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Attributes of drinking water, sanitation, and hygiene associated with microbiological water quality in stored drinking water in rural Mozambiquan and Ugandan schools

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Abstract

Contaminated drinking water causes morbidity and mortality worldwide, especially in low- and middle-income countries. Drinking water quality has been studied extensively in household settings, but little research is available on drinking water quality in schools. School settings are of particular importance, because children are more susceptible than adults to a variety of diseases from contaminated drinking water. Many school water, sanitation and hygiene (WaSH) interventions have been studied for their efficacy to reduce diarrheal disease incidence, but few have evaluated drinking water quality, which is an important exposure pathway between WaSH services and health outcomes. Using school surveys developed from internationally established WaSH indicators and field microbiological water quality tests, we studied 374 rural schools in Mozambique and Uganda to understand the association between specific WaSH services and drinking water microbiological contamination, specifically testing most probable number (MPN) of *Escherichia coli*, an indicator of fecal contamination. In Mozambique and Uganda, 71% and 83% respectively of rural schools had low risk drinking water quality (<1 *E. coli*/100mL); thirteen percent and seven percent had very high-risk water quality (≥ 100 *E. coli/100 ml*). When accounting for all WaSH services studied, schools that used an improved water source had 0.22 times less *E. coli* in stored drinking water in Mozambique (95% CI: 0.07, 0.65) and 0.12 times less *E. coli* in Uganda (95% CI: 0.02, 0.80). In Mozambique, use of a water source within 30 minutes and the presence of water and soap/ash for handwashing were also significantly associated with less *E. coli* in drinking water. The findings of this study provide public health practitioners with implementable WaSH services to improve school drinking water quality, which has implications for the health, learning environment, and cognitive development of school children in rural Mozambique and Uganda.

Introduction

Contaminated drinking water continues to cause substantial morbidity and mortality worldwide (Clasen et al., 2007; Hunter et al., 2010; Wolf et al., 2018a). As of 2019, drinking water sources of an estimated two billion people were contaminated with feces and over 800,000 people die annually from diarrhea caused by poor water, sanitation, and hygiene, including nearly 300,000 children (World Health Organization, 2019). Community settings, like schools and health facilities, have become spotlights for water, sanitation, and hygiene (WaSH) programming in low-income countries (LICs), to reduce disease exposure in commonly frequented areas outside the home. WaSH services that protect users from pathogen exposure remains limited in these settings (Guo et al., 2017; Morgan et al., 2017) and poorly financed (Alexander et al., 2016; McGinnis et al., 2017), and monitoring for quality of services in countries is severely limited (United Nations Children's Fund and World Health Organization, 2018; World Health Organization and United Nations Children's Fund, 2019).

Evidence from household studies for effective WaSH strategies informs interventions in school and other extra-household settings. Meta-analyses of household WaSH interventions show household water treatment, safe storage, and sanitation interventions reduce diarrheal risk (Hunter, 2009; Wolf et al., 2014) and intestinal protozoa infections (Speich et al., 2016). In cross-sectional studies, the type of water source and residual chlorine treatment are consistent predictors of *Escherichia coli* (*E. coli*) contamination (Gupta et al., 2007; Jeandron et al., 2019; Trevett et al., 2004), but various water storage and usage practices show no association in Honduras (Trevett et al., 2004).

Schools offer a particular opportunity for improving health related to WaSH and water quality, as children spend a significant portion of their daytime hours in school. While household water quality has improved, school-aged children remain exposed to waterborne disease through poor school drinking water quality. Further, children are more susceptible than adults to diarrheal disease and other waterborne illnesses (Jasper et al., 2012). Water treatment with hygiene or sanitation in schools decreases respiratory illness (Patel et al., 2012) and parasitic infections (Erismann et al., 2017; Freeman et al., 2013), and increases health-related knowledge and behaviors (Chard and Freeman, 2018; Hetherington et al., 2017), enrollment, and gender parity (Garn et al., 2013). School water treatment and handwashing interventions also reduce absenteeism, especially for girls (Trinies et al., 2016). School hand hygiene interventions alone also reduce absences due to a variety of respiratory and enteral infections (Talaat et al., 2011). Lastly, schools are a learning environment and have the potential to be places where safe WaSH practices are taught to students (Anthonj et al., 2021), and these teachings can then be shared with their families and communities (Bresee et al., 2016; Dreibelbis et al., 2014).

