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Temporal Variation In Densities Of Microbiological Indicators Of Pollution In Water Sources Within Naivasha Lake Basin, Kenya

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Abstract: This study investigated the temporal variation in community water source types within Lake Naivasha basin; borehole Direct (Dir), boreholePoints of Access (POA), and surface sources between April and July 2011.This was done through determination of the concentrations of total coliforms, *E. coli*, intestinalenterococci, *C. perfringens* and HPC in Lake Naivasha, Rivers Malewa and Gilgil and borehole water samples using Membrane Filtration Techniques(MFT) and Pour Plate Procedures. Physico-chemical parameters were also determined *insitu* for all water sources. Most of the physicochemical parameters did not show variation with respect to sampling time (months) except temperature and conductivity from borehole direct and borehole points of access ($P<0.05$). Surface sources recorded higher values in all the four months for all the parameters than borehole direct and POA sources. *E. coli* and *C.* Perfringens showed significant temporal variation only in the surface sources but not on the two borehole sources ($p<0.05$) while total coliforms and intestinal enterococci did not show any significant temporal variation in all the water source types ($p>0.05$). In conclusion, the study indicated presence of temporal variation in some microbiological parameters and recommended that other environmental parameters such as rainfall patterns be included to better understand and interpret the results.

Index terms: Faecal pollution, microbiological, temporal variation, water quality

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1.0 INTRODUCTION

Naivasha is one of the fastest growing towns in Kenya. The growth is fuelled by the increasing horticulture farming businesses, especially floriculture around the Lake(JICA, 2003 [1]). In addition, the area surrounding the lake offers a mild climate and natural beauty that has attracted tourists. Lake Naivasha also supports a productive fishery that provides jobs and income as well as being an important source of protein for local communities. Tourism activities in the region, rural to urban migration as a result of falling farm incomes from traditional cash crops, and commercial enterprises and good prospects for job opportunities have led to tremendous rise in population density in Naivasha district from 43,867 in 1969 to 376,246 in 2009 (Government of Kenya Census Report, 2009[2]). Provision of water in this area like in any other part of the country for socio-economic and ecological sustenance is indispensable. Its availability is primarily influenced by its quantitative distribution in time and space, and by its quality.

As is common with other areas in developing countries, direct surface water is still commonly used as a source for domestic water (WHO, 2002 [3]). In the case of Naivasha area, Malewa River provides the water for approximately 250,000 people within the townships surrounding the lake (M'cleen, 2001[4]; LakeNet, 2003 [5]). Other sources of water include bore holes, rain water harvesting, shallow wells and lake water. Despite these being the main sources of water for the communities in this area, they are under threat of pollution from anthropogenic activities in the heavily populated area and from the Naivasha basin catchment (Harper and Mavuti, 2004[6]). Faecal pollution from human and other animals in this area is recognized as the major water pollutant and it has a bearing on public health (M'cleen, 2001[4]).Lack of adequate access to sanitation, safe and clean drinking water imposes significant economic losses on the population. The World Health Organization (WHO) has estimated that 80% of all sicknesses and diseases in the world are caused by inadequate sanitation, faecal pollution and unavailability of water. About 1.7 million annual deaths are attributed to unsafe water supplies with most of these being due to diarrhoeal diseases which also affect 90% of children from developing countries to which majority of these cases are due to bacterial pathogens contamination (WHO, 2002 [3]). There is likelihood of contamination of both surface and ground water sources within Naivasha due to inadequate sanitation as communities here depend on bushes, pit latrines and septic tanks for sewage disposal (Mireri, 2005 [7]). This study investigated the temporal changes in the levels of bacterial indicators of faecal pollution in domestic water sources; boreholes, rivers Malewa and Gilgil as well as Lake Naivasha using standard procedures (APHA, 2005 [8]). Selected physicochemical parameters; temperature, dissolved oxygen, percentage saturation of dissolved oxygen, pH and electrical conductivities were also measured in situ using appropriate measurement probes.

