0.5	2	0.5	
Normal ( $WAZ \geq -1.0$ )	143	70.4	147	79.9	290	74.9	
Total population	203	52.5	184	47.5	387	100	
<b>Stunting category</b>							
Mild ( $-1.01 \leq HAZ < -2$ )	68	25.3	42	17.9	110	21.9	0.051
Moderate ( $-2 < HAZ < -3$ )	21	7.8	17	7.3	38	7.6	
Severe ( $-3 < HAZ$ )	8	3.0	2	0.9	10	2.0	
Normal ( $HAZ \geq -1.0$ )	172	63.9	173	79.3	345	68.6	
Total population	269	53.5	234	46.5	503	100	
<b>Anaemia status</b>							
Anaemia (hematocrit < 33%)	88	32.7	80	34.3	168	33.5	0.701
*Mean hematocrit	269	37.1±4.9	233	36.8±4.7	502	36.9±4.9	0.544

WAZ=Weight-for-age Z-scores; HAZ=Height-for-age Z-scores; WAZ indices were calculated for children aged 3-10 years, while HAZ indices were calculated for those aged 3-16 years; \* values are arithmetic means ± standard deviation.

**Table 2.** Prevalence of malaria and geo-helminths in relation to age of children.

Characteristic	Age category (years)			Total population	P value
	3-4	5-9	10-16		
Total number of children surveyed	45	290	168	503	/
Prevalence of malaria, % (n)	42.2 (19)	40.7 (118)	39.9 (67)	40.6 (204)	0.958
Light parasitaemia*, %	26.3	50.8	74.6	56.4	
Moderate parasitaemia, %	42.1	33.1	19.4	29.4	
Heavy parasitaemia, %	10.5	15.3	4.5	11.3	
Very heavy parasitaemia, %	21.1	0.8	1.5	2.9	0.000
Geometric mean parasitaemia, % (parasites/μl of blood)	1394 (120, 792000)	508 (40, 10400)	238 (40, 10440)	435 (40, 792000)	0.004
Prevalence of STHs**, % (n)	15.6 (7)	23.8 (69)	43.5 (73)	29.6 (149)	0.000

**Table 2.** Contd.

Prevalence of <i>Ascaris lumbricoides</i> , %	13.3 (6)	18.6 (54)	31.5 (53)	22.5 (113)	0.002
Light density (1-4999 epg†), %	66.7	74.1	58.5	66.4	
Moderate density (5000-49999 epg), %	33.3	25.9	39.6	32.7	0.456
Heavy density (≥50000 epg), %	0.0	0.0	1.9	0.9	
Geometric mean parasite density, % (epg)	4301 (1248-34464)	1628 (96-48192)	2373 (96, 50496)	2045 (96, 50496)	0.307
Prevalence of <i>Trichuris trichiura</i>	6.7 (3)	11.7 (34)	25.0 (42)	15.7 (79)	0.000
Light density (1-999 epg)	100	79.4	83.3	82.3	
Moderate density (1000-9999 epg)	0.0	20.6	16.7	17.7	0.647
Heavy density (≥10000 epg)	0.0	0.0	0.0	0.0	
Geometric mean parasite density (epg)	76 (48, 96)	191 (24, 3792)	169 (24, 6408)	173 (24, 6408)	0.738
Prevalence of co-infections‡	8.9 (4)	10.7 (31)	20.2 (34)	13.7 (69)	0.010

\*Classification of malaria parasiteamia: light parasiteamia (1-499 parasites/μl of blood), moderate parasiteamia (500-1999 parasites/μl of blood); heavy parasiteamia (2000-9999 parasites/μl of blood) and very heavy parasiteamia (≥10000 parasites/ μl of blood); \*\*Stool were examined using the Kato-Katz thick smear technique; †epg=egg per gram of faeces; ‡Co-infections= concomitant presence of malaria parasites and worms eggs.

**Table 3.** Univariate and multivariate logistic regression analyses of the association between malaria infection and independent factors.

Variable		Malaria positive (n=204)	Malaria negative (n=299)	Univariate analysis (OR***, 95% CI)	p-value	Multivariate analysis (Adjusted OR, 95% CI)	p-value	p-value for trend‡
Fever*	No	182 (39.3)	281 (60.7)	1				
	Yes	22 (55.0)	18 (45.0)	1.88 (0.98-3.61)	0.056	1.80 (0.86-3.78)	0.118	
Anaemia**	No	195 (39.9)	294 (60.1)	1				
	Yes	9 (69.2)	4 (30.8)	3.38 (2.29-4.97)	0.000	2.64 (1.71-4.07)	0.000	
Underweight categories†	Normal	119 (41.0)	171 (59.0)	1				
	Mild	45 (52.3)	41 (47.7)	1.57 (0.97-2.55)	0.065	1.49 (0.90-2.48)	0.117	
	Moderate to severe	1 (9.1)	10 (90.9)	0.14 (0.01 (1.13)	0.066	0.13 (0.01-1.12)	0.064	0.046
<i>Ascaris lumbricoides</i>	No	148 (37.9)	242 (62.1)	1				
	Yes	56 (49.6)	57 (50.4)	1.61 (1.05-2.44)	0.028	1.72 (1.01-2.91)	0.043	