Many researchers have tested the effect of school WaSH interventions on diarrheal disease incidence or missed school days (McMichael, 2019), but few have evaluated the quality of drinking water (microbiological or chemical) as an important intermediary in this causal relationship. As improvement of drinking water quality is a main mechanism for how school WaSH interventions improve health of school-aged children, we study microbiological stored drinking water quality in 374 rural schools in Mozambique and Uganda and analyze the association between school WaSH services and microbiological water quality. We highlight implementable WaSH services associated with better microbiological drinking water quality in rural schools in Mozambique and Uganda. These school WaSH services have the potential to reduce diarrhea incidence and absenteeism and improve educational outcomes in these settings.

Methods

Sampling, study population, data collection tools

The sampling methodology, survey instrument, and study population have been previously described (Morgan et al., 2017). Briefly, we conducted a stratified random sample of schools in specific rural regions of ten sub-Saharan African countries; here we describe results from the schools in

Mozambique and Uganda that had stored drinking water and collected a water sample from it. These two countries were selected from the original study as the random sample of schools with water quality samples from stored drinking water was sufficiently large in each. Data collection consisted of two components: a standardized survey instrument to evaluate access, quantity, quality, continuity, and reliability of WaSH services (previously described (Morgan et al., 2017)); and microbiological water quality testing of stored drinking water. GPS coordinates of schools were also collected.

Ethics

Free and informed participant consent was obtained from all school officials surveyed. The Institutional Review Board of the University of North Carolina at Chapel Hill approved this study protocol on June 3, 2014 (IRB Reference ID: 14-0763). This study was approved by the corresponding national governing bodies: the Uganda National Council for Science & Technology (UNCST) and the Directorate of Water in the Ministry of Public Works and Housing in Mozambique.

WaSH factors analyzed

We analyzed descriptive statistics of WaSH services of the 374 rural schools in Mozambique and Uganda, including water source type, access (distance to source), storage, removal; sanitation facility type and condition; and hand hygiene access. Estimates were weighted based on stratified random sampling in selected rural regions, to account for different probabilities of selection of schools. We tested the association of these WaSH factors in schools on school microbiological water quality using enumerated *E. coli.* We describe the variables and regression model here.

We used the WHO/UNICEF Joint Monitoring Programme categorizations of “improved” and “unimproved” types of water source and sanitation (World Health Organization, 2012). Improved-type drinking water sources decrease the risk of fecal contamination compared to unimproved-type drinking water sources, but do not guarantee microbial water safety (Bain et al., 2014; Shaheed et al., 2014). Improved-type water sources include piped water, boreholes, protected dug wells or springs, rainwater, and packaged water. Improved-type sanitation limits human contact with excrement, and include flush sewer systems, septic tanks, ventilated pit latrines, composting toilets, and pit latrines with slabs. We analyzed water storage by observing the use of a safe container (covered container, with a narrow opening, or with a wide opening and water treatment) and safe removal methods (pouring, spigot, tap, or long ladle for extracting water) (Centers for Disease Control, 2014). We analyzed hand hygiene facilities by assessing the presence of water and soap/ash for handwashing, a widely used indicator for hand-hygiene access (UNICEF and World Health Organization, 2018). We also analyzed the presence of materials for hand-drying.

The conditions of improved-type sanitation in schools were observed by the presence of the following aspects of each sanitation facility: doors, doors that can be closed, doors with locks, holes in the structure, stability of latrine slab, caving walls of the structure, latrine pits that were too large, latrine pits that were caving in, used paper on slabs, and flies swarming.

