# **Original Research Article**

# Analysis of Fecal Coliform Levels at Watering Points along the Upper Reaches of River Isiukhu in Kakamega County, Kenya

4 5

1

#### 6 Abstract

7 Diarrheal diseases often attributable to poor sanitary conditions and fecal contamination of drinking water remain a 8 leading cause of mortality for children younger than five years. Water contaminated with human faeces, for example 9 from municipal sewage, septic tanks and latrines, is of particular concern. Animal faeces also contain 10 microorganisms that can cause diarrhea. Kakamega County in Kenya has a diarrhea prevalence rate of 20.2%, which 11 is the highest in the country; a good proportion of these cases are believed to be water borne. This study was 12 designed to determine faecal coliform levels in water samples collected from watering points along the upper 13 reaches of River Isiukhu in Kakamega County, Kenya. Fifty-four samples were collected between August and 14 October 2015 from nine sampling points, comprising springs and watering points along the river. Water samples 15 were filtered on nitrocellulose filters by vacuum filtration; faecal coliform counts were conducted using membrane 16 filters cultured on mFC agar to establish contamination levels. The results indicated that counts ranged from 17 200cfu/100ml - 1450cfu/100ml in river sampling points and ranged from 0cfu/100ml - 44cfu/100ml in springs 18 sampling points. The faecal coliform counts for River Isiukhu and most springs surrounding it exceeded the WHO 19 recommended drinking water coliform(or E.coli) count value of 0cfu/100ml indicating that water from the upper 20 reaches of River Isiukhu and springs is not fit for drinking before treatment, especially during the wet seasons, based 21 on WHO drinking water standards.

22 Keywords: Faecal-coliform, Contamination, River-water, Spring-water, Season

23

#### 24 Introduction

25 Water has been classified as a natural resource and is important in sustaining life. Ashbolt et al., (2001) reported that 26 the accessibility and availability of clean drinking water not only plays a vital role in economic development and 27 social welfare, but is also an important component in health, food production and poverty alleviation. Despite its 28 significance, WHO (2006) revealed that safe potable water is not accessible by about 1.1 billion people in the world, 29 and the hourly toll from biological contamination of drinking water is 400 deaths of children below the age of five. 30 In most developing countries, Kenya being one of them, the demand for clean drinking water supply is growing 31 rapidly (Gelover et al., 2006). In addition, a small percentage of people in these countries access piped water. 32 Therefore, those who do not have access to safe drinking water, as well as those who have access but cannot afford 33 it; rely on other sources of water of questionable quality (Gadgil, 1998). It is often assumed that spring water 34 emerges from the ground clean and free of contaminants, especially in rural areas where industrial contamination is 35 not present (Wampler et al., 2010). Many rural Kenyans know that drinking untreated water from surface streams 36 and rivers is not safe but, they often assume that water emerging from the ground at a spring is clean and safe to 37 drink.

38

39 Although not absolute, several pathogenic microorganisms have been recommended as indices of faecal pollution

40 and act as indicators of microbiological quality of domestic water (WHO, 2003; Kabler and Clark, 1960; Abera et

41 al., 2011). According to WHO (2006), drinking water should contain zero fecal coliform and coliform organisms per

42 100 ml. However, human activities, particularly urbanization, and waste disposal and agricultural practices have

- 43 greatly increased inputs of microbial and other pollutants to terrestrial and aquatic habitats (Smol, 2009). Therefore,
- 44 the present study sought to establish the level of faecal coliform in sampling points in the upper reaches of River
- 45 Isiukhu.
- 46

# 47 Materials and Methods

#### 48 Study area

The study was carried out along River Isiukhu, Kakamega County, Kenya. The average annual rainfall of KakamegaCounty is 1800mm per annum and is bimodally distributed with peaks in April-May and August-September.