\*Fever: Temperature>37.5°C, \*\*anaemia: Parked cell volume or hematocrit<33%, \*\*\*OR: Odds ratio; †Mild underweight:-1.01SD≤WAZ score≥-2SD and moderate to severe underweight: WAZ<-2 SD in children aged 3-10 years; SD: standard deviation; ‡ p-value for trend for n>2 modalities.

increased chances of been anaemic, although this did not reach statistical significance (OR=1.64, 95% CI: 0.98-2.75; p=0.059). But, mean level of

haematocrit was significantly lower (mean=35.4±4.7%; p=0.000) in the latter compared to children with single *Ascaris* or *Trichuris*

infections (mean=39.7±4.1%). Table 3 shows the prevalence and intensities of malaria and STHs in relation to age of children.

infection, soil-transmitted helminths and undernutrition (underweight and stunting) in a total of 503 school children, aged 3-16 years, residing in Mfou health district, a forest area with perennial transmission of malaria, in Cameroon. Overall, malaria infection and soil-transmitted helminths (*A. lumbricoides* and *T. trichiura*) detected in these schoolchildren were associated with anaemia (malaria infection and to a certain extent malaria-helminth co-infections), undernutrition (underweight and stunting), and the area of residence (STHs).

The prevalence of malaria infection and STHs observed in these children are similar or lower compared to previous figures recorded in schoolchildren in Cameroon and elsewhere in Africa (Quakyi et al., 2000; Kimbi et al., 2005a, b; Egwunyenga and Ataikuru, 2005; Nkuo-Akenji et al., 2006; Achidi et al., 2008; Ayalew et al., 2011). In fact, in many areas where malaria infection and helminthiasis are co-endemic, schoolchildren usually harbour the heaviest burden of the infections, with associated morbidity, especially when co-infected (Mwangi et al., 2006; Achidi et al., 2008). The survey was conducted at the beginning of the rainy season when malaria transmission was more likely to be elevated. However, over the past years, there have been some fluctuations in climate, often resulting in prolonged dry season periods in the study locality, as well as elsewhere in the country.

No significant difference was observed in the prevalence of malaria infection between rural schoolchildren and those residing in the urban area of Mfou; thus, suggesting probably a similar rate of transmission between the two localities. In fact, the environment in Mfou urban (an emerging semi-urban area) is closely related to that of Mfou rural, with stagnant water bodies, streams and swampy areas that are pre-sent year round and provide breeding sites for *Anopheles* mosquitoes. Factors significantly associated with increased risk of malaria infection included: Anaemia and infection with *A. lumbricoides*; while association with mild underweight did not reach statistical significance. In fact, association between malaria infection and anaemia is well recognized as malaria parasite, *P. falciparum* lives inside the red cell, and requires haemoglobin for its development. Thus, completion of the parasite life cycle is associated with the break-down of red blood cells leading to anaemia. However, the cause of anaemia has been shown to be multifactorial, and we found a significant low mean level of haematocrit in children co-infected with malaria parasites and STHs compared to those with a single helminth infection. It is also possible that in these children with prevalent undernutrition, low dietary intake in iron, or malabsorption of iron could also contribute to anaemia. In fact, increased prevalence of anaemia was observed in undernourished children (underweight and stunted), compared to normal children, however, the difference did not reach statistical significance.

Looking at the association between malaria infection and STHs, particularly, infection with *A. lumbricoides*,

our data suggests increased susceptibility to asymptomatic malaria infection with concomitant *Ascaris* infection. In fact, there have been conflicting reports on the association between these two infections (Mwangi et al., 2006). Some studies conducted in patients with severe clinical forms of malaria (Nacher et al., 2002) or in schoolchildren (Murray et al., 1978) or children living in rural areas (Brutus et al., 2007) have suggested a protective effect of *A. lumbricoides* on malaria, while other studies have suggested a negative effect of this parasite or other worm infections on malaria (Spiegel et al., 2003; Sokhna et al., 2004; Le Hesran et al., 2004; Druilhe et al. 2005). As intensity of infection with *A. lumbricoides* seems to be important in this association as cited in these studies, we observed an increasing prevalence of malaria infection with increasing *Ascaris* load (moderate or heavy intensities), but this association was not statistically significant.

On the other hand, after adjusting for other factors, association between asymptomatic malaria infection and underweight (an indicator of both acute and chronic undernutrition) did not reach statistical significance ( $p=0.117$ ). In fact, mild underweight ( $-1.01 \leq WAZ < -2SD$ ) was associated with increased risk of malaria infection ( $p=0.065$ ), while moderate to severe underweight ( $WAZ < -2SD$ ) was instead associated with a reduced risk of malaria infection ( $p=0.066$ ). However, the limited sample size of this latter category ( $n=13$ ) did not allow us to make an interpretation of the observations. Previous studies on the relationship between undernutrition and asymptomatic malaria have often lead to controversial findings. Some studies have shown a positive association while others did not. For example, in a study conducted in Equatorial Guinean children, stunting was positively related to malaria infection, while no significant association was found with wasting and underweight (Custodio et al., 2009). Additionally, in another study conducted in Ghanaian children, Crookston et al. (2010), found no significant association between chronic undernutrition and asymptomatic malaria detected by polymerase chain reaction (PCR).

However, it should be noted that most of these studies were conducted in children aged less than 5 years, while our study included children up to 16 years, as late school entrance is common in rural primary schools in Mfou. Other factors that could explain differences observed could be the level of transmission, the study design, including the diagnostic method used for malaria (microscopy or PCR), childrens' diets and also intercurrent infections.

