1		teriological quality of household drinking water and
2		era risk in the Greater Accra Region, Ghana
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27 Abstract

Background: The bacteriological quality of drinking water plays a critical role in preventing waterborne diseases. In Ghana, there is water scarcity and many communities depend on contaminated water sources for their domestic use. This study aimed to assess the microbiological quality of household drinking water in both cholera endemic and nonendemic areas in Greater Accra Region.

Methods: A community-based cross-sectional survey was conducted among 432 33 34 randomly selected households. A total of 432 water samples were collected in both the wet 35 and dry seasons from the households and an additional 48 from other water sources within the area studied. The membrane filtration technique was used for the quantification of total 36 coliform counts, faecal coliform counts and vibrio counts. The bacteria were further 37 and characterized. The Kruskal Wallis rank test was used to determine any identified 38 significant variations in the means of the log-transformed bacteria counts among specific 39 factor variables. 40

Results: Household drinking water samples were contaminated with coliform counts 41 exceeding the recommended zero colony-forming units per 100 ml standard in most 42 communities across the cholera endemic and non-endemic areas. Vibrio counts were 43 44 detected in all household water stored in vessels. Further characterization identified predominantly Klebsiella penumoniae and Escherichia coli. The coliform contamination 45 levels were significantly higher in water stored in vessels compared to water directly 46 47 obtained from the source. The contamination levels were generally higher during the wet season than the dry season. 48

49 **Conclusion:** The household stored drinking and direct source water were highly

- 50 contaminated with coliform bacteria and a risk for transmission of pathogenic waterborne
- 51 diseases. Therefore, the need to implement an effective water treatment strategy to improve
- 52 on the quality of drinking water.
- 53 **Keywords:** Household water, contamination, coliform, cholera-risk, water source

55 Introduction

Water is a necessity for sustaining life and maintaining health. Access to safe and clean drinking water is a fundamental human right, yet billions of people globally, particularly in developing countries, are still deprived of this necessity [1, 2]. Among those who have access to water, millions drink from water sources which are contaminated with excreta [1], which increases the risk of contracting waterborne illnesses, such as cholera, which is still a serious public health concern in many parts of the world [3].

62 Cholera, is caused by the *Vibrio cholerae*, an acute diarrheal disease that can lead to severe 63 dehydration and even death if left untreated. The disease is primarily transmitted through 64 the consumption of contaminated water or food, with contaminated water being the most 65 significant source of infection. Cholera outbreaks are known to occur in regions where 66 sanitation and hygiene practices are suboptimal and access to safe drinking water is limited 67 [3, 4].

The Greater Accra Region, located in southern-most part in Ghana, is a densely populated area with rapid urbanization and inadequate water and sanitation infrastructure. The region has experienced recurrent cholera outbreaks over the years, resulting in significant number of deaths [5, 6]. The lack of access to safe drinking water, coupled with poor sanitation practices, makes the population vulnerable to cholera infection.

Understanding the bacteriological quality of household drinking water is crucial in assessing the risk of cholera transmission within communities. Bacteriological analysis provides insights into the presence and levels of indicator microorganisms, such as faecal coliforms and *Escherichia coli (E. coli)*, which serve as markers for faecal contamination [7]. High levels of these indicator bacteria indicate the potential presence of pathogens, including *V*. *cholerae* [7].

Several factors can contribute to the deterioration of drinking water quality, such as inadequate water treatment, contaminated sources and improper storage and handling practices at the household level [8-10]. Identifying these factors and assessing their impact on water quality is essential for designing effective interventions to reduce the risk of cholera transmission and improve public health outcomes.

This study aimed to investigate the bacteriological quality of household drinking water and its association with cholera-risk in the Greater Accra Region of Ghana. The presence and levels of faecal indicator bacteria were assessed in the household drinking water samples collected from different communities in the cholera endemic and non-endemic communities in the Greater Accra Region.

89 Materials and methods

90 Description of study area

This study was carried out in the Greater Accra Region (GAR), one of the 16 administrative regions of Ghana. It is in the south-most part of the country and bordered by the Central Region to the west, Volta Region to the east, Eastern Region to the north, and the Gulf of Guinea to the south (Figure 1). It is the smallest land area of the 16 regions, with estimated population to be 5,055,805 [11]. The capital city of Ghana, Accra is also located in the region with a daily influx of people from within and outside the country. The region is the most urbanized and has the highest population density in the country [12]. There are highly populated slums, squatters and informal settlements in parts of the region due to the rapidurbanization being experienced [13, 14].

100 The region has 29 districts of which two are metropolitans; namely Accra and Tema and 23 municipalities. Twelve communities such as; Agbogbloshie, James Town, Adabraka, 101 Kaneshie, Maamobi, Nima, Chorkor, Mamprobi, Agege and Dansoman in the Accra 102 103 Metropolitan Assembly, and Teshie and Nungua in the Ledzokuku Krowor Municipality located in the cholera endemic communities. In addition, Dawhenya North, Kofikope, 104 Awhiam, and Dawa in the Ningo Prampram district; Addokope, Dogobom, Anvaman East, 105 Adjumanikope in the Ada West district; Adedetsekope, Angomya-Ada, Kophemm Kasseh 106 and Asigbekope in the Ada East district all located in the non-cholera endemic communities 107 108 shown in Fig 1.

