Title: Interpreting drinking water quality samples: understanding contamination pathways at the point of collection

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Abstract

2.2 billion people lack access to safely managed drinking water, with microbiological water quality the major barrier to the achievement of SDG target 6.1. Microbiological water quality is strongly influenced by environmental and hand hygiene. We demonstrate the significant contribution of contamination from hygiene at water collection point localities and at the household level to drinking water quality. Comparison of water quality samples from 5,080 water systems at the 'point of delivery' (PoD) to those from the 'point of collection' (PoC) demonstrated that 60% of the *E. coli* positive results at the PoC were caused by contamination from local hygiene-related pathways, with seasonal variability. Additionally, household hygiene contamination pathways contributed a further 5% to 70% of *E. coli* positive detections in samples from the 'point of use' (PoU) based on analysis of 38 Multiple Indicator Cluster Survey (MICS) datasets. Better reporting of PoC and PoD methods is required to recognise this contribution from poor hygiene and ensure data are interpreted appropriately, thereby enabling more efficient targeting and prioritisation of interventions. Further revisions to drinking water quality sampling protocols are required to reflect the increase in (unchlorinated) piped water systems with potential for considerable biofilm growth, with implications for release of *Pseudomonas* and other opportunistic pathogens during sampling. There is immediate benefit to be gained from expanding communication and hygiene education to encourage cleaning of taps and spouts for drinking water.

Keywords

Drinking water, sampling methods, *E. coli*, faecal contamination, point of collection

1 Introduction

Faecal contamination is the primary limiting factor to get universal access to safely managed drinking water (Greenwood et al., 2024). An estimated 2.2 billion people lack access (UNICEF/WHO JMP, 2023), but adequate and available data on water quality is limited. Recent approaches to address these data gaps through modelling have estimated the number is closer to 4.4 billion people globally without access to safety managed drinking water (Greenwood et al., 2024). This large estimate is realistic given the seasonal variability in water quality (Bangladesh Bureau of Statistics and UNICEF Bangladesh, 2021; Charles et al., 2022), however, to advance drinking water safety we need to understand the implications of the data, and particularly, how it was sampled.

There are three common methods for sampling drinking water quality: sampling water from taps that have been cleaned, taps that have not been cleaned, and from a vessel served at the household level "as you would give a child to drink." Here and throughout this paper we use the term 'tap' to refer both to the devices that control outflow from reticulated systems and the spouts of handpumps. We propose that water samples that are collected from cleaned taps should be termed Point of Delivery (PoD) samples. International standards on water quality sampling generally recommend that sampling be done at this point. To ensure a clean tap, the tap should be disinfected by flaming (or appropriate disinfection method)

and flushed (APHA/AWWA/WEF, 2018; British Standards Institution, 2006; International Organization for Standardization, 2006), or a clean tap should be chosen (APHA/AWWA/WEF, 2018). These methods for *E. coli* and faecal indicator bacteria sampling are commonly derived from chlorinated, piped systems (Charles et al., 2020).

Water that is sampled from taps that have not been cleaned can be termed Point of Collection (PoC) samples. Samples collected in the household by asking for a cup of water "as you would give a child to drink" are termed Point of Use (PoU) samples. These sampling practices are common in international development, such as the Multiple Indicator Cluster Survey (MICS) approach to nationally representative water sampling (JMP/UNICEF/WHO, 2016). PoC sampling is included in international standards to "know the quality of the water as it is consumed" which is considered to be appropriate for "special situations, e.g. outbreaks" (International Organization for Standardization, 2006). Water systems in low- and middle- incomes countries typically include a higher proportion of small and non-piped water systems. Private on-premises connections are less prevalent and this has implications for primary contamination hazards.

Sampling drinking water from the PoC is becoming a de facto standard in international development work. Early guidance developed for small community supplies (WHO, 1997) differentiated approaches to sampling 'water as consumed', collected with flushing and cleaning with a cloth, from 'quality of water excluding contamination from the tap' which required flaming and flushing. Fifteen years later, guidance for the rapid assessment of drinking-water quality was aiming to expand understanding of the extent to which "quality of drinking-water from improved sources deviates from the assumption that it is safe" with large scale testing programmes; here the methodology was refined to cleaning the "tap or outlet with a clean, dry cloth" and then flush it before sampling (WHO and UNICEF, 2012). As these approaches expanded into UNICEF's MICS, the methodology for *E. coli* sampling reduced cleaning further to what is now accepted as PoC samples, specifying only that water be flushed for 30 seconds before sampling (JMP/UNICEF/WHO, 2016). Other guidance does not specify how to sample (e.g. CDC, n.d.).

While sampling at the PoC is increasingly common, the details of the sampling method are often not explicit in publications, reducing the comparability of results. A review of 171 articles that report faecal indicator bacteria results from PoC and PoU drinking water quality sampling in low- and middle-income countries found that 44% did not reference a standard method for sample collection and processing (Sikder et al., 2021). Sikder et al. (2021) did not comment on PoC cleaning practices but they noted that 61% of studies did not report anything about sampling sterility – here the focus was the sterility of sampling vessels and whether the sampler sanitized their hands. This lack of standardisation in reporting presents challenges in interpreting results, and limits the potential for aggregation of studies. A previous systematic review (Bain et al., 2014) of faecal contamination of drinking water in LMICs did not consider sampling methods, including papers using both PoD methods (e.g. Luby et al., 2006) and PoC methods (Mertens et al., 1990; Rufener et al., 2010). In more recent literature we find the same challenge, with recent literature on water quality in Bangladesh exhibiting variable methodology reporting between studies, often providing no data on sampling methods (Ercumen et al., 2017a; Escamilla et al., 2013; Ferguson et al.,

2011) , or reporting variable methods for either flushing (Ercumen et al., 2017b; Ferguson et al., 2011) or full decontamination for PoD sampling (Doza et al., 2020).