Moreover, infection with STHs, particularly *T. trichiura* was significantly associated with increased risk of underweight, but this did not reach statistical significance after adjusting for area of residence and age of the children ( $p=0.073$ ). Previous studies have suggested that intestinal helminthiasis compromise healthy nutrition and growth in infected children. The variety of mechanisms

used include reduced food intake due to malabsorption and/or reduced appetite, which result in higher levels of stunting (Stoltzfus et al., 1997; Stephenson et al., 2000; Crampton and Nesheim, 2002; Hotez et al., 2008; Hall et al., 2008; Brooker et al., 2010).

On the other hand, prevalence of STHs was significantly elevated in rural schoolchildren, with more than twofold increase in chance of being infected compared to children residing in urban Mfou. This is likely due to the sanitation conditions in rural areas where the majority of households do not have access to good potable water, and this offers the possibility for the spread and maintenance of these infections.

No infection with hookworm was detected in our study population and this is probably due to the low prevalence in the study area based on statistics of the clinical laboratory of Mfou district hospital, which is the referral hospital in the district. This could also be related to the fact that the Kato-Katz slides were not read immediately after preparation. It is likely that hookworm eggs deteriorate during the 15-24 h incubation period before slides reading. This likely lead to underestimation, as it has been suggested that hookworm eggs collapsed and disappeared shortly after the thick smear had cleared (Santos et al., 2005).

On the other hand, mild to severe underweight (WAZ $\leq$ -1.01SD) was significantly associated with the area of residence, sex and age of children. A sex-related increase in the prevalence of undernutrition was observed, with boys significantly more likely to be underweight or stunted, than girls. This observation has already been noted in previous studies (Stoltzfus et al., 1997; Genton et al., 1998; Wamani et al., 2007); and it has been suggested that the daily energy and micronutrients intakes of the study population could be different by sex. Also, malaria infection and geo-helminths could act as cofounders in this association, but we did not find any sex-related association with these two infections.

## Conclusion

The cross-sectional nature of our study limits the interpretation of some of the observations, but the data provide additional support to the fact that malaria infection and STHs exacerbate anaemia and undernutrition in schoolchildren of the study locality. Also, concomitant *Ascaris* infection increases the risk of malaria infection in these schoolchildren. Thus, current preventive strategies including use of long lasting insecticide treated bed nets in households and annual school-based deworming programs coupled with a nutritional component should be reinforced in this age group.

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Full Length Research Paper

## Mosquito biting and malaria situation in an urban setting in Zambia

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Unprecedented increased mosquito bites were observed in urban and peri-urban areas of Lusaka District and there was a perceived increase in malaria which necessitated a study in 2009 to determine the mosquito biting and malaria situation within urban and peri-urban settings. We analysed water bodies as sources of mosquito larvae (vegetable gardens, sewerage maturation ponds and foot paths) and, weather factors for possible effects on mosquito densities, species distribution and reviewed laboratory confirmed malaria cases and interventions implemented in the previous five years from 2009. We collected high densities of *Culex quinquefasciatus* mosquito larvae (mean; 250 per scoop) from sewerage maturation ponds overgrown with water hyacinth - an aquatic vegetation and collected large numbers of adult *Cx. quinquefasciatus* indoor (mean: 14.2, range: 2.8 to 34 per room) and up to 2,000 *Cx. quinquefasciatus* out-door in one open structure. However, we did not find adult *Anopheles* in any of these collections but we found *Anopheles* larvae along a footpath in one peri-urban location. There was no evidence of increased malaria cases despite reported increased mosquito biting but a district-wide and nationwide decline in malaria trends. A combination of factors; human and environmental, created suitable micro-habitats that increased densities of the *Culex* mosquito but not malaria vector species within urban and peri-urban settings. An intensified surveillance, monitoring and evaluation measure is vital to understanding malaria situation and delivering effective malaria interventions in different epidemiological settings.

**Key words:** Mosquito, malaria, *Culex quinquefasciatus*, larvae, water hyacinth.

### INTRODUCTION

Malaria is endemic in all the ten Provinces and has been a leading cause of mortality and morbidity in Zambia for several decades (MOH, 1988, 1992, 2000). *Plasmodium falciparum* constitutes the most predominant malaria species (98%) of all malaria infections in the Country with

98% of total infections transmitted principally by *Anopheles gambiae sensu lato* and *An. funestus* (MOH, 2010a). Malaria occurs throughout the year and transmission follows the main seasons; low transmission during the dry season from May to September and high transmission in wet hot season from November to April (MOH, 2005).

*Culex* mosquito species exist in Zambia. However, they do not constitute a major public health problem in Zambia as is the case with *Anopheles* vector species. *Culex* species

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are potential vectors of arthropod-borne viral (arboviral) infections in other ecological regions (Diaz-Badillo et al., 2011).