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2.0 MATERIALS AND METHODS

2.1 Study area

The research work was carried out within Lake Naivashabasin in Rift valley Province of Kenya (Figure 1). The Lake is one of the major water bodies in Kenya. It is a fresh water lake though it lacks a well established outlet and its watershed covers parts of both, the Rift Valley and the Central Provinces. The lake basin extends 6^0 North from the Equator and lies between $36^0 07'$ and $36^0 47'$ East of Greenwich Meridian. Lake Naivasha is located in the rain shadow of the Aberdare Ranges with a mean annual rainfall of about 1350millimetres and experiences long rain in Months of March to May and short rain in the months of September to October. The mean temperature around Lake Naivasha is approximately 25^0C with a maximum temperature of 30^0C, the months of December – March being the hottest period while July is the coldest month with a mean temperature of 23^0C. The Lake Naivasha watershed is drained by only two perennial rivers, namely Malewa River and Gilgil River with catchment areas of 1700km^2 and 400km^2 respectively. These two rivers get into the lake at its Nothern side. In addition, the lake is also drained by other seasonal rivers and streams with a major one beingKeratiriver to the North. The Lake, Rivers, shallow wells and ground water sources are a key to the provision of water to the Naivasha and Nakurumunicipalities as well as other adjoining human activities (Mireri, 2005[7]).

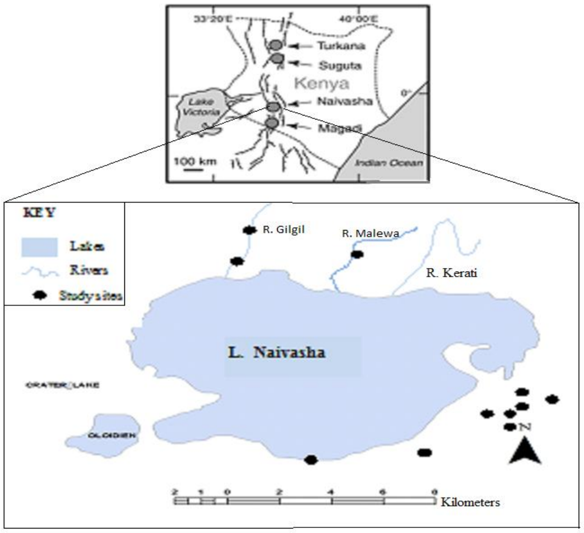

***Figure 1**: Map of Lake Naivasha showing study sites (modified from Martin et al., 2001*[9]*).*

2.2 Sampling

Water samples were obtained in triplicate from three different sources; Borehole direct sources and Borehole Points of Access sourcesbetwenn the months of April to July 2011 within Lake Naivasha Basin area. Samples were also obtained from surface sources which included lake Naivasha (at Kamerebeach), river Malewa (at the Naivasha-Nakuru highway bridge), river Gilgil (at the bridgenear Morendat Conference Centre and on the Naivasha-Nakuru highway bridge). Sampling was done on a weekly basis for duration of four months (April-July 2011). Sterilized water sample bottles were used to sample 30centimetres below the surface from the rivers and lake. Boreholes were sampled directly and at their points of use (POU). Water from the taps was sampled by first sterilizing the nozzle with 70% alcohol and then the bottles were aseptically filled-up. Physico-chemical parameters; temperature, dissolved oxygen, Electrical conductivity and pH of the water sources were measured in situ at the time of sampling using electrical probes. All samples were placed in a cool box with ice and transported to Egerton University, Department of Biological sciences laboratory for analysis.

2.3 Samples analysis

Analysis followed guidelines outlined in APHA 2005 [8]; Lawand*et al.,* 1997 [10] and Scott *et al.,* 2002 [11].This wasdone within 6-24 hours after sampling to avoid changes of the bacteria count due to growth or die off.Aseptic technique was observed in all the analysis procedures.