Water quality testing

We used *E. coli* as the microbial indicator as the organism is an indicator of fecal contamination, it does not grow naturally in the environment (Edberg et al., 2000), and field-based and laboratory testing are available, inexpensive, and are simple to conduct in rural areas of LICs. In each school, a 100mL water sample was collected from the stored drinking water consumed by students, used by the school, in the same way members of the school extract water for drinking. We analyzed water quality using Aquagenx (Chapel Hill, NC) Compartment Bags to determine the most probable number (MPN) of *E. coli* (Stauber et al., 2014) according to the manufacturer’s instructions (Aquagenx, 2013).

Regression model

WaSH indicators measured in the survey were treated as ordinal predictor variables, and MPN of *E. coli* in stored drinking water as a discrete outcome variable. Predictor variables were selected for the model based on potential for contamination of stored drinking water and were indicator variables for the following WaSH services: improved-type water source; treatment of stored water; safe container for stored drinking water; safe removal method of stored drinking water; water source within 30 minutes roundtrip; improved-type sanitation; water and soap/ash present for handwashing. The association between each WaSH factor and water quality was tested in bivariate (unadjusted) analysis using a negative binomial regression model (El-Shaarawi et al., 1981), because our outcome, concentration of *E. coli*, was discrete and overdispersed around zero. Unadjusted covariates that were significant at $p < 0.05$ or considered necessary to control for (e.g. treatment of water) were included in a multivariate (adjusted) model. Tests for collinearity of these factors were conducted before inclusion in an adjusted model, and interaction terms between predictors were evaluated. Model results are reported as incidence rate ratios.

Frequencies were calculated using PROC SURVEYMEANS and PROC SURVEYFREQ in SAS 9.4 (SAS Institute, Cary, NC, USA). The negative binomial regression model was computed using *nbreg* with the *irr* option in Stata 14 (StataCorp, College Station, TX, USA). Figures for descriptive statistics were generated using R 3.6.0. Maps were produced using the “sf” package in R 3.6.0. Schools with missing GPS points were mapped to their respective districts.

Results

School demographics

We studied 374 rural schools, 124 in Mozambique and 250 in Uganda, serving 206,487 total students (Table 1). Districts sampled were geographically disparate rural areas (Figure 1), and sampled schools were predominantly primary schools. The median number of students enrolled was 374 in Mozambique and 510 in Uganda (Table 1). In Mozambique, the median numbers of boy and girl students were 191 (IQR: 35, 366) and 172 (IQR: 66, 322), respectively. In Uganda, the median numbers of boy and girl students were 238 (IQR: 180, 372) and 263 (IQR: 190, 375), respectively.

[Table 1]

Descriptive statistics

Water quality

Water quality of stored drinking water in rural schools is not geographically clustered (Figure 1). Seventy-one percent of rural schools in Mozambique and 83% in Uganda had <1 *E. coli* MPN/100 mL (Figure 2A), the lowest health risk in the latest WHO classification (World Health Organization, 2017). Thirteen percent of rural schools in Mozambique and seven percent in Uganda had ≥100 *E. coli* MPN/100mL, WHO's highest health risk category. Fewer schools in each country fell into the intermediate risk categories. Boreholes, an improved-type water source, were the most common water source used by rural schools in both countries (Figure 2B). Stored drinking water of the highest health risk level (>100 *E. coli* MPN/100 mL) was found in rural schools with improved-type water sources in both countries (piped sources in Mozambique, boreholes and protected springs in both countries, and rainwater in Uganda).

[Figure 1]

[Figure 2]

Water source type and storage

Of schools with water sources, eighty-nine percent of rural schools in Mozambique and 95% in Uganda had an improved-type water source, and 92% and 85%, respectively, had a water source within 30 minutes of the school, including collection time (Figure 3A). Forty-eight percent of rural schools in Mozambique and 78% in Uganda had safe storage containers, and 19% and 62%, respectively, had means for safe removal of drinking water. Three percent of schools in Mozambique reported no storage of drinking water because they had on-plot water sources.

[Figure 3]

Ten percent and 17% of schools reported treatment of drinking water in Mozambique and Uganda, respectively. Treatment methods included boiling and chlorine in both countries, with one school in Uganda reporting filtration.