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ALN-Ablekuma North, ABC-Ablekuma Central, ABW-Ablekuma West, ACC-Accra
Metropolis, KKT-Korle Klottey, WGB-Weija Gbawe, OKN-Okaikoi North, AYWAyawaso West, LDK-La Dade Kotopon, ASH-Ashaiman

113 Study design

A community-based cross-sectional design was used for this study. The study area was 114 classified into cholera endemic and non-endemic communities. The cholera endemic 115 communities were cholera 'hotspots' in the Accra Metropolitan Area, Ledzokuku and 116 Krowor Municipal Areas which have consistently been reporting cases of cholera over the 117 past five years, preceding start of this study, with evidence of local transmission. The 118 cholera non-endemic communities were the Ningo Prampram, Ada West and East Districts 119 that have rarely recorded any case of cholera over the same period with no evidence of local 120 transmission in the area. Household drinking water from randomly selected households 121 were sampled for bacteriological water quality assessment. The study was conducted in the 122 wet (April to August 2019) and dry (January to March 2020) seasons. 123

124 Sample size determination

125 The sample size was determined using the Cochran's formula: 126 $N= \frac{z^2 p(1-p)}{e^2}$ 127 e^2 128 $N= \frac{(1.96)^2 X \ 0.838(1-0.838)}{(0.05)^2}$ 130 N= 208.608

By assuming z: 95% confidence interval, e: 5% margin of error and p: proportion of households drinking water contaminated with *V. cholerae* as 83.8% reported in Accra Metropolis [15], to obtain a minimum sample size of 209 for each of the endemic and also the non-endemic communities for the wet season and repeated in the dry season.

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137 Sampling technique

138 A multistage cluster sampling approach was used to determine the household of which to sample the water. The first cluster represented a cholera endemic or non-endemic 139 community. The second stage involved the random selection of 12 communities from both 140 141 the endemic and also the non-endemic communities. The households were subsequently listed for each community and 9 households randomly selected by a ballot. An additional 142 water sample was collected from the main drinking water source in each of the communities 143 (e.g public tap, pipe, or dam) as a control sample. The 12 selected cholera endemic 144 145 communities were; Agbogbloshie, James Town, Adabraka, Dansoman, Agege, Mamprobi, 146 Chorkor, Kaneshie, Nima, Maamobi, Teshie, and Nungua. Whereas the 12 non-endemic communities were; Addokope, Dogobom, Adjumanikope, Anyaman, Adedetsekope, 147 Agomya-Ada, Kopehem-Kasseh, Asigbeykope, Awhiam, Dawa, Kofikope, and Dawhenya 148 149 North. In all 216 households drinking water and 24 from the water source were collected during the wet season and repeated in the dry season from the same households and water 150 sources to determine the temporal variation in water quality. 151

152 Water sample collection

Households' drinking water samples were collected for bacteriological quality analysis. 153 Head of households or representative were asked to provide a cup of drinking water as they 154 would give to a child or guest to be collected to ensure that they will provide the best of 155 water quality to sample. Approximately 500 ml of the stored household drinking water was 156 sampled using the container usually used to scoop water from the storage container into a 157 sterile plastic bottle. In case the household drank sachet water, they were collected and a 158 selected edge of the sachet water decontaminated by cleaning with 70% ethanol, then cut 159 160 aseptically with sterile scissors and the water poured into the sterile plastic bottle. On the other hand, if the household drank bottled water, they were only collected and labelled. The 161 water samples from the source point were directly collected from the pipe, standpipe, or 162 unprotected well. The collection of water from the tap or pipe water was done by cleaning 163 the outside of the tap with 70% ethanol and allowing flow at maximum for about a minute 164 before directly collecting into the sterile plastic bottle [16]. A drop of sodium thiosulphate 165 (0.5 ml of 10% solution) was added to the pipe-borne water to neutralize residual chlorine 166 before transporting it to the laboratory [16]. The water samples collected from the field were 167 transported in insulated boxes at 4°C to the laboratory within six hours. In case of a delay, 168 169 the water samples were stored in the refrigerator ($2^{\circ}C$ to $8^{\circ}C$) and processed the next day (within 24 hours). 170

171 Ethical statement

Prior to the initiation of this study, ethical approval was given from the Ghana Health
Service Ethical Review Committee (GHS-ERC) under the approval number GHS-ERC
006/01/19. Community leaders granted initial permission, followed by written consent from

the household's head or representative, using a structured consent form. The informed consent form was explained to the participants in a dialect they could understand by a research assistant. The participants were provided with information regarding the study's purpose, rationale, selection process, procedures, and interview duration.

Those individuals who willingly agreed to take part in the study expressed their consent by either signing the consent forms or providing a thumbprint. A duplicate of the signed or thumb printed consent form was furnished to each participant. Participation in the study was entirely voluntary, with an assurance that participants could withdraw at any point with no consequences. Confidentiality of the gathered information was guaranteed to all participants.