Heterotrophic plate count; 1ml of each sample or its dilution was placed onto 80mm diameter plates with plate count agar and incubated at 37^0C for 48 hours. Colonies forming units (CFUs) counted were expressed as; cells per 1ml (APHA, 2005 [8]).

Membrane filtration technique; Aseptic filtration was done separately for each dilution by passing the sample through a membrane filter (47mm diameter, 0.45µm pore size) on a filtration unit. The filter was taken off using a pair of forceps and placed on the surface of the corresponding culture media. For total coliforms and *E. coli* counts, filters were placed onto chromocult agar (Merck) plates and incubated at 37^0C for 18-24 hours.Typical colonies appearing pink and dark blue were counted as total coliforms and *E. Coli*respectively and numbers expressed asCFUs/100ml (APHA 2005[8]). For intestinal enterococci counts, filters were placed onto enterococci agar (Merck) plates and incubated at 44^0C for24-48hours. Typical colonies appearing pink were counted as intestinal enterococci and numbers expressed as CFUs/100ml (APHA, 2005 [8]). For C. Perfringens counts, filters were placed onto Tryptose sulphite Cyclocerine (TSC) agar (Merck) plates. The filters were then placed in an anaerobic jar containing anaerocult strip and incubated at 44^0C for 18-24 hours.Black fluorescent counts of *C. perfringen* were made under 360nm UV light. Numbers were expressed as CFU/100ml as stipulated in (APHA, 2005 [8]).

2.4 Statistical analysis

Data analysis was done using Sigmaplot statistical package version 12. Non parametric test involving ANOVA on Ranks was used in testing the variability in the densities of microbiological and selected physicochemicalparameters' values between Months of sampling was based at 95% level of significance.

3.0 RESULTS

3.1 Physicochemical parameters

The median values of physicochemical parameters were as in Table 1. For the three water source categories, range of temperature values was between 20.8 to 23.6 °C. Dissolved oxygen values had a range of between 3.5 to 6.9 mg/l with surface sources recording higher values. pH values were moderately equal for the all sources with a range of between 8.0 to 8.1. Conductivity values ranged between 202.5µs/cm to 1382.5µs/cm with surface sources recording lower values. Most of the physicochemical parameters did not show variation with respect to sampling time (months) except temperature and conductivity from borehole direct and borehole points of access (Table 1).

Table 1: Monthly trends in the median values of physicochemical parameters (median values with different letters in the same column are significantly difference at P<0.05).

Water Source categories	Time (Months)	*N*	Temperature (°C)	DO (mg/l)	% DO	pH	Conductivity (µs/cm)
Borehole Direct Sources	APRIL	16	23.6 A	3.5 A	52.5 A	8.2 A	1232.0 A
	MAY	16	23.1 A	3.5 A	49.2 A	8.1 A	1174.5 B
	JUNE	16	22.5 B	3.9 A	55.8 A	8.1 A	1343.0 C
	JULY	16	22.7 B	3.6 A	56.1 A	8.0 A	1224.5 A
	P-value		**0.027**	**0.963**	**0.395**	**0.612**	**<0.001**
Borehole POA Sources	APRIL	20	23.6 A	4.9 A	71.6 A	8.3 A	1261.0 A
	MAY	20	22.8 AB	5.2 A	78.0 A	8.2 A	1181.0 B
	JUNE	20	22.4 B	5.2 A	74.2 A	8.5 B	1382.5 C
	JULY	20	22.5 B	5.0 A	73.2 A	8.2 A	1224.0 A
	p-value		**0.001**	**0.480**	**0.245**	**0.011**	**0.022**
Surface Sources	APRIL	16	21.8 A	6.6 A	94.7 A	8.1 A	254.0 A
	MAY	16	23.0 A	6.4 A	93.2 A	8.1 A	224.0 A
	JUNE	16	21.9 A	6.7 A	98.1 A	8.2 A	202.5 A
	JULY	16	20.8 A	6.9 A	94.8 A	8.2 A	229.0 A
	P-value		**0.470**	**0.401**	**0.887**	**0.896**	**0.257**