Zambia's national malaria control programme goal is to reduce the malaria incidence by 75% and all-cause child mortality by 20% of the 2010 baseline through a combination of interventions. The interventions being implemented are prompt diagnosis and effective treatment; Integrated Vector Management (using Long Lasting Insecticide Treated Mosquito Nets (LLINs), Indoor Residual Spraying (IRS) and environmental management in targeted areas; Malaria In pregnancy prevention; information, communication, education for Behaviour change communication and performance monitoring (MOH, 2006, 2010b). Effective delivery of these interventions has contributed to a rapid positive impact on malaria. Inpatient routine health facility data shows a 30% reduction in malaria cases and deaths between 2006 and 2008 (MOH, 2008). These findings from national routine information system corroborate population surveys which show a decrease in parasite prevalence and severe anaemia (that is, haemoglobin less than 8 g per decilitre) in children below age five years, and severe anaemia reduced from 13.8% in 2006 to 4.3% in 2008 (MOH, Malaria Indicator Survey, 2006, 2008).

Despite the national impact on the burden of malaria, communities in Lusaka reported that there was an increase in mosquito bites and perceived increased malaria cases in their residential areas from June to November, 2009. The World Health Organisation (WHO) in keeping with its core mandate of catalysing research supported the Ministry of Health (MOH) to determine the scope of the problem (WHO, 2008). The aim of this assessment was to investigate the underlying causes of the increased mosquito problem and to determine whether there was any increase in malaria cases in urban and peri-urban areas of Lusaka.

## METHODS

### Searches for mosquito larval sites

Searches for mosquito larvae were conducted in tertiary sewage ponds that is, the last ponds in the sewage treatment and settlement process; a dam, and in dug-out shallow wells within vegetable gardens in seven sites situated within urban and peri-urban residential areas of Garden Compound, Northmead, Mtendere, Kaunda Square, Matero, Ngwerere, and Chilenje South in Lusaka District (Figure 1). Of these residential areas, only one (Ngwerere) was more than 60 km away from the city centre of Lusaka. Coordinates were collected at each larval breeding site to use for mapping the areas for follow-ups. Surrounding areas in sewer ponds, disused vegetable gardening wells and a dam were searched for mosquito breeding using mosquito scoops (dippers). To scoop the mosquito larvae, the dippers were lowered within a depth of half a meter of the water bodies including; along the edges of a dam, sewer maturation ponds and garden wells. Larval densities were determined as the number of larvae per scoop.

Mosquito larvae were transferred to the Malaria Control Centre

Insectary and reared to the adult stage for identification.

### Sampling adult mosquitoes

Adult mosquitoes were collected indoor from surfaces of 10 selected houses per day in the morning hours at 06.00 to 07.00 h in five residential areas (Northmead, Mtendere, Kaunda Square, Matero, and Ngwerere). The Spray sheet mosquito collection method was used for adult mosquito collections (WHO, 1992).

These collections were placed into containers labelled with information that included; location, date, time of collection, number of people per room. The mosquito samples were transferred on the same day to an entomology laboratory at the National Malaria Control Centre, Lusaka, for species identification (WHO, 1992; Gillies et al., 1968; Edwards, 1941). Uncompleted bathrooms and toilet constructions were sampled to assess if there could be any disparity between these and other indoor densities of the culicine mosquitoes. Mean densities (average number of mosquitoes resting indoors during the day) were used as a proxy measure for assessing the "biting threat" to the people living in residential areas from houses that were randomly selected.

### Review of meteorological information

Meteorological data (maximum and minimum temperatures, rainfall and per cent relative humidity) for the period 2005 to 2009 was collected from the Meteorological Department database in Lusaka to assess weather patterns and trends, in order to ascertain any correlations with the problem of mosquito proliferation. The data is presented as average of three meteorological stations.

### Review of laboratory confirmed malaria cases

We obtained laboratory malaria confirmed data for the period 2004 to 2009 for children under the age five and above five years from a major referral centre (Matero) in Lusaka. Laboratory malaria blood microscopic examinations were conducted in accordance with the National Diagnosis and Treatment Policy (MOH, 2010b). We also analysed trends and seasonal variations of the malaria parasite prevalence on the data maintained at the National Malaria Control Centre and from National surveys conducted in the period 2004 to 2009.

### Review of malaria interventions implemented in Lusaka district

We reviewed Zambia Demographic and Health Survey (CSO et al., 2008), National Malaria Control Programme Reports and National Malaria Surveys carried out between 2004 and 2009 to gather information on malaria prevention and treatment interventions implemented in the district. Data included; ITN ownership and utilisation among different age categories (under five and 15 to 49 years pregnant women); IRS data from the National Malaria Programme reports and first line antimalarial medicine Artemether lumefantrine (AL) from National Medical Stores Limited.

### Definitions

IRS coverage is defined as proportion of households in target districts within urban and peri-urban sprayed within 12 months. Long Lasting Insecticide Treated Nets (LLINs) refers to factory pre-treated nets for malaria control.

We visited four clinics; Matero main-referral centre, Chipata,



**Figure 1.** The scatter of shallow wells in vegetable gardens within maturation sewer ponds.  
Source: Google Earth Maps.

Mtendere and Kaunda square to collect health worker perception on mosquito and malaria. At each of these health centres, five health workers, consisting of a medical doctor, a clinical officer and nurse were interviewed. The questionnaire guide requested the health worker(s) to comment on two main issues; (1) the mosquito and (2) malaria situation that is, to indicate whether there was an increase, decrease or no change regarding the mosquito and malaria situation in the past three years in their catchment areas and which antimalarial medicine was the recommended first line; its availability and efficacy. We verified availability of anti-malarials by checking the stock control cards in the pharmacy department. Each of the respondents interviewed had worked at their catchment area for at least three years.