185 Laboratory analysis

The water samples were analyzed in accordance with the American Public Health 186 Association (APHA) standard methods for examining drinking and waste water using the 187 membrane filtration method as described by Eaton, Clesceri [17]. In instances that the 188 growth was heavy and uncountable (i.e. greater than 200 colony count), 10 ml, 1 ml, 0.1 ml, 189 or 0.01 ml of the water was used and diluted with buffered peptone water to make up the 190 100 ml before filtration. The 0.45µm membrane filter was aseptically removed and placed 191 in the respective appropriate media. The inoculated Plate Count Agar (PC) and Violet Red 192 Bile (VRB) agar were incubated at 36^oC±1, while the Eosin Methylene Blue (EMB) agar 193 and Thiosulfate-Citrate-Bile-Sucrose (TCBS) agar were incubated at 44°C±1. The plates 194 were observed after 24 hours and 48 hours. The plates for PC and VRB were counted and 195 estimated respectively for the total viable count and total coliform. The EMB was estimated 196 197 for faecal coliforms (thermotolerant coliforms) and TCBS for suspected Vibrio species.

198 Sub-culturing of colonies

The morphological characteristics of the colonies on the filter membrane were described. 199 200 Colonies that appeared as; large, smooth, yellow, slightly flattened, and translucent peripheries on the TCBS medium were presumptively identified as Vibrio species. These 201 colonies were sub-cultured onto another TCBS plate for purification. The suspected Vibrio 202 203 colonies were further sub-cultured on Tryptone Soy Agar (TSA) (a non-selective media) for biochemical testing and serotyping. Other suspected coliform bacteria growing on the PC 204 205 agar, VRB, EMB were sub-cultured on MacConkey agar plate for purification. Distinct colonies were sub-culture on TSA for presumptive identification and biochemical testing 206 207 using the Mini Antigenic Profile Index 20 Enterobacteriaceae (API 20E) ((bioMérieux, 208 France) for confirmation.

209 **Quality control**

All the containers used for the collection of water samples from the field were sterilized byautoclaving and checking with an autoclave tape to ensure sterilization.

All the bacteriological media were prepared in accordance with the manufacturer's instructions and dispensed into disposable pre-sterilized plastic Petri dishes. A sterility test was performed on each batch of media prepared by incubating a plate at 35 to 37^oC overnight (18 to 24 hours) and examined for any contamination.

A media performance test was also conducted by culturing in-house or reference bacteria strains obtained from the National Public Health and Reference Laboratory, Ghana to assess the growth characteristics of the media. The reference strains and in-house control strains were used as reference points to validate the results of the biochemical tests. Blank membrane filtration was carried out after every fifth sample. This was to ensure
growths on the filter membrane were not influenced by the laboratory conditions. The
filtration techniques were carried out in a laminar flow hood to avoid contamination as much
as possible.

224 Data processing and analysis

Result of the analysis of the water samples were entered into Microsoft Excel. It was 225 226 exported into STATA software version 14.0 (State Cooperation, USA), cleaned and analyzed. Descriptive statistics such as means and standard deviation for continuous 227 variables and frequencies and percentages for categorical variables were computed. 228 Inferential statistics were applied to estimate the water parameters such as the total coliform, 229 faecal coliform and vibrio counts. Kruskal Wallis rank test, a non-parametric equivalent of 230 the Analysis of Variance (ANOVA) test was performed to determine whether there were 231 any significant differences between the means of the log of bacteria counts among some 232 selected factor variables. 233

234 **Results**

235 Assessment of the bacteriological quality of drinking water and

236 cholera risk in Greater Accra Region

In the endemic communities, household drinking water stored in vessels was also analyzed in both seasons. Of the 216 water samples; 110 were private pipes, 23 were public pipes, and 83 were sachet water. High levels of total coliform counts (TCC) were observed in ten communities with a mean count of 7.73 log cfu/100ml and 9.86 log cfu/100ml respectively in the dry and wet seasons. The mean counts for faecal coliforms (FCC) were generally high 242 in the private pipe water sources in both seasons except in a few communities such as Agege and Dansoman which did not record any counts. Vibrio counts (VC) were absent in the 243 private pipe water sources except in Mamprobi, which recorded 6.91 log cfu/100ml in the 244 245 wet season. The direct public pipe also had a high mean TCC of 7.82 log cfu/100ml in the dry season and 9.64 log cfu/100ml in the wet season. The mean FCC was also found to be 246 247 5.24 log cfu/100ml and 10.06 log cfu/100ml respectively in the dry and wet seasons. No VC was observed in the communities (Agbogbloshie and James Town) during both seasons. 248 249 The highest mean TCC and FCC in both seasons were found in stored public pipe water 250 compared to stored private pipe water and sachet water in the households. Vibrio counts 251 were present in all household stored drinking water samples (Table 1).

Study variable	Characteristics	TCC (log cfu/ 100 ml)		FCC (log cfu /100 ml)		VC (log cfu /100 ml)	
, ui iubic		Dry	Wet	Dry	Wet	Dry	Wet
Direct water sources	Community	Mean(sd)	Mean(sd)	Mean(sd)	Mean(sd)	Mean(sd)	Mean(sd)
Private pipe	Adabraka	8.16	10.04	5.35	9.30	0	0
	Nungua	7.24	12.10	4.50	10.57	0	0
	Agege	8.07	8.85	0	0	0	0
	Chorkor	7.60	8.52	4.60	0	0	0
	Dansoman	7.74	9.39	0	0	0	0
	Kanehsie	8.16	10.93	0	6.91	0	0
	Mamobi	8.04	10.46	4.79	8.29	0	0
	Mamprobi	6.91	9.61	5.13	0	0	6.91
	Nima	7.60	8.52	4.79	8.52	0	0
	Teshie	7.74	10.20	4.50	10.13	0	0
	Total	7.73(0.41)	9.86(0.36)	4.81(0.32)	8.95(1.34)	0	6.91
Public pipe	Agbogbloshie	8.04	10.60	5.13	10.34	0	0
	James town	7.60	10.78	4.79	9.80	0	0
	Total	7.82(0.31)	9.64(1.29)	5.24(0.65)	10.06(0.38)	0	0
Water stored in vessels	Private pipe	8.26(0.38)	10.76(1.13)	5.53(0.45)	10.04(1.09)	2.40 (0.69)	9.91(1.15
	Public pipe	8.49(0.37)	11.75(0.38)	5.80(0.44)	11.16(0.76)	1.77(0.88)	9.76(1.52
	Sachet	8.45(0.39)	10.51(1.08)	5.60(0.41)	10.18(0.99)	2.29(0.71)	10.49(0.57

