

The Impact Water, Sanitation and Hygiene Infrastructures Have on People Living with HIV and AIDS in Zimbabwe

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1. Introduction

Acquired Immune Deficiency Syndrome (AIDS) emerged in the 1980s as the most terrifying epidemic of modern times. AIDS as a disease is caused by a virus called the Human Immunodeficiency Virus (HIV). The total number of people living with HIV worldwide in 2009 was 33.3 million (UNAIDS, 2009). Sub-Saharan Africa continues to be the hardest hit by the global HIV and AIDS epidemic with an estimated 22.5 million infected people in 2009. AIDS was first reported in Zimbabwe in 1985 and by the beginning of the 1990s, around 10% of the adult population were thought to be infected with HIV (UNAIDS 2005). The estimated adult HIV prevalence rate (aged 15-49) during 2009 in Zimbabwe was reported to be 14.3% (UNICEF webpage).

Presently very little data is available on how water, sanitation and hygiene infrastructures are affecting the lives of people living with HIV and AIDS (PLWHA) in Zimbabwe. Literature has identified a series of linkages between water, sanitation and hygiene and HIV and AIDS (USAID/WSP, 2007). According to UNICEF (2005), a hygienic environment, clean water and adequate sanitation are key factors in preventing opportunistic infections associated with HIV and AIDS, and in the quality of life of people living with the disease. PLWHA are more susceptible to water-related diseases than healthy individuals, and they become sicker from these infections than people with healthy immune systems (UNICEF, 2005).

The urban areas of the Bulawayo metropolitan province of Zimbabwe consist primarily of high-density sub-urban areas and peri-urban settlements with municipal treated water supplies. Some peri-urban areas have unprotected water sources (rivers) and limited or totally inadequate sanitation. Although communal taps serve most peri-urban settlements, critical water shortages force these communities to rely on water from boreholes, unprotected wells and rivers for domestic use. Most of these households use pit latrines and some use the bush. Households in the rural areas mainly rely on boreholes as water sources, with a few households having piped water supply from Zimbabwe National Water Authority (ZINWA). This made the Bulawayo metropolitan the ideal study area to obtain representative data about the possible impact water, sanitation and hygiene infrastructures can have on the health of PLWHA in Zimbabwe.

Diarrhoeal diseases are the most common opportunistic infections experienced by PLWHA in Africa and elsewhere. Most of these diarrhoeal infections are either water borne or water washed. Patients with HIV infection or AIDS are commonly affected by gastrointestinal infections, with diarrhoea as the most common presentation (Janoff and Smith, 1988). Diarrhoea occurs in 30-60% of AIDS patients in developed countries and in about 90% of AIDS patients in developing countries (Framm and Soave, 1997). Enteric pathogens that cause diarrhoea include bacteria, parasites, fungi and viruses (Mitra et al., 2001). *Escherichia coli* (*E. coli*) are commonly used as an indicator of faecal pollution in water, indicating the possible presence of other bacterial pathogens that could have been shed into the water source. Although *E. coli* is used as an indicator it also has the capability of contributing to the diarrheal load in an area. Five classes of *E. coli* bacteria that cause diarrhoeal diseases are now recognised: enterotoxigenic *E. coli* (ETEC), enteroinvasive *E. coli* (EIEC), enterohaemorrhagic *E. coli* (EHEC), enteropathogenic *E. coli* (EPEC) and enteroaggregative *E. coli* (EAEC). Each class falls within a serological subgroup and manifests distinct features in pathogenesis and subsequent diarrhoea (Todar, 2002). The importance of *E. coli*, both as an indicator organism and as a diarrhoeagenic pathogen is well documented. The aim of this study is therefore to determine the impact water, sanitation and hygiene infrastructures and their associated health risks facing people living with HIV and AIDS through detection of pathogenic *E. coli* in domestic drinking water and on sanitation facilities in the and around Bulawayo in Zimbabwe.

2. Materials and methods

2.1 Ethical consent

Registration of the project and ethical clearance was obtained from the University of Venda's Health, Safety and Research Ethics Committee. The Medical Research Council of Zimbabwe and the City of Bulawayo, through the Director of Health Services provided the necessary authority to proceed with the study.

Staff at Opportunistic Infections Clinics (OICs) was used to identify the HIV and AIDS patients and only those that volunteered and signed consent forms were included in the study. The consent forms were translated into the vernacular language of the patient.

2.2 Data collection using questionnaires

Structured questionnaires were used to obtain information regarding household demographics, water sources used, water collection practices; time spent collecting water, water storage practices, costs involved in water and sanitation services, hygiene practices associated with sanitation and the level of hygiene understanding in each household.

2.3 Study area

The Bulawayo metropolitan in Zimbabwe was chosen as the study site for the project. A total of 414 households with individuals living with HIV and AIDS from high density suburban areas (n=150 households), peri-urban areas (n=121 households) and rural villages (n=142 households) in and around Bulawayo were included in this study.

2.4 Sample collection

Samples were collected aseptically from household water storage containers in sterile 1 litre plastic bottles and transported on ice to the laboratory for further analysis. Toilet seats were



Fig. 1. Map of Zimbabwe showing the location of the study area

swabbed using sterile methods and the swab placed into sterile containers with 100 ml phosphate buffered saline (pH 7.4) (PBS) and transported on ice to the laboratory for further analysis. Hands of 20% of the people living with HIV and AIDS in each of the three areas included in the study, were swabbed and the swabs placed into sterile containers with 100 ml PBS and transported on ice to the laboratory for further analysis.