3.2 Microbiological parameters

The median values for microbiological indicators of faecal pollution were as in Figure 2. Surface sources recorded higher values in all the four months for all the parameters than borehole direct and POA sources. *E. coli* and *C. perfringens* from the surface sources indicated a steady increase in median values form the first month through the second and the third to the fourth one (April to May, June to July). *E. Coli*and *C. perfringens*showed significant variationonly on the surface sources but not on the two borehole sources based on sampling time (months) (Table 2). Total coliforms and intestinal enterococci did not show any significant temporal variation in all the water source categories (Table 2).

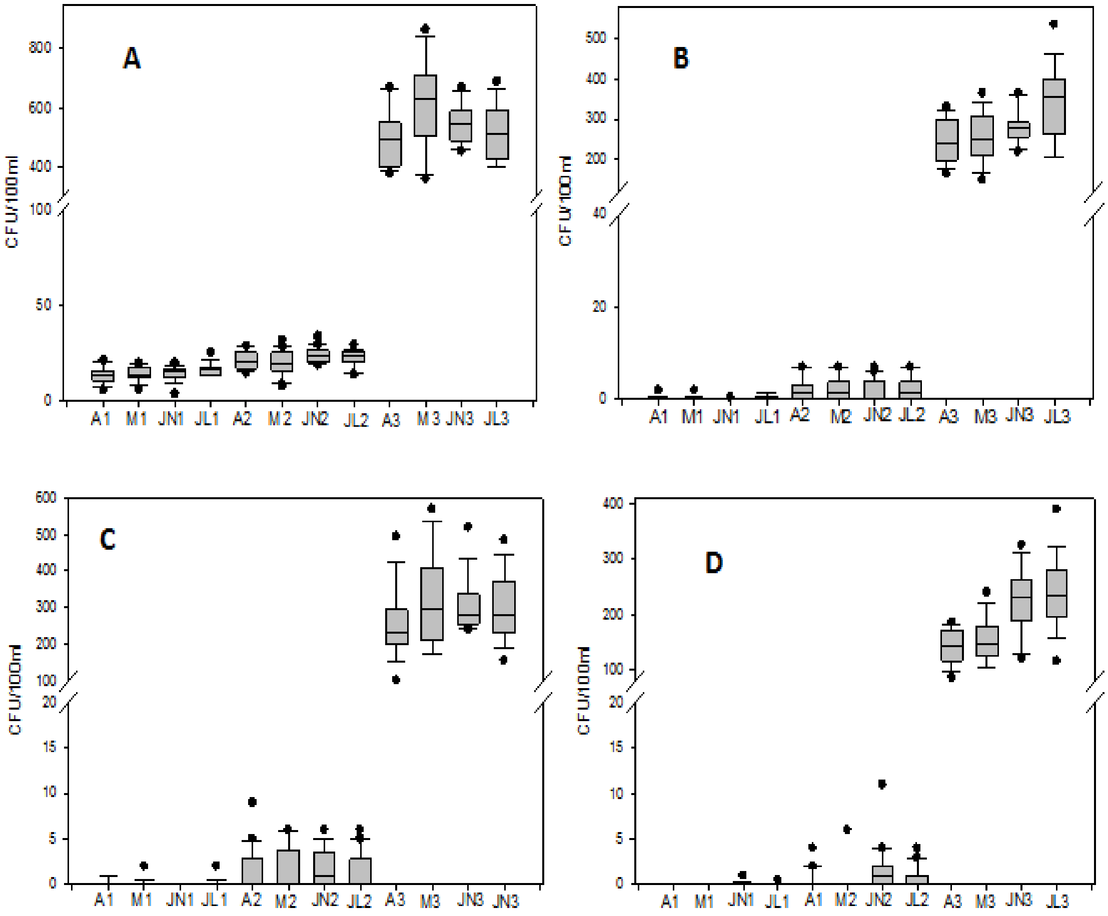

Figure 2: *Densities of faecal contamination indicators (A, B, C, and Drespectively for total coliforms, E. coli, intestinal enterococci and C. perfringens) in different water source types (1, 2 and 3 respectively for Direct borehole, boreholes POA and surface sources) at different months (A,M, JN and JL respectively for April, May June and July). The boxes span from 25th and 75th percentiles, with the median in between. The bars with whiskers span from estimated 10th and 90th percentile values and markers indicate out-liers.*