Table 1: Mean log of bacterial counts in the dry and wet seasons across some selected background characteristics in endemic
 communities

256	When the differences between the mean rank score of the log of TCC ($\Box^2 = 0.182$, p=0.669),
257	FCC ($\Box^2=2.591$, p=0.107) and VC ($\Box^2=0.200$, p=0.655) were assessed among the direct
258	water sources in the endemic communities, there were no statistical differences as shown in
259	Table 2. However, when the differences between the bacterial mean counts were assessed
260	for the stored household drinking water, the mean rank score of the log of TCC ($\Box^2=7.704$,
261	p=0.021), FCC (\Box^2 =2.591, p<0.001) and VC (\Box^2 =12.101, p=0.002) differed significantly
262	(Table 2). The bacterial counts were generally higher in the wet season than the dry season
263	during the period for both the water collected directly from the source and that stored in
264	vessels in the households.
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Table 2: Assessing the differences in log bacterial count among household drinking water stored in vessels and from direct water sources in endemic

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communities

C	ommunities				
		n	Rank Sum	\Box^2 with ties	P-Value
	To	tal coliform c	ount		
Direct source				0.182	0.669
Private Pipe		4	55.50		
Public Pipe		20	244.50		
Water stored in	ı vessels			7.704	0.021
Private Pipe	Stored	110	12068.50		
Public Pipe S	tored	23	3198.50		
Sachet water		83	8169		
	Fac	ecal coliform of	count		
Direct source				2.591	0.107
Private Pipe		4	70.50		
Public Pipe		20	229.50		
Water stored in	ı vessels		I	17.156	<0.001
Private Pipe	Stored	110	12170		
Public Pipe S	tored	23	3532.50		
Sachet water		83	7733.50		
L		Vibrio coun	t		
Direct source				0.200	0.655
Private Pipe		4	48		
Public Pipe		20	252		
Water stored in	ı vessels	I	1	12.101	0.002
Private Pipe	Stored	110	12094		
Public Pipe S	tored	23	3289		
Sachet water		83	8053		

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In the non-endemic communities also, 24-direct water sources were sampled from private pipe (4), public pipe (16), and unprotected wells (4) in both seasons (Table 3). The direct private piped water sources were found in Dawhenya and Kofikope and recorded a mean TCC of 8.57 log cfu/100ml in the dry season and 10.12 log cfu/100ml in the wet season. The mean FCC was 4.50 log cfu/100ml and 8.61 log cfu/ml in the dry and wet seasons respectively, with no recorded VC. The public pipe samples recorded a mean FCC of 8.44 287 log cfu/ml in the dry season and 9.64 log cfu/ml in the wet season. The mean FCC was 5.24 log cfu/100ml in the dry season and 9.39 log cfu/100ml in the wet season, although six of 288 the communities (Dawa, Dogobom, Adedetsekope, Asigbeykope, Awhiam, Adjumanikope) 289 290 did not record any feacal coliforms. Anyaman East community, on the other hand, recorded the highest TCC and FCC in both seasons. Except for Awhiam community that had a VC 291 292 of 6.91 log cfu/100ml, all other communities did not record any counts. The unprotected wells also recorded a mean TCC of 8.61 log cfu/100ml in the dry season and 10.20 log 293 cfu/100ml in the wet season. The mean FCC was 5.53 log cfu/100ml in the dry season and 294 295 9.45 log cfu/100ml in the wet season with a mean VC of 10.31 log cfu/100ml.

Study variable	Characteristics	TCC (log cfu/ 100 ml)		FCC (log cfu /100 ml)		VC (log cfu /100 ml)	
		Dry	Wet	Dry	Wet	Dry	Wet
Direct water sources	Community	Mean(sd)	Mean(sd)	Mean(sd)	Mean(sd)	Mean(sd)	Mean(sd)
Private pipe	Dawhenya	8.48	10.09	0	8.70	0	0
	Kofikope	8.66	10.16	4.50	8.52	0	0
	Total	8.57(0.13)	10.12(0.06)	4.50	8.61(0.13)	0	0
Public pipe	Addokope	8.63	9.21	4.79	0	0	0
• •	Dawa	8.29	0	0	0	0	0
	Dogobom	8.34	8.00	0	0	0	0
	Adedetsekope	8.27	0	0	0	0	0
	Anyaman East	8.81	10.60	5.70	9.39	0	0
	Asigbeykope	8.57	0	0	0	0	0
	Awhiam	8.48	10.76	0	0	0	6.91
	Adjumanikope	8.16	0	0	0	0	0
	Total	8.44(0.22)	9.64(1.29)	5.24(0.65)	9.39	0	6.91
Unprotected Dug well	Angomya-Ada	8.41	11.00	5.67	9.80	0	10.31
	Kopeheum Kasseh	8.81	9.39	5.39	9.10	0	0
	Total	8.61(0.28)	10.20(1.14)	5.53(0.19)	9.45(0.49)	0	10.31
Water stored in vessels	Private pipe	8.92(0.26)	11.10(0.68)	6.16(0.36)	10.27(0.71)	1.84(0.62)	9.92(1.24)
	Public pipe	8.77(0.26)	10.45(1.13)	6.03(0.34)	9.59(1.08)	2.23(0.92)	9.73(1.08)
	Sachet	8.88	12.21	5.74	10.93	0	0
	Unprotected dug well	8.81(0.31)	11.16(0.61)	6.10(0.40)	10.39(0.77)	2.48(0.59)	11.19(0.78)
	Dam	9.05(0.23)	11.83(0.13)	6.44(0.23)	10.60(0.18)	2.91(0.77)	9.24(0.34)