Sanitation type and quality

Sanitation facilities were predominantly of an improved type, though the conditions of sanitation facilities varied. Sixty-two percent in Mozambique and 91% in Uganda had improved-type sanitation facilities. Of schools with improved-type sanitation, the most notable problem was a lack of doors: only 46% of rural schools in Mozambique and 58% in Uganda had doors on all latrines. (Figure 3B).

Hand hygiene

The availability of handwashing facilities was notably absent: only 2% of rural schools in Mozambique and 14% in Uganda had water and soap or ash for handwashing present on the day of the survey. Only 2% of rural schools in each country had water, soap or ash, and drying materials for handwashing present.

Regression model

Several WaSH factors in rural Mozambique schools had unadjusted estimates that significantly correlated with *E. coli* MPN in stored drinking water (Table 2). Schools with an improved-type water source had 0.29 (95% CI: 0.13, 0.64) times the incidence rate of *E. coli* in stored drinking water compared with schools with unimproved-type water sources. Schools with water sources within 30 minutes for collection had 0.28 (95% CI: 0.12, 0.68) times the incidence rate of *E. coli* as schools with more distant water sources. Schools with safe storage containers had 3.71 (95% CI: 1.38, 9.92) times the incidence rate of *E. coli* compared with schools without safe containers, and schools with means for safe removal of stored water (e.g. with a tap or ladle) had 2.47 (95% CI: 1.10, 5.54) times the incidence rate of *E. coli* as schools that did not. Schools with water and soap/ash for handwashing on the day of the survey had 0.04 (0.01, 0.19) times the incidence rate of *E. coli* than with schools without these materials for handwashing. Schools that also had hygienic materials for hand-drying, in addition to water and soap/ash, on the day of the survey had 0.08 (0.06, 0.13) times the incidence rate of *E. coli* as schools without all three handwashing materials. In testing for collinearity, none of these variables had correlation coefficients above 0.8, and were all included in an adjusted model.

[Table 2]

In an adjusted model with the selected predictors of water quality in Mozambique, an improved water source (IRR: 0.22, 95% CI: 0.07, 0.65), water sources within 30 minutes (IRR: 0.25, 95% CI: 0.08, 0.81) and water and soap/ash for handwashing present (IRR: 0.12, 95% CI: 0.02, 0.73) remained significant, each associated with less *E. coli*. A similar adjusted model is observed when the handwashing indicator includes materials for drying. An improved water source and water sources within 30 minutes have similar incidence rate ratios, and water, soap/ash, and drying materials are associated with 0.27 times the incidence rate of *E. coli* (95% CI: 0.10, 0.76).

Predictors of water quality in schools in Uganda showed a different picture. Schools with piped water sources or other improved-type water sources had 0.09 (95% CI: 0.02, 0.55) times the incidence rate of *E. coli* as schools with unimproved water sources. Schools that treated water had 2.36 (95% CI: 4.22, 11.22) times the incidence rate of *E. coli* as schools that didn’t treat water. Lastly, schools with improved sanitation had 0.28 (95% CI: 0.10, 0.74) times the incidence rate of *E. coli* as schools with unimproved or no sanitation.

No collinearity was observed, and these three predictors were included in an adjusted model for Uganda. In the adjusted model, only an improved-type water source remained a significant predictor of water quality (IRR: 0.12, 95% CI: 0.02, 0.83). In both countries, neither the method of water treatment (boiling or chlorination) nor specific conditions of latrines on visual inspection were associated with amount of *E. coli* contamination.

Discussion

We provide one of very few studies of rural school drinking water quality in LIC settings and that assesses the WaSH factors associated with safer microbiological drinking water quality in rural schools. Drinking water quality in both countries was good overall: 71% of schools in Mozambique and 83% in Uganda had <1 *E. coli* MPN/100 mL, with most schools drawing drinking water from boreholes. These water quality estimates are similar to previously published data from these countries (Agensi et al., 2019; Holcomb et al., 2020). In both Mozambique and Uganda, an improved-type water source was associated with less *E. coli* in unadjusted and adjusted models, with a piped water source in Ugandan schools additionally associated with less *E. coli*. This finding is not surprising, based on water quality evidence from household monitoring (Kirby et al., 2016; Shields et al., 2015). In Mozambique, additional significant predictors included a water source within 30 minutes and handwashing materials present on the survey day (water and soap/ash). Proximity to water source affects health in multiple ways. First, longstanding evidence from households suggests closer water sources leads to increased water quantity and better hygiene, regardless of water quality (Stelmach and Clasen, 2015). Second, and more specific to waterborne diarrheal disease, schools with closer water sources likely refill storage containers more often, which presents fewer opportunities for contamination of water sitting in containers for longer periods of time. This is consistent with previous research in households that show an increase in disease incidence as distance from a water sources increases (Wang and Hunter, 2010).