2.5 Determination of total coliform bacteria and *Escherichia coli*

Total coliforms and *E. coli* were detected using the Colilert® Quanti-Tray/2000 System manufactured by IDEXX and supplied by DEHTEQ. Appropriate dilutions of the samples were made according to the procedures described by the manufacturers (IDEXX). The trays were incubated at 35°C for 18 to 22 hours and the positive (yellow and fluorescing) wells and *E. coli* bacteria were counted and converted into the most probable number of total coliforms and *E. coli* present in samples, using tables and formulae provided by the manufacturer. Included in the experiment were positive and negative controls (distilled water, *E. coli*, *Klebsiella* and *Pseudomonas*).

Selection of potential *E. coli* bacteria was made of fluorescing wells that could clearly be counted as the actual indicator in the selective growth media. The contents of positively identified *E. coli* wells were collected using 1 ml syringes and kept in clearly marked cryotubes at -70°C. A multiplex PCR method was performed according to Omar et al., (2010). The five diarrhoeagenic strains of *E. coli* and a commensal *E. coli* strain that were employed in the study are shown in Table 1.

All strains were obtained from the South African National Health Laboratory Services (NHLS) (2010), and stored at -70°C in a mixture of Plate Count Agar (PCA) with 10% (v/v) glycerol. The strains were routinely cultured on PCA and incubated under aerobic conditions at 37°C. Liquid cultures of the bacteria were obtained by inoculating 5 ml Luria-

Bacterial strain	Reference number	Genes targeted
Enterotoxigenic <i>Escherichia coli</i> (ETEC)	H10407	<i>Mdh, Lt, St</i>
Enteropathogenic <i>E. coli</i> (EPEC)	B170	<i>Mdh, eaeA</i>
Enterogaagregative <i>E. coli</i> (EAEC)	3591-178	<i>Mdh, Eagg</i>
Enterohaemorrhagic <i>E. coli</i> (EHEC)	C4193-1	<i>Mdh, eaeA, stx1, stx2</i>
Enteroinvasive <i>E. coli</i> (EIEC)	C316-58	<i>Mdh, ial</i>
Commensal <i>E. Coli</i>	Field isolate, API20E confirmed	<i>Mdh</i>

Table 1. Bacterial strains used for the experimental work

Bertani (LB) media in test tubes for 16 hours while rotating at 200 rpm at 37°C. Genomic DNA isolation was performed on 2ml of each sample using an adapted protocol as described in Omar et al., (2010). All buffers used, were prepared as described by Omar et al (2010).

2.6 DNA extraction and multiplex polymerase chain reactions (PCR)

The samples were centrifuged at 12,000xg for 2 minutes at room temperature to pellet any bacterial cells present. After removing the supernatant, each pellet was dissolved in 700 µl extraction buffer and left for 10 min at 37°C, after which 250 µl of 99% ethanol was added and thoroughly mixed. This was left for 10 minutes at 56°C, after which 40 µl celite suspension was added, mixed and left for 10 minutes with occasional mixing. Pellets were again collected by centrifugation at 12,000xg for 30 seconds at room temperature (the supernatant removed). The pellets were washed twice with 500 µl washing buffer. This was followed by two 70% (v/v) ethanol wash steps. The pellets were dried at 56°C for 10 minutes, mixed with 100 µl TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0), heated for 10 minutes at 56°C and briefly centrifuged to pellet the celite. The DNA containing supernatant was removed and the elution repeated once as described. All PCR reactions were performed in BIORAD Mycycler™ Thermal cycler as described by Omar et al., (2010).

DNA was analysed on a horizontal agarose slab gel [1% (w/v)] with ethidium bromide (0.5µg/ml) in TAE buffer (40 mM Tris acetate; 2 mM EDTA, pH 8.3). Electrophoresis was done for one hour in an electrical field strength of 5.9 V.cm⁻¹ gel and DNA was visualised with UV light (Gene Genius Bio Imaging system, Vacutec). The relative sizes of the DNA fragments were estimated by comparing their electrophoretic mobility with that of the standards run with the samples on each gel. Gene ruler (Fermentas) was used as standards.

2.7 Data analysis

The data from the questionnaires and microbiological data on commensal and pathogenic *E. coli* strains was coded and collated before being entered into an MS Excel spread sheet. The data was imported into the Stata Release 8.0 statistical software package for cleaning and editing. For categorical data, frequencies of occurrence of response were calculated. For numerical variables the data was summarised using the arithmetic, geometric and harmonic means and a corresponding 95% confidence interval. Testing was done at the 0.05 level of

significance. All statistical analyses were done by Professor Piet Bekker from the Medical Research Council Statistical Unit in Pretoria, South Africa.

3. Results and discussion

3.1 Age and gender distribution of study population

As indicated in Table 2, the average family size of the urban household was 5.2 people, with females constituting 57% and males 43% of the urban study population. There was an average of 2.2 males and 3 females in the urban households. The peri-urban household average family size was 4.7 people, with females being 54% and males 46% of the peri-urban study population. There was an average of 2.5 females and 2.2 males in the peri-urban households. In rural areas the average family size was 5.3 people; females formed 54% and males 46% of the household composition. There was an average of 2.9 females and 2.4 males in the rural household. The urban and rural family sizes were the same, with the peri-urban family size being slightly smaller, due to a number of single member families found in peri-urban areas. In all areas there were more females than males in the households; the peri-urban and rural household composition was the same, whilst the urban household composition had a larger difference between the female and male members of households.