Table 3: Mean log of bacterial counts in the dry and wet seasons across some selected background characteristics in non-endemic
 communities

299 The differences in the mean rank score between the log of FCC among the direct water sources were assessed and found to be statistically significant ($\Box^2 = 10.212$, p=0.006). 300 Whereas the mean rank score of the log of TCC and VC among the direct water sources did 301 not differ ($\Box^2=5.171$, p=0.075: $\Box^2=2.029$, p=0.363) as shown in Table 4. Two hundred and 302 sixteen (216) households stored drinking water in the non-endemic communities made up 303 of 34 private piped water, 136 public pipe water, 36 unprotected dug well, 8 dam water and 304 2 sachet waters stored in the household. Except VC, which was not found in sachet water, 305 TCC, FCC, and VC were generally present in all household water stored in vessels for the 306 307 period. The bacterial counts in the stored water samples were higher than those collected from the direct water sources. The TCC and FCC were generally observed to be higher 308 309 during the wet season than the dry season. The differences between the bacterial mean 310 counts were assessed for the household drinking water stored and a statistically significant difference was found in the mean rank score of the log of TCC and FCC ($\Box^2=14.165$, 311 p=0.007: \Box^2 =33.901, p<0.001), whiles the mean rank scores for log of VC (\Box^2 =6.051, 312 313 p=0.195) was not significant (Table 4).

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Table 4: Assessing the differences in log bacterial count among household drinking water stored in vessels and from direct water sources in non-endemic

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water stored in vessels and from direct water sources in non-endemic communities

communities		Rank Sum	\Box^2 with ties	P-Value
	n Total colifor			P-value
	I otal collior	m count	C 171	0.075
Direct source		(7.50	5.171	0.075
Private pipe	4	67.50		
Public pipe	16	163		
Unprotected dug well	4	69.50		
Water stored in vessels			14.165	0.007
Unprotected dug well	36	4423.0		
Dam water	8	1189.5		
Private pipe	34	4357.5		
Public pipe	136	13168.0		
Sachet	2	298.0		
	Faecal colifor	·m count		
Direct source			10.212	0.006
Private pipe	4	63.50		
Public pipe	16	155.50		
Unprotected dug well	4	81		
Water stored in vessels			33.901	<0.001
Unprotected dug well	36	4972.0		
Dam	8	1257.5		
Private pipe	34	4743.0		
Public pipe	136	12205.5		
Sachet	2	258.0		
	Vibrio co	ount		
Direct source			2.029	0.363
Private pipe	4	46		
Public pipe	16	195.50		
Unprotected dug well	4	58.50		
Water stored in vessels			6.051	0.195
Unprotected dug well	36	4206.5		
Dam	8	1152.0		
Private pipe	34	3525.5		
Public pipe	135	14226.0		
Sachet	2	110.0		

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Out of 120 household drinking water samples that were collected, in the endemic communities for each of the seasons, 76 (63.3%) and 82 (68.3%) positive growths were found respectively in the wet and dry seasons. In some of the water samples, multiple growths of the bacteria were found. Among the positive isolates, the predominant organism occurring in both seasons was *Klebsiella pneumoniae* 24 (31.6%) followed by *Escherichia*

- *coli* 13 (17.1%). There were no confirmed *Vibrio cholerae* strains in the household drinking
- 331 water samples in the endemic communities (Table 5).

Table 5: Microorganisms isolated from the household drinking water samples collected in the endemic communities

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	Wet Season		
Organisms	Frequency	Percent	
Acinetobacter baumanni/calcoaceticus	1	1.3	
Citrobacter freundii	6	7.9	
Enterococcus faecalis	1	1.3	
Enterobacter sp	9	11.8	
Enterococcus sp	3	4.0	
Escherichia coli	13	17.1	
Klebsiella pneumoniae	24	31.6	
Proteus mirabilis	8	10.5	
Proteus vulgaris	1	1.3	
Pseudomonas aeruginosa	9	11.8	
Serratia ficaria	1	1.3	
Total	76	100.0	
	Dr	y Season	
Acinetobacter baumanni/calcoaceticus	3	3.7	
Citrobacter freundii	7	8.5	
Enterobacter sp	11	13.4	
Enterococcus sp	10	12.2	
Escherichia coli	16	19.5	
Klebsiella pneumoniae	23	28.1	
Proteus mirabilis	7	8.5	
Pseudomonas aeruginosa	5	6.1	
Total	82	100.0	