HPC median values were also highest in surface sources than in the two borehole sources as in Fig. 3. Only surface sources indicated steady increase in HPC median values from the month of April through to the month of July. There was significant temporal variation in HPC median values in all the three water source types (Table 2).

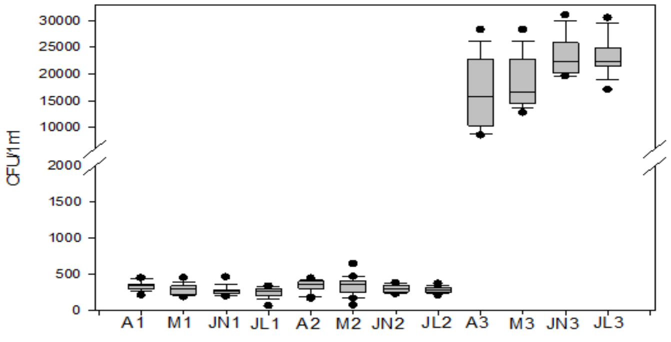

Figure 3: *Densities of HPCin different water source types (1, 2 and 3 respectively for Direct borehole, boreholes POA and surface sources) at different months (A,M, JN and JL respectively for April, May June and July). The boxes span from 25th and 75th percentiles, with the median in between. The bars with whiskers span from estimated 10th and 90th percentile values and markers indicate out-liers.*

Table2: Monthly trends in the median values of microbiological parameters (value with different letters in the same column are significantly difference at P<0.05).

Sources	**Time (Months)**	***N***	***E. coli***	**Total Colliforms**	**Intestinal Enterococci**	***C. perfringens***	**Heterotrophic Plate Counts**
Borehole Direct Sources	APRIL	16	0.0 A	14.0 A	0.0 A	0.0 A	237.5 A
	MAY	16	0.0 A	14.0 A	0.0 A	0.0 A	300.0 B
	JUNE	16	0.0 A	16.0 A	0.0 A	0.0 A	262.5 C
	JULY	16	0.0 A	16.5 A	0.0 A	0.0 A	272.5 C
	P-value		**0.32**	**0.122**	**0.32**	**0.566**	**< 0.001**
Borehole POA Sources	APRIL	20	1.5 A	20.5 A	0.0 A	0.0 A	365.0 A
	MAY	20	1.5 A	19.8 A	0.0 A	0.0 A	365.0 A
	JUNE	20	0.0 A	24.0 A	1.0 A	1.0 A	305.0 AB
	JULY	20	0.0 A	23.5 A	0.0 A	0.0 A	285. 0 B
	p-value		**0.906**	**0.084**	**0.860**	**0.06**	**0.005**
Surface Sources	APRIL	16	237.5 A	492.5 A	230.0 A	141.3 A	15800.0 A
	MAY	16	250.0 A	627.5 A	295.0 A	145.0 A	16637.0 A
	JUNE	16	277.5 AB	545.0 A	277.0 A	230.0 B	22250.0 B
	JULY	16	357.5 B	515.0 A	280.0 A	232.5 B	22300.0 B
	P-value		**0.008**	**0.057**	**0.182**	**< 0.001**	**0.001**

3.3 Relationship between feacal contamination indicators and physicochemical parameters

Relationships between parameters measured in different water sources are shown in Tables 3-5. From these results, minor significant relationships occurred between parameters in all borehole waters (direct and point ofaccess). Physico-chemical parameters did not show any great influence on microbial parameters. However, it is notable that HPC from direct borehole waters had positive and significant correlation with temperature as in Table 3. There were significant correlations between *E. coli* (EC) and all the microbiological faecal contamination indicators in surface water as in table 5.