In all areas the children aged between 5 years and 12 years were the largest age-group. There were significantly less men in the age group 41 to 50 years (15.5%) in rural areas compared to peri-urban (23.1%) and urban (25.3%) areas. This may be due to rural to urban migration of this age group in search of employment. In the age group 61 to 70 years there was a significantly higher percentage of males in rural areas (10.6%) compare to urban and peri-urban areas (3.3% in both areas). This again may be related to migrant work, with this age group having returned from urban areas where they had been working.

3.2 Age and gender distribution of PLWHA

Most of the PLWHA were in the age groups 31 to 40 years and 41 to 50 years. These results agree with household surveys in 28 sub-Saharan Africa which revealed that in all but five countries peak HIV prevalence occurs between the ages 30 and 34 for women and in the late 30s and early 40s or men (Macro International, 2008). These are the sexually active members of society and the results of this study showed that the HIV in Bulawayo could be predominantly sexually transmitted.

Of the one hundred and fifty PLWHA from urban areas one hundred and nine (73%) were female and forty-one (27%) were male. One of the participants was in the age group 16 to 22 years (0.7%); twelve were in the age group 21 to 30 years (8%); sixty-one were in the age group 31 to 40 years (40.7%); fifty-six were in the age group 41 to 50 years (37.3%); fifteen were in the age group 51 to 60 years (10%); three were in the age group 61 to 70 years (2%) and two were above 70 years of age (1.3%).

In peri-urban areas, twelve (9.3%) of the one hundred and twenty-two participants were in the age group 21 to 30 years; fifty-two (42.6%) were in the age group 31 to 40 years; forty-three (35.2%) were in the age group 41 to 50 years; eleven (9%) were in the age group 51 to 60 years and two (1.6%) were in each of the age groups 61 to 70 years and above seventy years of age.

Age in years	<u>Urban</u> Average family size = 5.2 Female = 57% Male = 43%			<u>Peri-urban</u> Average family size = 4.7 Female = 54% Male = 46%			<u>Rural</u> Average family size = 5.2 Female = 54% Male = 46%		
	F (n)	M (n)	Per age (%)	F (n)	M (n)	Per age (%)	F (n)	M (n)	Per age (%)
Infant 0-1	5	6	11 (2%)	10	6	16 (3%)	13	7	20 (3%)
Toddler >1-5	31	40	71(9%)	27	24	51 (9%)	39	31	70 (9%)
Child >5-12	71	69	140 (18%)	65	67	132 (23%)	59	90	149 (20%)
Adolescent >12-16	60	40	100 (13%)	26	17	43 (8%)	34	36	70 (10%)
Adolescent >16-20	57	38	95 (12%)	21	25	46 (8%)	39	33	72 (10%)
Adult 21-30	58	43	101 (13%)	40	33	73 (13%)	50	43	93 (13%)
Adult 31-40	75	33	108 (14%)	49	38	87 (15%)	55	31	86 (12%)
Adult 41-50	50	38	88 (11%)	40	28	68 (12%)	46	22	68 (9%)
Adult 51-60	23	13	36 (5%)	11	12	23 (4%)	24	25	49 (7%)
Adult 61-70	13	5	18 (2%)	8	4	12 (2%)	15	15	30 (4%)
Adult >70	4	6	10 (1%)	8	7	15 (3%)	16	8	24 (3%)
Total	447	331	684 (100%)	305	261	566 (100%)	390	341	731 (100%)

Table 2. Demographics of study population

In rural areas, six (4.3%) of the one hundred and forty-two participants were in the age group 16 to 20 years; twenty-five (17.7%) were in the age group 21 to 30 years; forty-six (32.6%) were in the age group 31 to 40 years; thirty-nine (27.7%) were in the age group 41 to 50 years; twenty-one (14.9%) were in the age group 51 to 60 years and four (2.8%) were in the age group 61 to 70 years.

3.3 CD4 counts of PLWHA

The cells mostly affected by HIV when it infects humans are the CD4 cells. After having been affected for a long time the number of CD4 cells decreases indicating that the immune system is being weakened. Normal CD4 counts are usually between 500 and 1 600 (AIDS InfoNet, 2010). An HIV positive person with CD4 count of less than 200 is considered to have AIDS, and preventive therapy should be started even if the person has no symptoms

(Lab Test Online, 2009). Of late most health care providers begin antiretroviral therapy (ART) when the CD4 count goes below 350 (AIDS InfoNet, 2010).

The CD4 count results of all the 150 people living with HIV and AIDS in urban areas were obtained (Table 3). The geometric mean for the CD4 count results was 221. In peri-urban