## RESULTS

### Search for mosquito larval sites

Two hundred and fifty *Cx. quinquefasciatus* species larvae (or more per scoop) were collected under an overgrown water hyacinth; aquatic vegetation and in maturation sewer ponds (Figure 2). These *Culex* mosquito larvae were collected along the edge of the water body in three township locations. Culicine larvae were also found

in shallow wells within vegetable gardens located 5 to 10 m from sewer maturation ponds (Figure 2). However, larval counts were fewer in the garden wells (30 to 50 per scoop) than in maturation sewer ponds (250 *Culex* larvae per scoop). No *Anopheles* larvae of the malaria-carrying mosquitoes were collected in disused vegetable gardening wells, edges of a dam, stagnant water pools located between ridges, and tertiary sewer treatment ponds/sites sampled. However, scanty anopheles (ten larvae) and no culicine larvae were collected in one scoop from shallow (less than one meter deep) temporary water pool located in one peri-urban (Mtendere) along footpath, approximately five hundred meters away from a settlement site. These were reared at the laboratory; reared mosquitoes identified as *Anopheles gambiae sensu lato*.

### Sampling adult mosquitoes

A total of 1,701 adult mosquitoes were caught –door during the entire three weeks study. The mean in-door





**Figure 2.** Proliferation of the aquatic vegetation (hyacinth) in the sewage treatment ponds. The insert is a scoop showing the presence of larvae (appearing as dark spots) in the pond-water.  
Source: Investigator, Nkhuwa DCN.

#### Perceptions on mosquito and malaria situation among health workers

resting density of these adult mosquitoes was 14.2 mosquitoes per room (Table 1). 80% of the female adult mosquitoes were fully fed (engorged). All the mosquitoes captured were identified into the *Cx. quinquefasciatus* group; no adult *Anopheles* species were found in all the collections. The mean room occupancy was 3 people. A total of 321 people lived in the areas where mosquitoes were sampled. Culicine mosquitoes densities resting outdoor were too numerous to count. Approximately 2,000 adult *Cx. quinquefasciatus* were collected in a small (two metres square) bathroom adjoining a house which constituted the highest count within the five residential areas sampled. A similar high density of at least 170 culicine mosquitoes and more were collected from a small disused broken cement drain leading to treatment sewerage ponds, measuring two square metres, situated approximately 5 km from one residential area (Kaunda square) (Figure 3). Favoured adult mosquito resting sites included hyacinth vegetation in the sewage maturation ponds inside pit latrines and dug-out garden wells.

#### Review of meteorological information

Temperature, rainfall and relative humidity trends in Lusaka are shown in Figure 4. The data are averages of three Meteorological Stations (Lusaka International Airport, City Airport and Mount Makalu) collected between July 2005 to September 2009.

#### Review of laboratory confirmed malaria cases

Annual confirmed malaria cases showed a decreasing trend in Lusaka district for both children under the age of five and those above five (that is, all malaria cases) since 2004 with slide positivity at 16% in 2005 and 1.5% in 2009 (Figure 5). MOH, Malaria Indicator Survey (2006, 2008) showed a similar trend in parasite prevalence data for children under the age of five years (22%) in 2006 compared with 10% in 2008.

#### Malaria interventions in Lusaka district

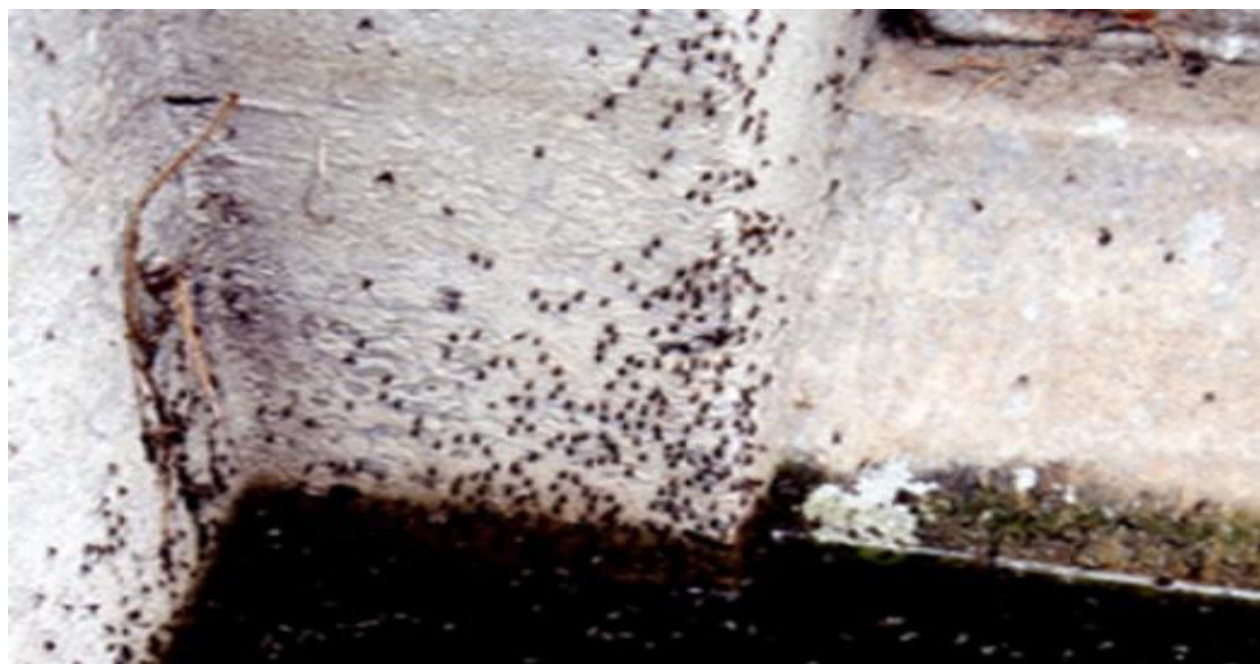
The number of formal housing structures sprayed in Lusaka

**Table 1.** Adult culicine mosquito resting densities and larvae from breeding sites in five residential areas, November, 2009, Lusaka, Zambia.