Table 3: Correlation between microbiological and physico-chemical parameters in borehole direct sources

	EC	**TC**	**IE**	**CP**	**HPC**	**TEMP**	**DO**	**Ph**
EC	_	_	_	_	_	_	_	_
TC	0.300	_	_	_	_	_	_	_
IE	0.223	0.090	_	_	_	_	_	_
CP	-0.64	0.080	-0.044	_	_	_	_	_
HPC	0.059	-0.139	-0.064	0.152	_	_	_	_
TEMP	0.047	-0.006	0.176	-0.032	0.258**	_	_	_
DO	0.042	-0.014	0.002	0.080	0.062	0.138	_	_
Ph	-0.014	0.163	0.008	-0.173	-0.248**	-0.365**	-0.144	_
E. COND	-0.120	0.076	0.150	-0.184	-0.320**	-0.101	0.083	0.478

**Correlation is significant at P<0.05, (2 tailed, N=64)

Table 4: Correlation between microbiological and physico-chemical parameters in borehole point of access sources

	EC	TC	IE	CP	HPC	TEMP	DO	Ph
EC	_	_	_	_	_	_	_	_
TC	0.041	_	_	_	_	_	_	_
IE	0.148	0.194	_	_	_	_	_	_
CP	0.099	0.311**	0.215	_	_	_	_	_
HPC	0.127	0.234	-0.106	-0.012	_	_	_	_
TEMP	-0.013	0.000	0.195	-0.021	0.134	_	_	_
DO	0.084	0.364**	-0.003	-0.079	-0.169	-0.74	_	_
Ph	0.043	0.065	0.247**	0.111	-0.178	-0.029	0.206	_
E. COND	0.059	0.129	0.099	0.055	0.198	0.030	-0.086	-0.004

**Correlation is significant at P<0.05, (2 tailed, N=64)

Table 5: Correlation between microbiologicall and physico-chemical parameters in surface sources

	EC	TC	IE	CP	HPC	TEMP	DO	Ph
EC	_	_	_	_	_	_	_	_
TC	0.278**	_	_	_	_	_	_	_
IE	0.384**	0.112	_	_	_	_	_	_
CP	0.420**	0.085	0.282**	_	_	_	_	_
HPC	0.560**	0.056	0.447**	0.538**	_	_	_	_
TEMP	-0.085	-0.147	0.132	-0.242	-0.035	_	_	_
DO	-0.088	-0.052	-0.280	0.168	-0.010	-0.142	_	_
Ph	-0.234	-0.059	-0.122	0.037	-0.176	0.023	0.143	_
E. COND	0.078	0.005	0.272	0.085	0.100	0.241	-0.312**	0.049