HIV and CD4 count tests	Urban	Peri-urban	Rural
Where first HIV test was done	n = 150	n = 121	n = 142
New Start Centre	70 (46.7%)	59 (48.8%)	64 (45.1%)
Local clinic	49 (32.7%)	48 (39.7%)	51 (35.9%)
Hospital	26 (17.3%)	13 (10.7%)	27 (19.0%)
Private Doctor	1 (0.7%)	1 (0.8%)	0 (0%)
Other	4 (2.6%)	0 (0%)	0 (0%)
Where first CD4 count test was done	n = 149	n = 121	n = 142
Not done	0 (0%)	0 (0%)	1 (0.7%)
New Start Centre	8 (5.4%)	5 (4.1%)	7 (4.9%)
Local clinic	78 (52.4%)	49 (40.5%)	17 (12%)
Hospital	47 (31.5%)	20 (16.5%)	90 (63.4%)
Private Doctor	13 (8.7%)	0 (0%)	1 (0.7%)
Other	3 (2%)	47 (38.9%)	26 (18.3%)
Frequency of follow-up tests	n = 150	n = 121	n = 133
Not done	0 (0%)	3 (2.5%)	0 (0%)
Once a year	57 (38%)	16 (13.2%)	41 (30.8%)
Every 6 months	21 (14%)	5 (4.1%)	8 (6%)
Every 3 months	6 (4%)	3 (2.5%)	6 (4.5%)
Not done	43 (28.7%)	92 (76%)	77 (57.9%)
Other	23 (15.3%)	2 (1.7%)	1 (0.8%)
Last CD4 count test	n = 149	n = 121	n = 114
Not done	2 (1.34%)	47 (38.84%)	0 (0%)
Within last month	38 (25.5%)	72 (59.5%)	21 (18.4%)
>1 month, <3 months	13 (8.7%)	13 (10.7%)	23 (20.2%)
3 to 6 months	10 (6.7%)	4 (3.3%)	13 (11.4%)
6 to 12 months	32 (21.5%)	14 (11.6%)	24 (21.1%)
>12 months	56 (37.6%)	18 (14.9%)	33 (28.9%)

Table 3. Details of places of testing and or frequency for HIV and CD4 tests

areas 119 PLWHA out of 121 in the study had their CD4 count results recorded and the mean was 224. All the 141 PLWHA in rural areas had their CD4 count results obtained and the mean was 235. The mean CD4 count in all the areas was below the normal CD4 count of between 500 and 1 600, however it was above 200, at which one is said to have AIDS (AIDS InfoNet, 2010).

PLWHA that had taken their first test for HIV at a New Start Centre were 47% in urban areas, 49% in peri-urban and 45% in rural areas. Those that were first tested for HIV at their local clinic were 33% in urban areas, compared to 40% in peri-urban areas and 36% in rural areas. Those that had their first test for HIV done at a hospital were 17% in urban areas, 11% in peri-urban areas and 19% in rural areas. The rest (3% in urban areas and 1% in peri-urban areas) had been first tested for HIV at a private doctor or some other testing facility.

The first CD4 count tests were done at the local clinic for 52% of PLWHA in urban areas, 41% of those in peri-urban areas and 12% of those in rural areas. Those that had the first CD4 count test done at a hospital were 32% in urban areas, 17% in peri-urban areas and 63% in rural areas. CD4 tests done by private doctors were 9% in urban areas and 1% in rural areas. Some PLWHA (2% in urban areas, 39% in peri-urban areas and 18% in rural areas) had their first CD4 count test done as special request because they had been commenced on treatment using the WHO staging without the CD4 count test.

After the initial CD4 count test getting a follow-up test was not easy, and most of the PLWHA had not had regular follow-up CD4 count tests. In urban areas 29% had not done a follow-up CD4 count test, compared to 76% in peri-urban areas and 58% in rural areas.

Those that had a CD4 count test done once every year were 38% in urban areas, 13% in peri-urban areas and 31% in rural areas. Those that had a follow-up test every 6 months were 14% in urban areas, 4% in peri-urban areas and 6% in rural areas. Those that a follow-up test every 3 months were 4% in urban areas, 2.5% in peri-urban areas and 6% in rural areas. In urban areas 15% had not had regular follow-up CD4 count tests, compared to 1% in peri-urban and rural areas.

3.4 Sanitation in study population

The Urban Councils Act and the municipal bylaws require all houses to have a flush toilet before being occupied. These provisions by and large complied with in the urban areas. Problems arose during periods of water unavailability when the households had to resort to pouring water to flush these toilets. Being situated in a water scarce region, Bulawayo needs a relook into the appropriate toilet technology for the city. A technology that would not be dependent on water would best suit the city. There is no specific policy on sanitation for peri-urban areas due to their semi-formal and in some cases informal nature. However, as in rural areas, the ventilated improved pit (VIP) latrine offers an appropriate option. The guidelines for rural sanitation recommend a VIP latrine for each household.

In urban areas 98% of the households had a water closet connected to a public sewer, the rest did not have toilet facilities and used the bush. In peri-urban areas 48% of the households had a VIP latrine, 2% had a water closet connected to a septic tank and 1% had a substandard pit latrine. The remaining 49% of the households had no toilet facilities and used the bush. In rural areas 66% of the households had VIP latrines, 15% had pit latrines with a slab and 4% had pit latrines without a slab. One household had a water closet connected to a septic tank. The remaining 14% of the households had no toilet facilities and used the bush.

Approximately 98% of households in the urban areas had access to improved sanitation; however, 49% of households in peri-urban areas and 34% in rural areas did not have access to an improved sanitation facility. The need for access to improved sanitation is critical for

HIV and AIDS patients. Nearby latrines are necessary for weak patients. In some patients, diarrhoea is chronic, further weakening them. In order for HIV infected people to remain healthy as long as possible and for people with AIDS to reduce their chances of getting diarrhoea, adequate sanitary facilities are of the utmost importance (Kamminga and Wegelin-Schuringa, 2003). The hygiene infrastructures were generally inadequate, especially with regards to hand washing facilities.