Locality (urban or peri-urban)	Mean mosquitoes collected per room	Rooms sampled	Water source	Larvae per dip
Garden (Peri-U)	34.00	20	Sewer ponds	+250
North Mead (Urban)	2.80	20	Sewer ponds	+100
Mtendere (Peri-U)	6.87	15	*	0
Kaunda Sq. (Urban)	29.68	28	Sewer ponds	+250
Chilenje (Peri-U)	4.00	14	*	0
Average	14.20			

\*No sewer ponds were available for larval searches in Mtendere and Chilenje sites. However, Anopheline larvae mosquitoes were collected in peri-urban areas in Mtendere areas from a shallow water pool. (Abbreviation; peri-U = peri-urban).

Source: Data collected by PI - FM and other investigators CS.



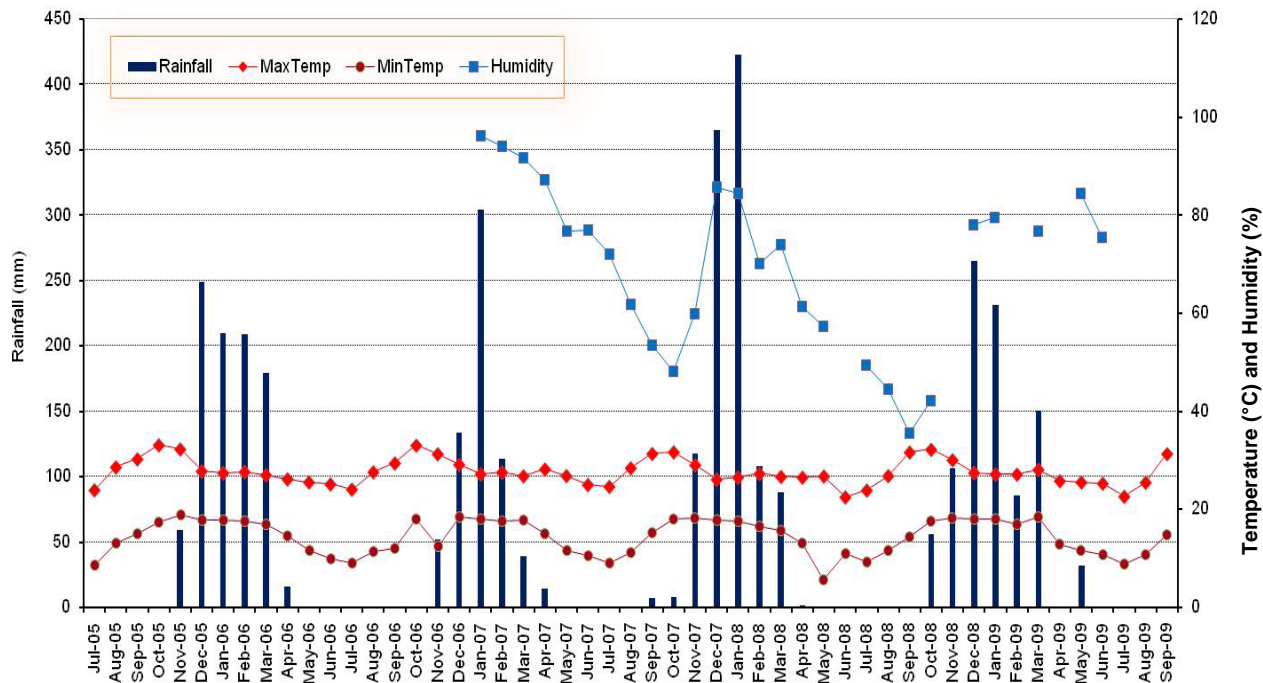
**Figure 3.** Mosquitoes' hideouts disused manholes, located five meters from sewer ponds at Kaunda Square. Adult mosquitoes appear as black spots.

Source: Principal Investigator (PI), Masaninga F.