335

A hundred and twenty (120) drinking water samples were sampled from the non-endemic 336 337 communities in each of the seasons (wet and dry). 104 (86.7%) isolates were identified during the wet season (Table 6). Of the 11 different bacteria identified in the wet season, 338 Escherichia coli 18 (17.3%) was the most common isolate followed by Klebsiella 339 pneumoniae 17 (16.4%). In the dry season, 88 (73.3%) had positive growth with Klebsiella 340 pneumoniae (22.7%) as the most frequently occurring followed by Escherichia coli 341 (18.2%). There were also multiple growths in some of the water samples. No Vibrio 342 cholerae strains were isolated in the household drinking water samples. 343

	Wet Season		
Organisms	Frequency	Percent	
Acinetobacter baumanni/calcoaceticus	2	1.9	
Citrobacter freundii	7	6.7	
Enterobacter sp	13	12.5	
Enterococcus sp	11	10.6	
Escherichia coli	18	17.3	
Klebsiella pneumoniae	17	16.4	
Proteus mirabilis	15	14.4	
Proteus vulgaris	2	1.9	
Pseudomonas aeruginosa	16	15.4	
Staphylococcus aureus	2	1.9	
Yersinia pestis	1	1.0	
Total	104	100.0	
	Dr	y Season	
Aeromonas hydrophilia	2	2.3	
Acinetobacter baumanni/calcoaceticus	1	1.1	
Citrobacter freundii	7	8.0	
Enterobacter sp	14	15.9	
Enterococcus sp	10	11.4	
Escherichia coli	16	18.2	
Klebsiella pneumoniae	20	22.7	
Proteus mirabilis	7	8.0	
Pseudomonas aeruginosa	11	12.5	
Total	88	100.0	

Table 6: Microorganisms isolated from the household drinking water samples collected in the non-endemic communities

346

347 **Discussions**

The bacteriological quality of water can be affected by the natural presence of microorganisms or from contamination by human activities, which could pose a risk to cholera and other diarrhoeal diseases [18]. In this study, we assessed the bacteriological quality in direct water sources and household drinking water storage.

In the cholera endemic communities, the water source collected for drinking was from a private pipe or public pipe. In the non-endemic communities on the other hand the water sources were from private, public and unprotected wells. This study demonstrated that the direct pipe, public pipe, unprotected well water sources collected from most of the

communities during the dry and wet season were contaminated with total and/or faecal 356 coliform counts above the WHO and Ghana standards of zero (0) cfu/100 ml for drinking 357 water both in the endemic and non-endemic communities, making them unwholesome for 358 359 consumption [19, 20]. These findings are consistent with other studies that recorded high total and faecal coliform counts in directly collected piped and well water [21, 22]. The high 360 bacterial counts in the piped water sources may be attributed to the practice of laying 361 362 pipelines in gutters and the lack of maintenance of the pipelines. Inadequate chlorination may also account for these high bacterial counts as reported by Karikari and Ampofo [23] 363 364 in an earlier study that revealed unsuitable chlorine levels and the presence of faecal coliform in the Accra water network. The difference in bacteria counts between the direct 365 pipe and public taps in the endemic communities did not show any statistical significance, 366 367 indicating that their contamination levels were similar.

The same was the bacteria counts in the non-endemic communities except faecal coliform 368 counts that showed significant differences in private, public and unprotected well water. The 369 private and public piped water are supplied by Ghana Water Company and may just reflect 370 their service delivery that needs improvement to reduce the risk of cholera and diarrheal 371 372 disease transmission. The unprotected wells may have run-off water contamination of the wells especially during the rainy season that might account for the high counts from the 373 374 direct well water. This notwithstanding, the direct main public tap water in Dawa, 375 Aedetsekope, Asigbeykope, and Adjumanikope did not record any coliform counts and was considered wholesome by the standards in the wet season. These communities have a 376 community public standpipe located at a central point in the community and either 377 378 connected to a borehole or directly to a high-level elevated tank, thus have few pipeline connections, unlike in the endemic communities where the public standpipes were 379 connected to the general pipeline distribution and subject to more contamination. 380

With regards to seasonality, higher total and faecal coliform counts were generally observed 381 in the wet season than in the dry season in both the endemic and non-endemic communities. 382 This finding is in agreement with other studies that similarly found higher total and faecal 383 coliform counts in the wet season than the dry season [24, 25]. This may occur because of 384 the poor environmental conditions around some of these water sources in the communities. 385 386 In this study, the water stored for drinking by the households was also assessed for the level of contamination. This source water was collected and stored mostly in plastic containers 387 388 and a few in earthenware/pot. The study results revealed that irrespective of the water source (a private pipe, public pipe, sachet water or well) the contamination levels for total 389 and faecal coliforms were higher than that directly collected from the water source. In the 390 endemic communities, our study found a significant difference between the bacterial 391 392 contamination levels and the water sources stored in the household. This was similarly observed in the non-endemic communities, except the Vibrio count which did not show any 393 significant differences with the water sources stored. This study is consistent with the 394 findings of Meierhofer, Wietlisbach [26] in Kenya which found higher contamination levels 395 in the storage containers. The reports of Boateng, Tia-Adjei [27] and Agensi, Tibyangye 396 397 [28] in Tamale, Ghana and Kisoro, Uganda respectively, are similarly in line with this study findings. The cleanliness of the storage vessels, methods of drawing water from the storage 398 399 container and the length of the storage were implicated for the high bacterial counts [9, 10]. 400 This study findings also revealed that sachet water recorded similarly high levels of contamination with total and faecal coliforms as the private pipe and public tap water. 401 402 Earlier studies in Ghana and Nigeria have variously indicated sachet water to be 403 contaminated with faecal coliform and other pathogenic bacteria and not wholesome [29-31]. Semey, Dotse-Gborgbortsi [32] explained in their study that the contamination might 404 be due to the unhygienic handling by factory workers during production. It may also occur 405