In unadjusted models, rural schools in Mozambique with a safe storage container had 3.52 times the *E. coli* incidence rate in stored drinking water than schools that did not use a safe storage container; schools with means to remove drinking water safely had 2.47 times the *E. coli* incidence rate compared with schools that did not. The adjusted model for Mozambique controlled for these safe storage components, as well as water source type, source within 30 minutes for collection, treatment of stored water, and hand hygiene. In the adjusted model, use of a safe storage container and safe removal of water no longer significantly increased incidence rate of *E. coli.* Further, schools with handwashing facilities (water and soap/ash present for handwashing) had 0.12 times the *E. coli* incidence rate compared with schools without hand hygiene, while controlling for other significant WaSH predictors. This evidence suggests that water and soap/ash for handwashing reduces incidence rate of *E. coli* in school drinking water in Mozambique, regardless of safety of water storage. Handwashing with soap and water reduces the fecal contamination of hands of students and teachers accessing stored water, which can lead to less *E. coli* contamination of stored drinking water. Although studies of handwashing in households have established this relationship (Wolf et al., 2018b), our findings are the first to show that in schools, handwashing materials are associated with substantially less *E. coli* in stored water, regardless of stored water practices.

In unadjusted models of rural schools in Uganda, treatment of water and improved-type sanitation were significant predictors of *E. coli,* in addition to water source type. Reported water treatment was associated with increased incidence rate of *E. coli* in drinking water, contrary to expectation. In the adjusted model, when controlling for use of a piped water source or other improved-type water source, improved-type sanitation, and treatment of water, water treatment was not associated with increased *E. coli;* only a piped water source or other improved-type remained significant. These findings suggest that of the WaSH services studied in Uganda, water source type most significantly predicts *E. coli* incidence rate in drinking water. In both countries, specific conditions of latrines were not significantly associated with amount of *E. coli* contamination, a finding that has been similarly observed with sanitary inspections and water quality of water handpumps (Kelly et al., 2021).

Limitations

There are limitations to this research. First, we did not sample microbial contaminants at the various points of study (water sources, door handles and other surfaces of sanitation facilities, hygiene facilities), nor at timepoints between storage and use, so we cannot isolate exact times or places of contamination. This is worth noting given the JMP "improved" and "unimproved" type classification does not involve sanitary inspection of water sources or sanitation facilities. Evidence from households suggests such recontamination of drinking water after collection is common (Levy et al., 2008). We also did not sample water at different points in time throughout the year, as seasonality can affect water quality. Second, while most indicators used were observations, we rely on self-reported time to water source due to insufficient GPS collection at the water source, which is less accurate than Euclidean distance measured with GPS (Ho et al., 2014). Finally, as this study was part of a larger multi-site, multi-country evaluation, we were unable to test other microbiological or physiochemical parameters associated with poor health outcomes through additional laboratory evaluation, but would recommend it in future studies.

This study concerns WaSH services associated with water quality in rural schools, in order to inform future school WaSH interventions that can be most successful in improving water quality, and thus, reducing disease incidence and absenteeism. Past studies that found school WaSH interventions had no effect on reducing disease incidence or absenteeism evaluated whole WaSH programs, not individual WaSH services, or relied on self-reported disease (Chard et al., 2019; Garn et al., 2017), while implementation of specific interventions, such as handwashing with soap and additional toilets in a recent study of schools in Nepal, has been shown to reduce intestinal parasitic infections (Shrestha et al., 2020). To avoid inaccuracies of subjective disease recall in the study of school WaSH interventions, future studies could focus on objective measures of disease, such as enteric pathogen antibodies (Chard et al., 2018) or on water quality (such as *E. coli* in stored water as a proxy for disease risk).