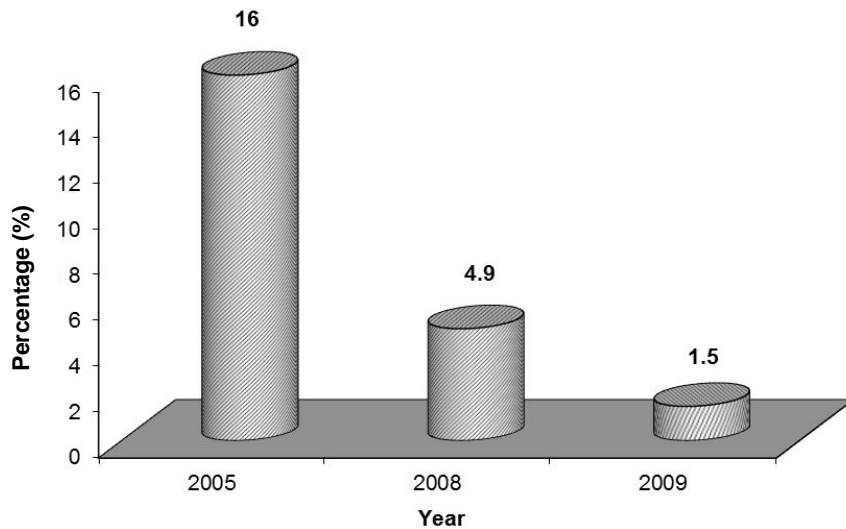
Lusaka District increased from 7,000 in 2003 to 322,000 structures in 2008 while the informal structures increased from 8,800 to 85,000. The IRS coverage in Lusaka district was 11% in 2006 and 30% in 2008. Likewise, the national IRS coverage was 6.5% in 2005 and 42% in 2008. Organochlorine (DDT) and pyrethroids insecticides (alphacypermethrin, lambdacyhalothrin and K-orthrine) have been used for indoor spraying since the re-introduction of IRS in 2003. DDT has been targeted to rough, non-plastered wall surfaces while pyrethroids targeted smooth cement-plastered surfaces. National ITNs data for the period 2006 to 2008 showed increased national household ownership from 44% (N = 2957) to 62% (N = 4,405). Likewise Lusaka district ITN use increased among children of age below five years from

15% (N = 321) to 42% (N = 363) and in pregnant women from 11 to 46%. ITN utilisation by wealth index showed a similar trend; an increase in the lowest quintile from 22% (N = 581) to 39% (N = 1,082) for children and from 21% (N = 88) to 40% (N = 103) for pregnant women. The increasing trend in ITN use was true for the highest wealth index as well; increasing from 30% (N = 408) to 40% (N = 611) in children and from 25% in 2006 (N = 75) to 49% in 2008 (N = 416) in pregnant women. National distribution of *A lumefantrine* by the Medical Store Limited averaged 2.1 million treatment courses in the period 2004 to 2009 (Figure 6). The average annual distribution of the AL treatment courses for all the age groups for Lusaka district was 291,000 (range: 292,882 to 318,574).





**Figure 4.** Recent trends in rainfall, maximum and minimum temperatures and relative humidity for Lusaka as averages of the three Meteorological stations, namely; Lusaka International Airport, City Airport and Mount Makulu between 2005 to 2009. Source: National Meteorological Department, Lusaka, Zambia.

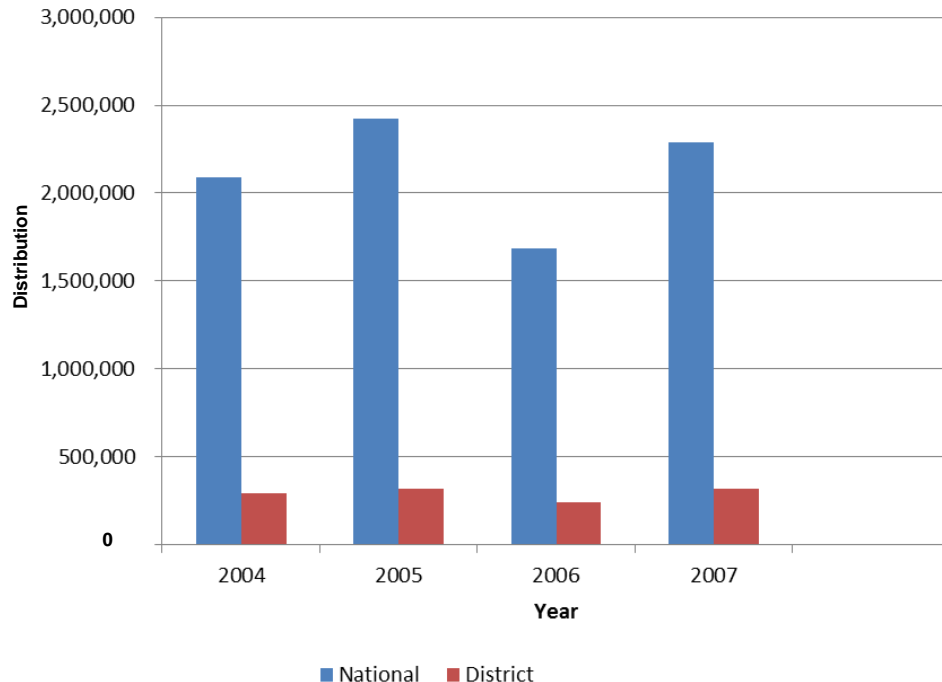


**Figure 5.** Percent slide positive tested by microscopy for *Plasmodium falciparum* malaria cases by year, Mtendere Clinic, Zambia (Number. tested = 4934 in 2005; 10,043 in 2008; 8,314 in 2009). Source: Mtendere Clinic, Ministry of Health, Lusaka Zambia.

**Perceptions on mosquito and malaria situation among health worker**

All the 20 health workers interviewed indicated that the number of mosquitoes biting them during the late afternoon

into the evenings was a record high, when compared to the past three years and high biting occurred even in the dry-cool season (June). However, interviews indicated an apparent decrease in malaria cases within their catchment health facilities (clinics) during these past



**Figure 6.** National compared to District (Lusaka) distribution of the first line (Artemether lumefantrine) 2004 to 2007.