51 Temperatures range from a minimum of 10.3°C to a maximum of 30.8°C with an average of 20.5°C. Anthropogenic

- 52 activities are done along the river such as agriculture, livestock watering, laundry and bathing and sewage disposal.
- 53

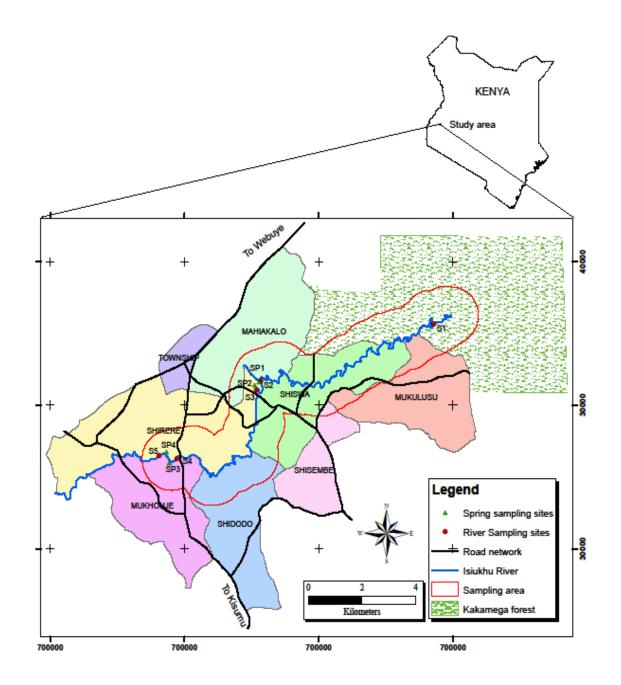
### 54 Sample collection

The sampling sites were River Isiukhu and springs along it. The sites were randomly chosen and they represented the upper reaches of the river as shown in Table 1. In the study, samples were collected in three successive months (August, September and October). Approximately 250ml of water samples were aseptically collected in duplicates from watering points using sterile containers and immediately stored in an ice box before being transported to the laboratory for analysis. Water samples were collected from a depth of about 10-20 cm below the water surface at each site to avoid potential contamination from surface water.

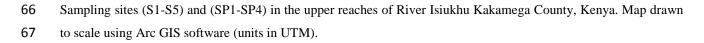
61

#### 62 Table 1: River and spring sampling sites

Site	Label
Forest	S1
Confluence of Lianila stream and Isiukhu River	S2
Savona resort area	S3
Amalemba	S4
Waste water entry point	S5
Spring 1	SP1
Spring 2	SP2
Spring 3	SP3
Spring 4	SP4



65 Figure 1



# 71 Research Design

72 The present study adopted randomized sampling design, where Fifty four duplicate samples were collected between

the month of August and October 2015 from nine sampling points along the river and springs that represented theupper reaches of River Isiukhu.

75 Determination of faecal coliform levels at watering points and springs along the upper reaches of River76 Isiukhu.

77

78 Faecal coliform levels in water samples were determined by the membrane filter procedure using mFC agar (Hi 79 Media, India). The samples collected from the river were diluted to 1:100. The biomass from both spring and the 80 diluted river-water samples (100ml) was concentrated via subsequent filtration on nitrocellulose filters, 47mm 81 diameter with pore size of 0.45µm by vacuum filtration. The vacuum pressure for filtration was between 50mm to 82 70mm Hg in order to avoid rupture of bacterial cells that has been observed at pressures above 80 mm Hg (Kepner 83 and Pratt 1994). Following the method recommended by the American Public Health Association in 2006 (Wehr et 84 al., 2004; Britton and Greeson 1987) filters were then aseptically placed with grid side up onto the surface of the 85 plates of mFC. All plates were incubated inverted in watertight plastic bags submerged in a 44.5°C water bath for 86 22-24 hours. Fecal coliform colonies that were observed in any shade of blue color were counted using a Quebec 87 colony counter and recorded as colony-forming units per 100ml. The formula used was:

88

$$CFU/100mL = \frac{Colonies \ counted}{mL \ filtered} \ge 100$$

89

90

91

# 92 Data analysis

Data obtained on coliform counts was subjected to one-way ANOVA followed by Turkey's post hoc test at 95%confidence level using Winks software version 7.