These results support the findings of water quality studies in other household and extra-household settings that show sanitation facilities and hand hygiene influence water quality, in addition to water source type and water treatment (Guo and Bartram, 2019; Holcomb et al., 2020). As we highlight water, sanitation, and hygiene services associated with school drinking water quality, our findings complement a recent study evaluating school system factors and water quality (Cronk et al., 2020), which found fewer schools in Mozambique and Uganda to have the lowest WHO risk water quality (61% and 56%, respectively, compared with our findings of 71% and 83%), despite sampling occurring after the present study. These findings, specifically a suggested worsening in school drinking water quality, are important for policymakers, school administrators, and public health practitioners in Mozambique and Uganda, as they provide specific WaSH services with potential to improve water quality and subsequently the health, learning environment, and cognitive development of their young people.

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Table 1: Demographics of rural schools studied

Country	Districts sampled	Schools with water quality samples (n)	Median total students (IQR)	Median male students (IQR)	Median female students (IQR)	Median number of teachers (IQR)
Mozambique	13	124	374 (133, 698)	191 (35, 366)	172 (66, 322)	6 (3, 15)
Uganda	10	250	510 (372, 757)	238 (180, 372)	263 (190, 375)	10 (7, 13)

IQR: Interquartile range

Figure 1: Geographic locations of surveyed schools in Uganda (A) and Mozambique (B).