3.5 Water in study population

Zimbabwe does not have national standards for water accessibility; the SPHERE project recommended standard of 500m to a water point was therefore used (World Vision, 2006). Whenever water was available it was easily accessible from taps within the house (67%) or within the yard (33%) in urban areas. In peri-urban areas most of the households collected water from communal standpipes (70%) and boreholes (5%) to their homes over distances that varied from less than 500m to more than 2km. Up to 25% of the households collected water from unprotected wells or springs. Those that collected water from a water source more than 500m away from their homes were 9% of the households in peri-urban areas and 27.5% in rural areas. Table 4 summarises information on water availability. Travelling more than 500m to a water source is likely to be tedious especially to young children, the elderly and those weakened by illness. In peri-urban and rural areas containers of up to 25 litres were used to collect water and were mainly carried on the heads. This was a considerable strain especially for the vulnerable people. Whilst water sources were accessible to most households in all the three areas, the unavailability of water was a major constraint, especially in urban areas where 60% of the households reported that water was not always available. In peri-urban areas 36% of the households and 13% in rural areas said water was not always available. During the period August to September 2008, in urban areas and peri-urban areas supplied with municipal piped water was available only two days a week and in some cases only for a few hours. Some of these communities had gone for more than a month with intermittent water supplies. The intermittent water supply meant that even though water was piped to the house it had to be collected and stored for those periods when it was not available from the tap.

This resulted in increased water handling with a subsequent increase in opportunities for contamination (USAID, 2006). Unsanitary and inadequately protected water containers could contribute to the contamination of water at the point-of-use (Dunker, 2001). Inadequate storage conditions have been shown to result in the increase in the number of some microorganisms such as heterotrophic bacteria and total coliform bacteria over time (Reiff et al., 1996). Potable water is critical for PLWHA; they need it for taking antiretroviral (ARV) medication, bathing, washing soiled linen and clothing and for essential hygiene, which reduces exposure to opportunistic infections (WRC, 2005).

Various factors affected the availability of water in urban and peri-urban areas during different periods of the study. At one point the major supply dams had dried up resulting in water shortages. There was also a time when the municipality was reported to have run out of water treatment chemicals, and were unwilling to distribute water that had not been fully treated. In all the areas solving the water problems being experienced was taking quite long, with 39% in urban areas, 83% in peri-urban areas and 77% in rural areas having reported that it took more than a month to have the problems fixed. During these periods of water being unavailable from the usual sources 5% of households in urban areas collected water from unprotected wells, compared to 26.5% in urban areas and 10.5% in rural areas. Resorting to unsafe water sources puts the PLWHA at a risk of contracting water-borne diseases.

Water supplies	Urban % (n=150)	Peri-urban % (n=121)	Rural % (n =142)
Source			
Piped water into dwelling	66.67	0	2.82
Piped water to yard	32.67	0	11.97
Public tap/standpipe	0	70.25	23.94
Tubewell/borehole	0	4.96	50
Protected well	0	0	4.23
Unprotected dug well	0.67	23.97	6.34
Protected spring	0	0	0
Unprotected spring	0	0.83	0
Distance from home			
0 - 50m	99.33	23.97	21.83
>50 -100m	0	32.23	21.13
>100 - 200m	0.67	16.53	16.2
>200 - 300m	0	7.44	6.34
>300 - 400m	0	2.48	1.41
>400 - 500m	0	8.26	5.63
>500m - 1km	0	4.13	15.49
>1km - 2km	0	3.31	10.56
>2km	0	1.65	1.41
Time taken to water source and back			
<30 min	0.67	73.55	51.41
30 min - 1hr	0	16.53	27.46
>1hr	0	9.92	4.23
Water on premises	99.33	0	16.9
Frequency of unavailability			
	n=148	n=44	n=18
Always available	39.19	0	0
Everyday	5.41	11.36	38.89
Every other day	3.38	4.55	11.11
Twice a week	19.59	13.64	38.89
More than twice a week	25	47.73	5.56
Other	7.43	22.73	5.56
Time taken to fix problem			
	n=148	n=36	n=13
No problem	38.51	0	0
On the same day	8.11	2.78	0
1 day - 3 days	8.73	2.78	0
>3 days - 5 days	0	2.78	7.69
>5 days - 1 week	0	0	0
>1 week - 2 weeks	0	2.78	0
>2 weeks - 1 month	2.7	5.56	15.38
>1 month	39.19	83.33	76.92

Table 4. Details of sources of water supplies, water accessibility and availability

In urban areas under normal circumstances water was available on the premises and no collection times were applicable. However during periods of water shortages water had to be collected from alternative sources such as boreholes (31%) or water tankers supplied by the municipality (61%). During these time 79% of the respondents said they collected water in the morning, 23% at midday, 35% in the afternoon and 48% said they collected water in the evening.

In 3% of the households it was the responsibility of girls to collect water. Women were responsible for water collection in 30% of the household whilst men were responsible for water collection in 4% of the household. In 10% of the households women and girls were responsible for water collection. Women and boys collected water in 1% of the households and in another 1% it was women and children. The PLWHA collected water in 76% of the households. The main reasons why the PLWHA did not collect water were because they were too weak to do so (67%) or as men they did not consider it their chore (28%).