95 96

# 97 Results

98Table 2 shows the mean counts of faecal coliforms from five different sampling points in River Isiukhu for three99successive months. There was a significantly high (p<0.05) faecal coliform concentration between sampling point,</th>100S5 and all other sampling points in the river in the month of August. Sampling point, S2 also recorded significantly101higher (p<0.05) coliform counts when compared to S1 in the month of October. In the month of September, S2 and</th>102S5 had visibly higher coliform counts per 100ml, though they were not significantly different from the rest of the103samples. Generally, fecal coliform counts were lowest at S1 and highest at S5in all the three month of the study.

104

Sample code	Environment	Aug.	Sept.	Oct.
S1	Forest (site 1)	$200{\pm}0.00^{a}$	600±141.42 <sup>a</sup>	600±424.26 <sup>a</sup>
S2	Confluence of Lianila stream and	$300{\pm}141.42^{ab}$	$1050{\pm}1343.50^{a}$	$1700 \pm 424.26^{b}$
	River Isiukhu			
S3	Savona resort area	$550 \pm 494.97^{ab}$	$750\pm636.40^{a}$	$950{\pm}353.55^{ab}$
<b>S4</b>	Amalemba area	450±353.55 <sup>ab</sup>	$900 \pm 848.53^{a}$	$1050{\pm}636.40^{ab}$
S5	Waste water entry point	1450±212.13 <sup>c</sup>	1050±353.55 <sup>a</sup>	950±212.13 <sup>ab</sup>

105 Table 2: Means of Faecal coliform count given in colony forming units /100mls for five sampling points from

106 **River Isiukhu for three consecutive months** 

108

#### 107 \*Means followed by the same letter within the same column are not significantly different at P < 0.05

#### 109 Faecal coliform count for four sampling points from springs along River Isiukhu

110 Table 3 shows the mean counts for fecal coliform from four different springs for three consecutive months. 111 Generally, the month of October recorded higher coliform count compared to September and August. The month of 112 August recorded few counts across all the springs, in the three months studied. However, it is important to note that 113 there was no significant difference in the counts for all the months studied. In the month of August, spring SP1 114 recorded no coliform counts, however, increasing counts were recorded in the month of september and October.

115

#### 116 Table 3: Means of Faecal coliform count given in colony forming units /100mlfor four sampling points from

#### 117 spring for six sessions

118

vironment	Aug. Sept.	Oct.
fluence of Lianila s	and $0\pm 0.00^{a}$ $5\pm 7.07^{a}$	$6.5 \pm 4.95^{a}$
er Isiukhu		
ona resort area	$3\pm1.41^{a}$ $3\pm2.83^{a}$	44±46.67 <sup>a</sup>
alemba area	2.5±3.54 <sup>a</sup> 6.5±4.95	<sup>a</sup> 6.5±4.95 <sup>a</sup>
ste water entry point	5.5±4.95 <sup>a</sup> 6.5±2.12	<sup>a</sup> 36.5±43.13 <sup>a</sup>
ste water entry point	5.5±4.95 <sup>a</sup> 6.5±2.12	a

119 \*Means followed by the same letter within the same column are not significantly different at P < 0.05

120

#### 121 Discussion

122 Generally, coliforms are the most common group of indicator organisms used in water quality monitoring (Sibanda 123 et al., 2013). Furthermore Alotaibi (2009) elaborated that the presence of fecal coliform indicates the presence of 124 potential fecal contamination and the presence of possible pathogenic microorganisms and to determine the health 125 risk to the consumers. In the present study, significant variation of fecal coliform counts in River Isiukhu was 126 reported during the three months of the study. This difference is attributed to rainfall variation within the three

127 months. Lower rainfalls are normally experienced in the month of August than in the month of October. Similar 128 observations have been made elsewhere, for instance, Wolf (1999) reported that in the dry season, there were fewer 129 incidences of faecal pollution in the water supply system but high fecal contamination of drinking water increases 130 during wet season.