if the source water used in the packaging is already contaminated without any further 406 treatment. Dzodzomenyo, Fink [33] have further indicated that some of the sachet water 407 companies are not formally registered by the Food and Drug Authority and might be 408 violating the guidelines of the standard for guality water production. It is therefore not 409 surprising that Aslan, Rochani [29] study which examined the sachet bag observed some 410 bags not to have a batch number, date of manufacture and no information on treatment. This 411 412 is worrisome as the shift to drinking sachet water might not necessarily be protective against cholera. 413

414 Further analysis was conducted to confirm the suspected Vibrio isolates that were mostly detected in the household drinking water storage in both seasons. The most frequently 415 isolated organisms in both seasons were Escherichia coli and Klebsiella pneumoniae. The 416 presence of these organisms indicates the possibility of the presence of potentially harmful 417 bacteria and a high risk of its transmission The suspected Vibrio counts observed were 418 confirmed to be negative for Vibrio cholerae, instead were identified as Pseudomonas 419 aeruginosa, Proteus sp. and Aeromonas hydrophilia. Our findings did not detect the 420 presence of V. cholerae and contrasts that of Yirenya-Tawiah, Darkwa [15] that found 421 422 Vibrio cholerae O1 strains in household drinking water storage in some communities in Accra just before the major cholera outbreak in 2014. The absence of *V. cholerae* in the 423 424 current study could explain the reason for the absence of a cholera outbreak during the study period. The findings further strengthen the importance of regular environmental surveillance 425 to predict cholera outbreaks. 426

427 Strength and limitation

The findings of this study indeed have significant implications for understanding householddrinking water quality and its relationship with cholera transmission in the Greater Accra

Region. By examining the current state of water quality, the study provides valuable insights 430 that can inform policymakers, public health authorities and other stakeholders in their efforts 431 to combat cholera and improve overall water and sanitation practices in the region. 432 However, the type of storage vessels used was not accounted for and could be a significant 433 limitation. Different types of storage vessels may have varying levels of susceptibility to 434 contamination, and this could impact the overall quality of the water. To address these 435 436 limitations, future research or follow-up studies could consider incorporating the assessment of storage vessel types and their impact on water quality. 437

438 Conclusion and recommendations

The household drinking and direct water sources were contaminated with total and faecal 439 coliform counts that exceeded the Ghana and WHO recommendation in both the endemic 440 and non-endemic communities. The bacterial counts followed a seasonal pattern peaking 441 during the wet season than the dry season, possibly resulting from contamination of water 442 sources from run-off flood waters. Escherichia coli and Klebsiella pneumoniae were the 443 most common coliforms recovered with no V. cholerae strains isolated from both the direct 444 or stored household drinking water. The presence of these coliform bacteria gives an 445 indication of faecal contamination and a high risk of waterborne diseases. 446

It is important therefore, to educate households members to treat their drinking water at the point-of-used to avoid drinking contaminated water and reduce the risk of transmission of cholera and other diarrhoeal pathogens. It is also important for the Ghana Standard Authority to regularly monitor the bacteriological quality of the sachet water manufacturing companies to ensure their operations conform to the quality standards.

452

453 Data availability

The raw data supporting the study could be made available by authors without any

455 reservation.