In peri-urban areas water was collected in the morning in 92% of the households, at midday in 6%, in the afternoon in 20% and in the evening in 36% of the households. Collection was done more than once a day in several households. Women were responsible for water collection in 42% of the households compared to 14% of the households where men were responsible. In 15% of the households women and girls were responsible for water collection. In 8% of the households all family members collected water. In 3% of the households it was women and children who collected water. Women and boys collected water in 2% of the households. The PLWHA collected water in 87% of the households. Where they did not collect water it was mainly because they were too weak to do so (56%) or they were ill at that moment (19%) or as men they did not consider water collection as their chore (19%).

In rural areas water was collected in the morning in 99% of the households, at midday in 17%, in the afternoon in 26% and in the evening in 45% of the households. In most households water was collected more than once a day. Women were responsible for collecting water in 54% of the households. In 10% of the households it was women and girls who collected water. Men were responsible for collecting water in 6% of the households. In 3% of the households all family members collected whilst in another 3% it was women and boys who collected water. Boys collected water in 2% of the households, and in another 2% of the household it was children who collected water. Women and children collected water in 1% of the households. In 81% of the households the PLWHA collected water. The reasons for not collecting water were that they were too weak to do so (46%) or they were ill at that moment (31%) or being men they did not consider it their chore to collect water. In all the areas more women than men were responsible for water collection, and 28% of men who did not collect water considered collecting water not to be one of their chores. This indicated that water collection to some extent remains a gender issue. Even though the majority of the PLWHA in all areas collected water, the fact that some did not do so because they were too weak to do so or they were ill at the time shows their vulnerability if water is not easily accessible.

In urban areas in 29% of the households water for drinking was boiled before it was consumed, compared to 18% in peri-urban areas and 17% in rural areas. The households where drinking water was disinfected with chlorine before consumption were 24% in urban areas, 21.5% in peri-urban areas and 23% in rural areas (Table 5). The disinfectant, in the form

of *Aquatabs* (167mg sodium dichloroisocyanurate) was supplied either by the municipality or by some NGO supporting the PLWHA. Allowing the water to stand and settle was used by 8.3% of the peri-urban households, 4.9% of rural households and 0.7% of urban households.

Although the water was kept for long periods of time under sub-optimal conditions, the majority of households did not treat the water before consuming it. Water stored in the home tends to be more contaminated than water at the collection point, suggesting that substantial contamination takes place during transport, storage and use. Those households collecting water from communal supplies must, of necessity transport water to their homes and store it. Those households with water on the premises may have intermittent or poor quality water.

Proper handling and storage and household-level disinfection is therefore necessary to maintain the quality of water (USAID, 2006). It has been observed in a wide variety of settings that water quality improvements at the point-of-use are likely to have a positive impact on health at all levels of the water supply system (USAID, 2006). There are physical and chemical treatments available as interventions to improve the quality of water at the point of use (Sobsey, 2002).

Treatment method	Urban % (n=150)	Peri-urban % (n=121)	Rural % (n=142)
Boiling	29	18	17
Bleach/chlorine	24	21.5	23
Straining with cloth	4	0	0
Filter (sand, ceramic etc)	0	0	1.4
Solar disinfection	0	0	0
Stand and settle	0.7	8.3	4.9

Table 5. Domestic pre-treatment of drinking water

3.6 Total coliform and *Escherichia coli* counts

In urban areas 77% of the hands swabs from PLWHA were positive for total coliforms, compared to 83% and 72% in peri-urban and rural areas respectively. A total of 3% of the hand swabs in urban areas, 10% in peri-urban areas and 17% in rural areas were positive for *E. coli*. In 88% of the urban toilets, 83% of the peri-urban toilets and 73% of the rural toilets the toilet seat swabs were positive for total coliforms. *E. coli* was found in 41% of the urban, 58% of the peri-urban and 42% of the rural toilet seat swabs. The unsatisfactory cleaning of the toilets and the lack of use of disinfectants could be the reason for these results. Toilet seats can be classified as disease transfer points because they are regularly touched by the bare skin of more than one person. The spread of bacteria such as *E. coli* can not be controlled unless a disinfecting process is performed on these surfaces (CDC, 2008).

The results from the study indicated that the water collected from household storage containers was positive for total coliform bacteria in 93% of urban households, 98% of peri-

urban households and 89% of rural households. The water samples positive for *E. coli* were 33% in urban areas, 67% in peri-urban and 41% in rural areas. The microbial quality of water did not meet the standards set out in the WHO guidelines which Zimbabwe uses. In all water intended for drinking, treated water entering the distribution system or treated water in the distribution system, total coliform bacteria and *E. coli* must not be detectable in any 100 ml sample (WHO, 2008). These results indicated contamination of the water from a faecal source, indicating that the water might potentially be contaminated with pathogenic microorganisms. This is likely to expose the PLWHA to infection with diarrhoeal diseases and other opportunistic infections (Kamminga and Wegelin-Schuringa, 2003). The quality of drinking water is a well-recognised factor in the transmission route for infectious diarrhoea and other diseases (WHO, 2003).