131

Results in the present study further indicated significant differences in means for fecal coliforms in samples taken from the different sampling points in River Isiukhu. This finding agrees with a previous study carried out on River Danube which showed lower bacterial pollution on upstream of the River and higher levels of fecal pollution in the middle part and downstream of the River (Kavka *et al.*, 2002). However, the finding in this study disagrees with that of Uwimpuhwe (2014) where total and fecal coliform from different sampling points in River Nyabarongo showed insignificant differences. This difference could be attributed to the fact that the current study of River Isiukhu included survey of sampling points with different anthropogenic activities taking place at every point.

139

140 According to the WHO guideline values for bacteriological parameters, the total and fecal coliform bacteria should 141 be 0 cfu/100ml in water intended for drinking. However, in this study the fecal coliform counts for River Isiukhu, 142 exceeded the WHO recommended drinking water guideline value. These results are supported by previous studies 143 conducted in rural areas (Abera et al., 2011; Chigor et al., 2011). The studies found that microbiological parameters 144 counts for river water in rural areas were above the permissible limits and were a potential hazard to public health 145 (Chigor, 2011). Even though, WHO recognizes that these targets would be difficult to achieve in some cases, 146 especially in rural communities with untreated water provisions, and recommends that in these settings, the 147 guidelines values would be seen as goals for the future, but not an immediate prerequisite (WHO/UNICEF, 2008)

148

149 Although findings from the study reported insignificant difference in fecal coliform counts from different springs 150 during the three months studied, the water was unsafe to drink based on the World Health Organization (WHO) 151 drinking water standard. Both protected and unprotected springs had bacterial counts in excess of the WHO 152 standard, suggesting that water treatment from all sources is necessary to ensure clean and safe drinking water. The 153 spring water contamination could be due to what Narain Rai and Sharma (1995) termed as lack of sanitation or 154 improper waste disposal. The researchers further explained that 40% or more of the disease out breaks were 155 attributed to consumption of polluted ground water. Furthermore, presence of coliforms in drinking water sources 156 indicates need for treatment and proper sanitation which is necessary for drinking (Christine et al., 2006). Further the destruction of microbial ecosystems through deforestation, high spring water temperatures, averaging 24.4 °C, 157 158 may be contributing to the observed bacterial abundance.

159

Findings from the study indicate that springs have more fecal coliform contaminants during wet season. This finding
is not different from that of Ofoma *et al.*, 2005) where spring water was highly contaminated with fecal coliform
during wet seasons compared to dry seasons. They suggested that the contamination was due to pollution; most

163 likely from sewage as well as solid waste discharge during run-offs. This is non-point sources typically wet-weather

where they diffuse in nature, in that they do not enter water bodies from any single point (e.g. urban litter, contaminated refuse, domestic pet/wildlife excrement and failing sewer lines). This suggests that emphasis on points of use (POU) treatment methods, decontamination of protected and unprotected springs, and behavioral interventions to improve sanitation practices are needed.

168

# 169 Conclusion

170 Results obtained from the study indicated significant contamination of River and spring water at different sites. The 171 levels of faecal coliforms were higher than the accepted levels. In addition, higher faecal coliform levels were 172 recorded during wet than dry seasons, concluding that water from the upper reaches of River Isiukhu and spring is 173 not fit for drinking before treatment especially during wet seasons based on the WHO drinking water standard and 174 the water quality. It is therefore recommended that water from both the stream and springs in the upper reaches of 175 River Isiukhu should be treated before use.