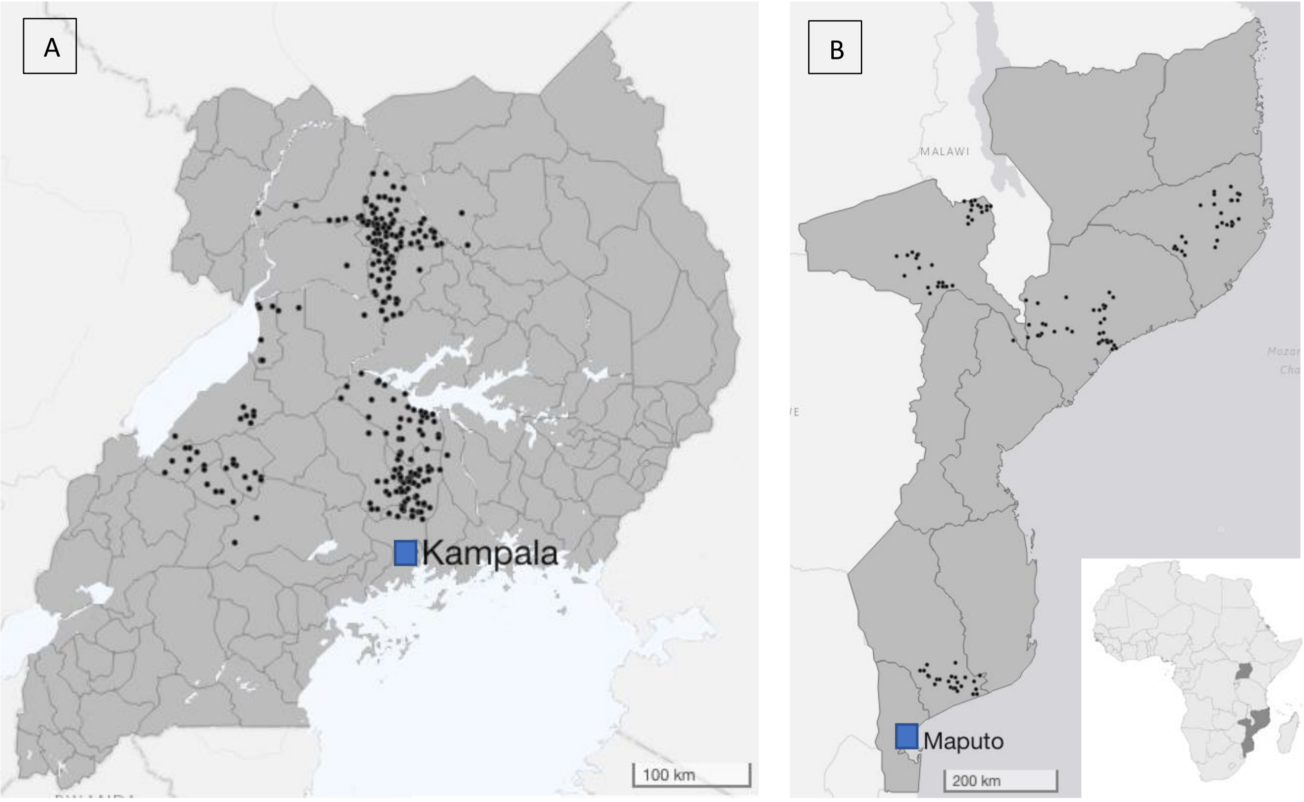


Figure 2. A: Weighted percent of rural schools with drinking water in each WHO water quality risk level based on *E. coli* MPN. B: Weighted percent of rural schools in each WHO water quality risk level by school water source type. Boreholes were the most common drinking water source type in schools. Other drinking water sources were less frequent; as such, the y-axis is shown on a smaller scale.

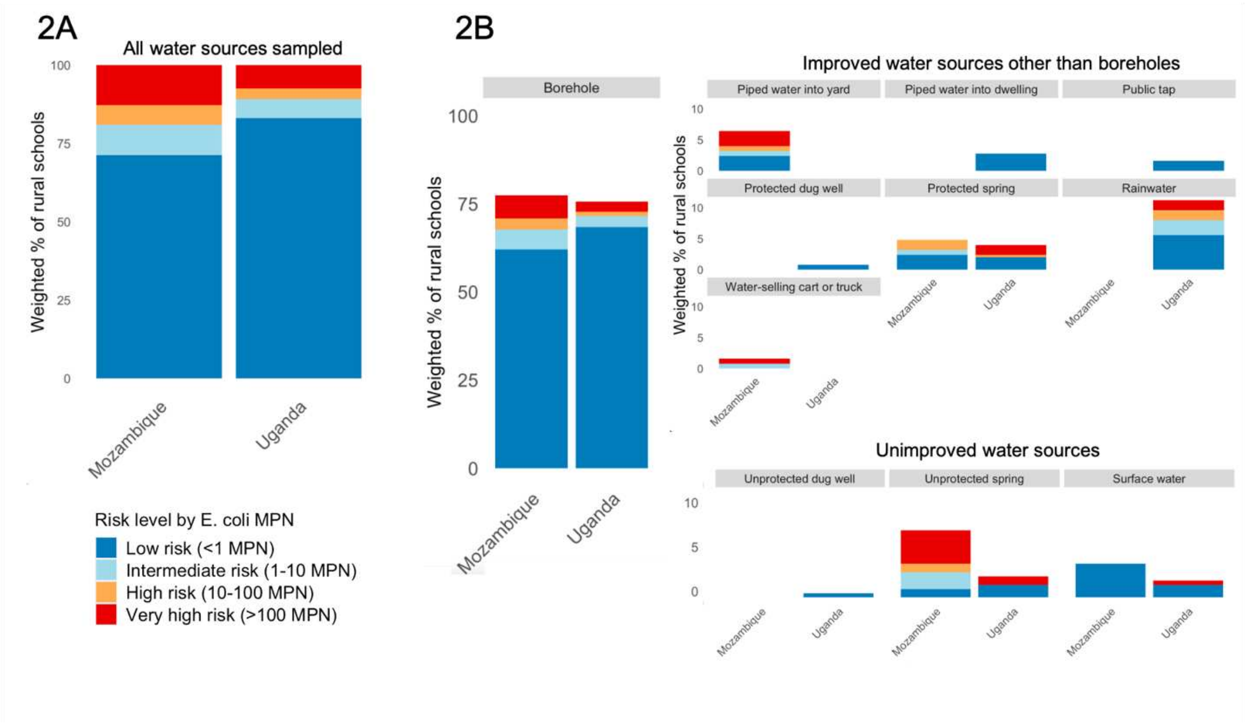
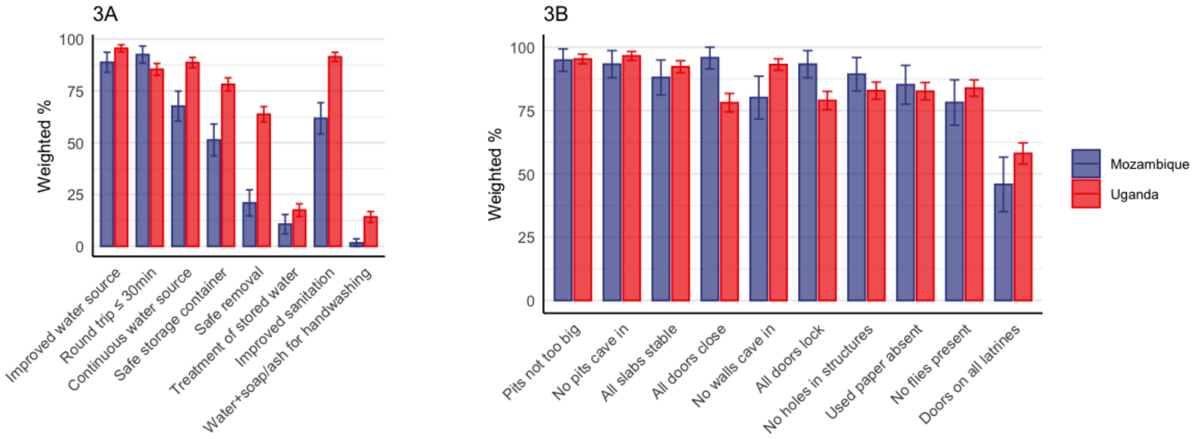