456 **Conflict of interest**

457 The authors declare that they have no conflict of interest.

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480 **Reference**

- 1. Bain, R., et al., *World Health OrganizationGeneva, SwitzerlandBartram, J.(2014)*. Global assessment of exposure to faecal contamination through drinking water based on a systematic review, EM&IH J. Volume19, Issue8vol. **19**: p. 917-927.
- 2. Wright, J., S. Gundry, and R. Conroy, *Household drinking water in developing countries: a systematic review of microbiological contamination between source and point-of-use.* Tropical medicine & international health, 2004. **9**(1): p. 106-117.
- 3. D'Mello-Guyett, L., et al., *Prevention and control of cholera with household and community water, sanitation and hygiene (WASH) interventions: a scoping review of current international guidelines.* PLoS One, 2020. **15**(1): p. e0226549.
- 4. Cronin, A., et al., A review of water and sanitation provision in refugee camps in association with selected health and nutrition indicators—the need for integrated service provision. Journal of water and health, 2008. **6**(1): p. 1-13.
- 5. Mengel, M.A., et al., *Cholera outbreaks in Africa.* Curr Top Microbiol Immunol, 2014. **379**: p. 117-44.
- 6. Ghana Disease Surveillance Department, *Cholera Hotspots in Greater Accra Region*. 2015, Ghana Health Service. p. 1 5.
- 7. Organization, W.H., *Guidelines for drinking-water quality: first addendum to the fourth edition.* 2017.
- 8. Yeboah, S.I.I.K., P. Antwi-Agyei, and M.K. Domfeh, *Drinking water quality and health risk assessment of intake and point-of-use water sources in Tano North Municipality, Ghana.* Journal of Water, Sanitation and Hygiene for Development, 2022. **12**(2): p. 157-167.
- 9. Lartey, K.A., Assessing the Quality of Household Drinking Water in Selected Communities in the Akuapem South District, in Department of Biological, Environmental and Occupational Health Sciences, School of Public Health. 2019, University of Ghana: University of Ghana <u>http://ugspace.ug.edu.gh</u>. p. 1-64.
- 10. Asfaw, H.S., M.A. Reta, and F.G. Yimer, *High enteric bacterial contamination of drinking water in Jigjiga city, Eastern Ethiopia*. Ethiopian Journal of Health Development, 2016. **30**(3): p. 118-128.
- 11. Ghana Statistical Service, 2010 Population and Housing Census District Analytical Report. Accra Metropolitan. 2014.
- 12. Ghana Statistical Service, 2010 Population and Housing Census, National Analytical Report. 2013.
- 13. Amoako, C., Brutal presence or convenient absence: The role of the state in the politics of flooding in informal Accra, Ghana. Geoforum, 2016. **77**: p. 5-16.
- 14. Accra Metropolitan Assembly and UN Habitat, *Participatroy Slum upgrading and revention: Millennium city of Accra, Ghana. UN Habitat Annual report, Accra.* 2011.
- 15. Yirenya-Tawiah, D.R., A. Darkwa, and M. Dzodzomenyo, *Environmental surveillance for Vibrio cholerae in selected households' water storage systems in Accra Metropolitan Area (AMA) prior to the 2014 cholera outbreak in Accra, Ghana*. Environmental Science and Pollution Research, 2018. **25**(28): p. 28335-28343.
- 16. Cheesbrough, M., *District Laboratory Practice in Tropical Countries*, ed. 2nd. Vol. Part 2. 2006, UK: Cambridge University Press, Cambridge.
- 17. Eaton, A.D., et al., *Standard Methods for the Examination of Water & Wastewater*. 2005: American Public Health Association.
- 18. WHO, Guidelines for drinking-water quality: first addendum to the fourth edition. WHO library Cataloguing-Publication Data. 2017.
- 19. WHO, *Guidelines for drinking-water quality*. 1993: World Health Organization.

- 20. Ghana Standard Authority, *Limits for drinking water (GS 175-1)*. 2009.
- 21. Tekpor, M., et al., *Bacteriological quality of drinking water in the Atebubu-Amantin district of the Brong-Ahafo region of Ghana.* Applied Water Science, 2017. **7**(5): p. 2571-2576.
- 22. Oyedum, U., N. Adabara, and F. Kuta, *Comparative study of coliform contamination of public boreholes and pipe borne water systems in Bosso town, North Central, Nigeria.* Journal of Applied Sciences and Environmental Management, 2016. **20**(2): p. 234-238.
- 23. Karikari, A. and J. Ampofo, *Chlorine treatment effectiveness and physico-chemical and bacteriological characteristics of treated water supplies in distribution networks of Accra-Tema Metropolis, Ghana.* Applied Water Science, 2013. **3**(2): p. 535-543.
- 24. Dongzagla, A., S. Jewitt, and S. O'Hara, *Seasonality in faecal contamination of drinking water sources in the Jirapa and Kassena-Nankana Municipalities of Ghana*. Science of the Total Environment, 2021. **752**: p. 141846.
- 25. Odonkor, S.T. and T. Mahami, *Escherichia coli as a Tool for Disease Risk Assessment of Drinking Water Sources*. International Journal of Microbiology, 2020. **2020**.
- 26. Meierhofer, R., B. Wietlisbach, and C. Matiko, *Influence of container cleanliness, container disinfection with chlorine, and container handling on recontamination of water collected from a water kiosk in a Kenyan slum.* Journal of water and health, 2019. **17**(2): p. 308-317.
- 27. Boateng, D., M. Tia-Adjei, and E.A. Adams, *Determinants of household water quality in the Tamale Metropolis, Ghana.* Journal of Environment and Earth Science, 2013. **3**(7): p. 70-77.
- 28. Agensi, A., et al., Contamination potentials of household water handling and storage practices in kirundo subcounty, kisoro district, Uganda. Journal of environmental and public health, 2019.
 2019.
- 29. Aslan, A., et al., *Sources of microbiological contamination in sachet water from Ghana*. Journal of Water, Sanitation and Hygiene for Development, 2020. **10**(2): p. 202-208.
- 30. Mosi, L., et al., *Microbiological assessment of sachet water "pur e water" from five regions in Ghana [version 2; peer review: 2.* 2019.
- 31. Oluwafemi, F. and M.E. Oluwole, *Microbiological examination of sachet water due to a cholera outbreak in Ibadan, Nigeria.* 2012.
- 32. Semey, M.D., et al., *Characteristics of packaged water production facilities in Greater Accra, Ghana: implications for water safety and associated environmental impacts.* Journal of Water, Sanitation and Hygiene for Development, 2020. **10**(1): p. 146-156.
- 33. Dzodzomenyo, M., et al., *Sachet water quality and product registration: a cross-sectional study in Accra, Ghana.* Journal of water and health, 2018. **16**(4): p. 646-656.