- 176
- 177

- 179
- Abera S, Zeyinudin A, Kebede B, Deribew A, Ali S, Zemene E (2011). Bacteriological analysis of drinking water
   sources. Afr. J. Microbiol. Res. 5(18): 2638-2641.
- 182 Alotaibi EL (2009). Bacteriological assessment of urban water sources in KhamisMushait Governorate,
  183 southwestern Saudi Arabia. Int J Health Geogr. 8(1): 1.
- Ashbolt NJ, Grabow WO, Snozzi M (2001). Indicators of microbial water quality. IWA Publishing. 289-316.
- Britton LJ, Greeson PE (1987). Methods for collection and analysis of aquatic biological and microbiological
  samples. U.S. Geological Survey Techniques of Water Resources Investigations. 5 (A4): 37-40.
- 187 Chigor VN, Umoh VJ, Okuofu CA, Ameh JB, Igbinosa EO, Okoh AI (2012). Water quality assessment. surface
  188 water sources used for drinking and irrigation in Zaria, Nigeria are a public health hazard. Environ. Monit.
  189 Assess. 184(5). 3389-3400.
- Christine L, Moe, Richard D, Rheingans (2006). Global challenges in water, sanitation and health. Journal of Waterand Health. 04.IWA Publishing 2006.
- 192 Gadgil A (1998). Drinking water in developing countries. Annual Review of Energy and the Environment
   193 Resources. 23(1): 253-286.
- 194 Gelover S, Gomez LA, Reyes K, Leal MT (2006). A practical demonstration of water disinfection using TiO 2 films
  195 and sunlight. Water Res. 40(17): 3274-3280.
- Kabler PW, Clark HF (1960). Coliform group and fecal coliform organisms as indicators of pollution in drinking
  water. J Am Water Works Assoc. 52(12): 1577-1579.

#### UNDER PEER REVIEW

- Kavka GG, Kasimir GD, Farnleitner AH (2006). Microbiological water quality of the River Danube (km 2581-km
  15). Longitudinal variation of pollution as determined by standard parameters. Eigenverlag ICPDR: 415 –
  421.
- Kepner RL, Pratt JR (1994). Use of fluorochromes for direct enumeration of total bacteria in environmental samples.
   past and present. Microbiol Rev. 58(4): 603-615.
- 203 Narain Rai, J.P. and Sharma, H.C.(1995) Bacterial Contamination of Ground Water in Rural Areas of North
   204 Uttarpradesh. Indian Journal Environmental Health. 37(1):37-41.
- Ofoma AE, Omologbe DA, Aigberua P (2005). Physico-chemical quality of groundwater in parts of Port Harcourt
   City, eastern Niger Delta, Nigeria. Water Resour. Res. 16: 18-24.
- 207 Sibanda T, Chigor VN, Okoh AI. Environ Monit Assess (2013) 185: 6579. <u>https://doi.org/10.1007/s10661-012-</u>
   208 <u>3048-4</u>
- 209 Smol JP (2009). Pollution of lakes and rivers: a paleoenvironmental perspective. John Wiley & Sons.
- Uwimpuhwe M (2014). Microbiological assessment of water quality and prevalence of waterborne diseases in rural
   areas of Masaka, Rwanda (Doctoral dissertation).
- Wampler PJ, Sisson AJ(2011). Spring flow, bacterial contamination, and water resources in rural Haiti.
   Environmental Earth Sciences. 62(8): 1619–1628.http://doi.org/10.1007/s12665-010-0645-9
- Wehr HM, Frank JF, American Public Health Association (Eds.) (2004). Standard methods for the examination of
   dairy products (pp. 327-404). Washington, DC: American Public Health Association.
- WHO (2006). Microbial aspects. In Guidelines for drinking water quality. World Health Organisation 3<sup>rd</sup> ed. Vol. 1,
   Geneva:
- WHO/UNICEF Joint Water Supply, Sanitation Monitoring Programme, World Health Organization, UNICEF
   (2008). Progress on drinking water and sanitation: Special focus on sanitation. World Health Organization.
- 220 Wolf AT (1999). The transboundary freshwater dispute database project. WATER INT. 24(2): 160-163.
- World Health Organization (2003). Water, sanitation and health. Geneva, World Health Organization. World Health
   Organization. (2006). Guidelines for drinking-water quality: First addendum to volume 1, Recommendations
   (Vol. 1). World Health Organization.
- 224

225