Figure 3: A: Descriptive statistics of water quality predictors. B: Condition of improved sanitation facilities.



Error bars indicate 95% confidence intervals of population-level weighted percentages.

Table 2: Incidence rate ratios (IRR) for *E. coli* most probably number in stored drinking water by water, sanitation, and hygiene service

	Mozambique			***Uganda***		
	N	**Unadjusted IRR (95% CI)**	**Adjusted IRR (95% CI)**	**N**	**Unadjusted IRR (95% CI)**	**Adjusted IRR (95% CI)**
Improved water source	**124**	**0.29 (0.13, 0.64)**	**0.22 (0.07, 0.65)**	**250**	**0.09 (0.02, 0.55)**	**0.12 (0.02, 0.80)**
Water treatment	118	1.07 (0.30, 3.84)	0.39 (0.11, 1.33)	**250**	**2.36 (4.22, 11.22)**	1.75 (0.68, 4.52)
Continuous water source	118	5.53 (0.80, 38.15)		250	2.24 (0.33, 15.21)	
Safe Container	**124**	**3.52 (1.32, 9.40)**	3.58 (0.84, 15.19)	250	2.13 (0.71, 6.43)	
Safe Removal	**124**	**2.47 (1.10, 5.54)**	1.01 (0.29, 3.55)	249	1.98 (0.81, 4.86)	
Round trip < 30 min (self-reported)	**124**	**0.28 (0.12, 0.68)**	**0.25 (0.08, 0.76)**	250	0.61 (0.23, 1.65)	
Improved sanitation	123	0.94 (0.48, 1.87)		**250**	**0.28 (0.10, 0.74)**	0.44 (0.15, 1.29)
Feces absent on all latrines	123	3.26 (0.87, 12.22)		250	1.57 (0.58, 4.22)	
Water + soap/ash for handwashing observed	**122**	**0.04 (0.01, 0.19)**	**0.12 (0.02, 0.73)**	250	1.09 (0.36, 3.35)	
Water +soap/ash +drying materials observed for handwashing	**122**	**0.08 (0.06, 0.13)**	**0.27 (0.10, 0.76)**	250	2.30 (0.38, 13.93)	

§In Uganda, the variable for main water source included a category for piped sources. In Mozambique, this variable was binary, with only two options, improved and unimproved water sources.

Bold covariates are significant at $p < 0.05$

CI: confidence interval

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