Source: National Malaria Control Centre and Medical Stores Limited, Lusaka, Zambia.

three years. They reported Artemether lumefantrine to be the recommended first line anti-malarial medicine for the treatment of uncomplicated malaria, and that it was generally available, providing adequate therapeutic cure, with no recorded treatment failure.

## DISCUSSION

There was no evidence of increased malaria cases as perceived by the communities in the Lusaka environment, in the period under review but on the contrary, a declining trend of *P. falciparum* malaria slide positivity occurred in Lusaka between 2004 and 2009. This finding corroborates with the nation's Ministry of Health (MOH) Management Information System. This routine system has recorded declines in-patient malaria cases and deaths of 46 and 47%, respectively between 2006 and 2008. Similarly, WHO reported national declines of 55 and 60% in 2008, when the 2008 malaria data was compared to the 2001 to 2002 reference point (WHO, 2009; WHO Zambia Country Office, 2009); this decline continued to be reported (WHO, 2010). Nationally, representative malaria surveys report a general decline in *P. falciparum* parasite prevalence in children of age 0 to 59 in Zambia (MOH, Malaria Indicator Survey, 2006, 2008).

This assessment on malaria-mosquito situation in Lusaka showed scarce anopheline malaria vector breeding in the areas sampled. The observed paucity of

malaria-transmitting-anopheles vectors in the city corroborates the local declines in the malaria situation, particularly for the month of November when sampling was conducted. The apparent absence of adult *Anopheles* malaria vectors show changes in the epidemiology of vectors that may be related to increased city expansion that has occurred in the last four years. This may have contributed significantly to the destruction of mosquito breeding sites. Furthermore, interventions such as ITNs, IRS and Artemisinin combination therapy (ACTs) for treatment of uncomplicated malaria have significantly been scaled-up above the national and regional targets (Abuja Declaration and the Plan of Action, 2000; MOH, 2010a; WHO, 2009; Barnes et al., 2009). Continued vector and disease surveillance are vital to guide effective delivery of preventive, diagnostics and treatment interventions (MOH, 2002, 2010b; WHO, 2010; 2004).

High densities of culicine mosquitoes were observed outdoors in sewerage treatment ponds overgrown with water hyacinth, disused holding holes, water drains and in pit latrines. Breeding sites were situated close to homes; within a distance of 10 to 20 m. Socio-economic activities, including vegetable gardening located 5 to 10 m away from the sewage ponds contributed to the proliferation of mosquito populations during the dry season, when these gardening or vegetable growing activities increased. Health workers at different health facilities confirmed the unprecedented increased numbers of

mosquitoes biting people that were experienced starting in the afternoon around 16.00 to 17.00 h which we identified as *Cx. quinquefasciatus*. *Cx. quinquefasciatus* transmits filariasis and viral infections in African and other regions (WHO, 2001; Service, 1986).

WHO disease distribution maps show the occurrence of filariasis of up to 11% prevalence on the Copperbelt Province and 6% in the North-Western Province of Zambia (WHO Zambia Country Office, 2009). Therefore, our findings highlight the need to develop comprehensive vector control strategies for malaria and other mosquito-borne diseases and to enforce relevant Public Health Act to promote a healthier environment (Mosquito extermination Act, CAP 537, 1964; Laws of Zambia, 1964).

Eighty percent of the female culicine mosquitoes collected indoors were blood-fed. Even though no tests were done to determine the source, the blood was most likely human, given the paucity of domestic animals in the area. That the public complaints about mosquitoes occurred in winter and daytime biting was a clear indication that the biting was unlikely to be related to malaria.

In the 1960s and early 1970s, there was limited breeding of mosquitoes in the city of Lusaka, with no or occasional bites around households; malaria was also a notifiable disease (MOH, 2002). Intensive surveillance ensured early response to even a single reported case in this and other cities. During this period, there were fewer unplanned settlement residential areas and relevant statutory instruments were enforced through the local government which obliged each household to maintain hygiene standards and to prevent and control mosquito breeding. However, mosquito breeding and malaria cases increased during the late 1970s to 1990s due to worsening socio-economic factors concomitant with rapid urbanisation; this urbanisation has continued to expand in the major cities in Zambia (MOH, 1992, 2000; UN Habitat, 2012).

Weather data in this study suggest an increase in total seasonal rainfall for 2007 to 2008 and 2008 to 2009 seasons compared to the preceding years. However, there was no apparent change in temperature patterns. These and meteorological data on relative humidity were insufficient to make inference on mosquito occurrence. The changing malaria epidemiology and sporadic increases in culicine mosquito density, low numbers of both confirmed malaria cases and malaria vectors necessitate effective interventions in Lusaka including; Integrated Vector Management, Systematic monitoring of vector and malaria case investigation, strengthened community participation and adherence to legal provisions to prevent mosquito breeding and malaria proliferation in the city.

## Conclusion

Decreasing malaria prevalence in Lusaka environment following effective anti-vector measures (IRS and ITNs), diagnosis and malaria treatment necessitates an

intensification of surveillance, monitoring and evaluation to sustain the impact on the disease but also to strengthen understanding of the malaria-mosquito epidemiology to curb future proliferation of nuisance mosquitoes.

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