Sample type	Urban	Peri-urban	Rural
	Number (%)	Number (%)	Number (%)
Hand swabs	24 (77%) n=31	25 (83%) n=30	21 (72%) n=29
Toilet seats	132 (88%) n=150	47 (83%) n=57	88 (73%) n=120
Water storage container	140 (93%) n=150	118 (98%) n=121	127 (89%) n=142

Table 6. Total coliform counts given as colony forming units per 100 ml (cfu/100 ml)

Sample type	Urban	Peri-urban	Rural
	Number (%)	Number (%)	Number (%)
Hand swabs	1 (3%) n=31	3 (10%) n=30	5 (17%) n=29
Toilet seats	61 (41%) n=150	33 (58%) n=57	50 (42%) n=120
Water storage container	50 (33%) n=150	81 (67%) n=121	58 (41%) n=142

Table 7. *E. coli* counts given as colony forming units per 100 ml (cfu/100 ml)

Contamination of water at household levels has been shown to be a risk factor in the transmission of the Hepatitis E virus (DREF, 2008). More than half of the hospital beds in the developing countries are estimated to be occupied by patients suffering from ailments associated with water of poor quality (UNDP, 2006). Improving the quality of water at household level would go a long way in addressing diarrhoeal diseases, particularly

amongst PLWHA. The storage conditions and the unhygienic maintenance of containers could have contributed to the poor results.

3.7 Prevalence of pathogenic *Escherichia coli* strains

The one *E. coli* positive sample found from the hand swab samples in urban areas and all three isolates from the peri-urban areas did not test positive for virulence genes. In rural areas one of the three *E. coli* positive hand swab samples tested positive for atypical EPEC. The presence of atypical EPEC (which is more frequently isolated from diarrhoea cases than typical EPEC) on the hands of PLWHA indicates the potential risk of contracting diarrhoea.

In toilet seat samples, atypical EPEC was the most prevalent pathogenic *E. coli* strain found in urban areas at 25%, followed by typical EPEC at 24%. EAEC was also fairly prevalent at 18%, with ETEC following at 10%. Almost 83% of *E. coli* strains identified in urban areas were pathogenic, compared to 40% in peri-urban areas and 69% in rural areas. The higher presence of pathogenic *E. coli* strains in urban areas could contribute to higher diarrhoea prevalence.

In household water storage container samples from the urban areas, the most prevalent pathogenic strain was typical EPEC (25.5%), followed by atypical EPEC (11%), EAEC (9.1%), ETEC (0.8%), EHEC (1.8%) and EIEC (1.8%). The remaining 49% of the samples had commensal *E. coli*. In peri-urban water storage samples ETEC was 4.9%, atypical EPEC was 3.7% and EHEC was 2.5%. EAEC was the least prevalent at 1.2%. The rest of the *E. coli* positive samples (87.7%) only had commensal *E. coli* present. In rural areas the most prevalent pathogenic *E. coli* strain detected in water samples was EAEC (14%), followed by atypical EPEC and EHEC (5% each). ETEC was 3.4% and the least prevalent was EIEC at 1.7%.

E. coli from toilet seats could be transferred to water during storage and handling resulting in these poor results, especially with inadequate hand-washing after using the toilet. The presence of pathogenic strains of *E. coli* in water at household level indicated the risk that the PLWHA were exposed to diarrhoeagenic bacteria, reinforcing the need for appropriate household level water supply and storage. Most of the households in all the areas had to store water in their dwellings. In urban areas 99.3% of the households had water stored inside the house, 96% in peri-urban areas and 99.3% in rural areas also stored water inside the house. Those that stored water outside the house were 3.3% in urban areas, 9% in peri-urban areas and 2% in rural areas. A few households stored water both inside the house as well as outside the house. The same containers used for water collection were also used for storage. In most households in the urban and peri-urban areas the same room that was used for water storage was also used for various other uses such as sleeping, cooking and in some cases, even bathing. In general water was not adequately protected from contamination.

Though in most of the households water containers were found clean, both inside and outside of the container, quite a number of household containers were found with either loose particles of dirt inside (27% in urban areas, 40% in peri-urban areas and 43% in rural areas) or with a biofilm (3% in urban areas, 7% in peri-urban areas and 9% in rural areas). The external conditions of containers were found to be unsatisfactory in more than half of the households in all the three areas. In about half of the households in all areas (51% in urban areas, 54% in urban areas and 48% in rural areas) both wide-mouthed containers and those with screw-type mouths were used. In 41% of the urban households, 22% of the peri-urban households and 21% of the rural households, only containers with screw tops were

used. Wide mouth bucket type containers were used in 8% of the urban households, 24% of the peri-urban households and 31% of the rural households.

The main water uses mentioned by the respondents in all the areas were cooking, drinking, bathing and laundry, dishwashing and cleaning. Some households in urban areas mentioned toilet flushing as a use whilst some peri-urban households mentioned drinking water for animals as a use. The largest volume of water was used for laundry in most households, however in most cases laundry was done only once or twice a week and in some rural households laundry was done at the water source. Bathing used the second largest amount of water in most of the households in all the areas.

The average total volume of water containers in each urban household was 95 litres. The average actual volume of water found in each household was 87 litres. The average volume used by each urban household was 127 litres per day. Considering that the average family size of an urban household was 5, the per capita per day volume of water used was 25.4 litres, which is satisfactory if the SPHERE project recommended volume of 15 litres per capita per day is used. Even if the average actual volume of water found in each household is used, the volume per capita per day of 17.4 litres is still above the SPHERE recommended standard.

In peri-urban areas the average total volume of water containers in each household was 79 litres and the average actual volume found was 61 litres. The average total volume of water used in each household was 121 litres. The per capita per day volume of water used was 25.7 litres. Based on the actual volume of water found in each household the per capita per day water available was 13 litres, which is below the SPHERE recommended volume.