***Correlation is significant at P<0.05, (2 tailed, N=64)*

4.0 DISCUSSION

From the results obtained in this study all the physico-chemical parameters from all the water sources were within the recommended standards by WHO, US-EPA and NEMA for drinking water. These results were also similar to those obtained in water sources from Aboekuta, Nigeria (Shittu*et al.,* 2008[12]). Higher levels of electrical conductivity were recorded from boreholewater sources than surface water sources as expected. This is attributed to high levels of dissolved minerals found in ground water as well as high fluoride contents in particular which have been recorded in borehole waters within the region (Matofari, 2006[13]). High values of dissolved oxygen in rivers and borehole waters at the points of access (POA) was due to aeration caused by the flow of water in the rivers and high pressures as the water flows from over-headreservoirs to the lower distribution taps connected with hose pipes where sampling was conducted. Lack of significant temporal variation in DO, % oxygen saturation and PH could be attributed and significant variation in temperature and conductivity of borehole direct and borehole points of access sources may be attributed to the influence of rainfall events as had been noted in studies by Kistemann*et al.,* (2002)[14] on the microbial load of drinking water reservoir tributaries during extreme rainfall and runoff. Within the four months of study duration, temporal variation in the density of faecal contamination indicators was not evidenced in both the borehole direct and borehole-POA sources for all the microbial parameters except HPC. This was an indication that there was no weather variable that had significant influence on the density of microbiological indicators of faecal contamination for the entire study period. This would probably indicate thatsanitary completion of bore holes in the region reduced the risks of contamination to a great extent with pathogens by providing a barrier to the influences of recharge pathways close to the source (Howard *et al*., 2006[15]). However sanitary conditions in some bore holes were appalling as evidenced by presence of pit latrines, strewn refuse and storm water close by which were possibly responsible to faecal pollution in some cases. The degree of contamination at the BH-POA did not change significantly with respect to sampling times. The reason for this could have been that the population of the local community which used/accessed the borehole during the entire study period and water accessibility factors remained the same. Within the Surface sources, all the parameters had showed significant variation with respect to sampling time except total coliforms and intestinal enterococci. This is because unlike the borehole sources, surface sources based on their microbiological quality are more susceptible to influence of weather variables such as rainfall. Rainy spells within the duration of this study were noted to have significant influence on the microbiological quality of surface water and such cases have been noted through studies in United States (Hatfield and Prueger, 2004[16]). A study by John and Jerry (1991)[17] and Obiri-Danso*et al*., (2009)[18] had also pointed out the influence of rainfall on microbiological densities. The positive and significant correlation between HPC and temperature in borehole direct sources means that temperature may be one of the factors influencing microbial growth in borehole water as had been justified by…..Randal *et al*., (2006)[19] who had found a strong correlation between *E. coli* load rate (colony-forming units [CFU]/d), antecedent precipitation, and turbidity.. The fact that there was no significant correlation between temperatures with faecal indicator micro-organisms means that faecal contamination in direct borehole water was of minor consequence in causing public health concerns. The significant correlations between EC and all microbial faecal indicators of pollution apart from TC in surface water sources could probably be an indicator that contamination with organic matter is of faecal origin. Lack of significant correlations between EC and TC in surface water might indicate that TC may have proliferated in the environment and there may be no direct relationship with EC which is of recent origin. This low correlation between microbiological indicator organisms had also been a major factor of discussion in a study within the wooded watersheds of the Southern Piedmont (Dwight and Dinku, 1999[20]). The ratios of *E. coli* to intestinal enterococci of different water sources were closer to 0.7 than 4.0. This showed that the major source of pollution into the water sources within Lake Naivasha basin were more of non-human warm blooded animals' origin. Indeed, domestic and wild animals were a common occurrence at the precincts of water sources. Vendors were also using donkeys for transportation of water to the consumers. In addition, domestic and wild animals were also noted to be visiting the lake and rivers for drinking purposes. This result was the same as what was found on a river in North Area of Wigry National Park where it was noted that source of faecal contamination was majorly coming from flows arising from other arable-forestry-pasture-meadow catchments as expressed by a larger number of streptococci than *E. coli*, (Niewolak, 1999[21]).

5.0 CONCLUSION AND RECOMMENDATIONS

This study has indicated that there is temporal variation in microbiological parameters withrespect to time of sampling in surface sources Borehole water in Naivasha can be considered to have minimal risk of waterborne disease transmission as it didn't seem to be threatened by faecal pollution during the study period. The significant correlation between HPC and EC may also be used to derive a conclusion that as the water sources get faecally polluted, high levels of pollution by organic materials may also be rampant. However, the study duration could have been too short to give an elaborate understanding of this. It is therefore recommended that a study be taken for a longer time and other environmental parameters such as rainfall patterns to be fully in-cooperated to better understanding and interpretation of similar results in future studies.

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