Each rural household had an average total volume of containers of 70 litres. There was an average of 48 litres of water found in each household. According to the respondents, an

<i>E. coli</i> strain	Hand swab samples			Toilet seat samples			Water storage container samples		
	Urban	Peri-urban	Rural	Urban	Peri-urban	Rural	Urban	Peri-urban	Rural
Commensal <i>E. coli</i>	1 (100%)	3 (100%)	2 (40%)	13 (16.4%)	18 (60%)	16 (31.3%)	27 (49%)	71 (87.7%)	41 (70.7%)
Atypical Enteropathogenic <i>E. coli</i>	0 (0%)	0 (0%)	1 (20%)	19 (24.0%)	2 (6.6%)	7 (13.7%)	6 (11%)	3 (3.7%)	3 (5.2%)
Typical enteropathogenic <i>E. coli</i> (bfp and eae)	0 (0%)	0 (0%)	0 (0%)	20 (25.3%)	0 (0%)	0 (0%)	14 (25.5%)	0 (0%)	0 (0%)
Enterohaemorrhagic <i>E. coli</i>	0 (0%)	0 (0%)	0 (0%)	2 (2.5%)	0 (0%)	1 (1.9%)	1 (1.8%)	2 (2.5%)	3 (5.2%)
Enterotoxigenic <i>E. coli</i>	0 (0%)	0 (0%)	0 (0%)	8 (10.1%)	6 (20%)	3 (5.8%)	1 (1.8%)	4 (4.9%)	2 (3.4%)
Enteroaggregative <i>E. coli</i>	0 (0%)	0 (0%)	2 (40%)	14 (17.7%)	4 (13.3%)	22 (43.1%)	5 (9.1%)	1 (1.2%)	8 (13.8%)
Enteroinvasive <i>E. coli</i>	0 (0%)	0 (0%)	0 (0%)	3 (3.8%)	0 (0%)	2 (3.9%)	1 (1.8%)	0 (0%)	1 (1.7%)

Table 8. Pathogenic *E. coli* strains detected in hand swabs, toilet seat and water storage container samples

average of 157 litres of water was used in each house every day. Based on this, the per capita per day volume of water used is 30 litres, however based on the average volume of water found in each household, the figure drops to 9.2 litres, far below the SPHERE project recommendation. The water volumes found in the rural house households were the lowest compared to the other areas.

In urban areas in 29% of the households water for drinking was boiled before it was consumed, compared to 18% in peri-urban areas and 17% in rural areas. The households where drinking water was disinfected with chlorine before consumption were 24% in urban areas, 21.5% in peri-urban areas and 23% in rural areas. The disinfectant, in the form of *Aquatabs* (167mg sodium dichloroisocyanurate) was supplied either by the municipality or by some NGO supporting the PLWHA. Allowing the water to stand and settle was used by 8.3% of the peri-urban households, 4.9% of rural households and 0.7% of urban households. Proper handling and storage and household-level disinfection is therefore necessary to maintain the quality of water (USAID, 2006). It has been observed in a wide variety of settings that water quality improvements at the point-of-use are likely to have a positive impact on health at all levels of the water supply system (USAID, 2006). There are physical and chemical treatments available as interventions to improve the quality of water at the point of use (Sobsey, 2002).

4. Conclusions

The presence of total coliforms and *E. coli* in water, toilet seat and hand swab samples was determined using the Colilert® Quanti-Tray/2000 System. Multiplex PCR was used to identify pathogenic strains of *E. coli*. The technique was found to be appropriate in detection of virulence genes to identify various pathogenic *E. coli* strains from water, sanitation and hygiene samples. The presence of total coliforms and *E. coli* in household samples, especially in water samples and hand swabs indicated the potential risk of enteric diseases that the PLWHA are exposed to. Pathogenic *E. coli* strains found in household samples indicated the risks to which PLWHA are exposed to, more so in view of their compromised immune status.

There is therefore need for hygiene education at the household level on the importance of household water storage to prevent contamination. Appropriate household water treatment systems, such as filters or disinfectants are needed, especially in households where there are PLWHA, to ensure that the water is safe for human consumption. The presence of total coliforms in hand swabs was indicative of inadequate hand washing, especially non-use of soap and disinfectants. This increases the potential of faecal-oral transmission of enteric pathogens. Hygiene education on appropriate and effective hand washing needs to be reinforced in all the communities. Adequate cleaning and disinfection of toilets seats, which can be disease transfer points, is needed to reduce the potential of disease transmission.

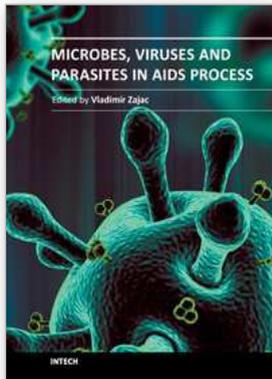
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Microbes, Viruses and Parasites in AIDS Process

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The main goal in compiling this book was to highlight the situation in Africa in terms of AIDS and opportunistic diseases. Several chapters reveal great poverty, an apocalyptic situation in many parts of Africa. Global migration of people resulted in their exposure to pathogens from all over the world. This fact has to be acknowledged and accepted as African reality. New, unconventional hypotheses, not determined by established dogmas, have been incorporated into the book, although they have not yet been sufficiently validated experimentally. It still applies that any dogma in any area of science, and medicine in particular, has and always will hinder progress. According to some biologists, in the future, AIDS is very likely to occur in a number of variations, as a direct result of the ongoing processes in the global human society. Thus, we urgently need a comprehensive solution for AIDS, in order to be ready to fight other, much more dangerous intruders.

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