PoD, PoC and PoU sampling methods support understanding of different hazards. Wright et al. (2004) suggested that the PoD samples can lead to an understating of 'source contamination', however, this misrepresents the utility of these different types of samples. Mertens et al. (1990) demonstrated the difference in water quality from communal tubewells between PoD and PoC: at the PoD only 5% of samples were contaminated with faecal coliforms and contamination was at 'low levels' compared to at PoC, when around 50% of samples were contaminated, with average contamination similar to levels found in unprotected shallow wells. In their work, over 30 years ago, Mertens et al. reflected that communal water points are sites of activity for humans and animals, highlighting contaminated hands as a potentially important source of contamination. Contamination at the PoC is important for public health, but it should be recognised that the contamination is coming from a wider range of pathways than just the water system, with local hygiene being a primary consideration. At the PoU, still further pathways of contamination contribute to the quality of water assessed, including potential sources of contamination in the household management of drinking water, in transport and storage, as well as localised hygiene issues. It is widely recognised that contamination commonly increases between the PoC and PoU (Bain et al., 2021; Wright et al., 2004).

These three sampling methods evaluate the risks to the drinking water consumer in different ways, capturing different contamination pathways. In this paper, we analyse how different contamination pathways impact on water quality sample results, drawing on secondary data from international and project level surveys to compare results from PoD, PoC and PoU samples. We explore how interpretations of these different types of sample results differ for drinking water management and the implications for advancing drinking water safety. Lastly, we share results on the unintended results of cleaning taps for sampling in unchlorinated piped systems. Overall, we conclude that the role of local hygiene conditions in drinking water contamination at the point of collection are largely overlooked.

2 Materials and methods

To compare contamination of PoD, PoC and PoU drinking water quality samples with *E. coli* we utilised multiple secondary data sets. *E. coli* is the most precise indicator of recent faecal contamination in drinking water (Charles et al., 2020). To frame the analysis we broadly characterise the contamination pathways in Figure 1, where PoD captures contamination from the water catchment environment, such as groundwater contamination from latrines, and from infrastructure, such as leaks in pipes. PoC additionally captures pathways associated with 'local hygiene conditions'; this potentially includes contamination from users hands and animals that touch the tap. PoU capture pathways associated with collection container contamination, storage behaviours, and 'household hygiene', the latter is defined for the purposes of this paper as the contribution of hygiene to drinking water contamination in the process of accessing the water and the hygiene of the drinking vessel.

[insert here] Figure 1. Drinking water contamination pathways contributing to sample results at PoD, PoC and PoU

There are three components to the analysis. Firstly, we explore the relative contribution of the 'local hygiene' contamination pathways to drinking water quality through comparison of samples at the PoD and PoC to expand the empirical evidence for the difference in these measures. Secondly, we focus on the contribution of the 'household hygiene' pathway at the household level, addressing the contribution to water quality at the PoU through comparison of with samples at PoC that indicated no contamination. Lastly, we recognise the lack of reflective evidence that is available on these methods, and particularly on their adaptation to the diverse range of water systems in use. For this we contribute an analysis of PoD and PoC sampling method results in different types of water systems, to advance discussion on considerations when sampling.

2.1 Contribution of contamination pathways from PoD and PoC

Datasets that included paired waterpoint samples for *E. coli* for PoD (sampled after disinfection or decontamination and flushing) and PoC (sampled with flushing but no cleaning) were used to compare the contamination pathways. This sampling approach is applied by the Laboratory of Environmental Health, iccdr,b across their research and quality assurance programmes in Bangladesh (Table 1). 'Tap' is used throughout to identify the point of delivery for the water for simplicity, but data is almost exclusively from handpumps on tubewells (>99%) with the exception of Study 8. These data are largely from the Rohingya refugee camps in Bangladesh's Cox's Bazar peninsula, thus they are not representative of the national level of contamination (Bangladesh Bureau of Statistics and UNICEF Bangladesh, 2021). Study 8 in hospitals is of water piped through the hospital for use in healthcare provision; drinking water points are often separate from these larger piped networks.

The sampling methodology instructs that waterpoints be flushed for approximately two minutes before the PoC sample is taken. The waterpoint is then decontaminated, either flamed with alcohol-soaked cotton wool for metal or cleaned with an alcohol wipe for plastic outlets, in line with international standards (APHA/AWWA/WEF, 2018; International Organization for Standardization, 2006). The waterpoint is then flushed and the PoD sample is taken. All samples are stored in coolers with ice maintaining the temperature at 4 to 10°C for transport and analysed within 24 hours of sampling in the Laboratory of Environmental Health at icddr,b using membrane filtration (see individual studies for more details). For the analysis in this paper, the data were pooled across the datasets with PoD-PoC pairs analysed to compare the different levels of contamination using descriptive statistics in SPSS (version 28). Results were plotted in a Sankey plot using the networkD3 package with R version 4.1.1. Results are presented using the WHO risk categories: <1 cfu/100 mL = low risk; 1 – 10 cfu/100 mL = intermediate risk; 11 – 100 cfu/100 mL = high risk; and >100 cfu/100 mL = very high risk.

Table 1. Summary of Bangladesh water quality datasets used in the analysis. All data collected and analysed by icddr,b.

	Site	System types	Sampling months	N of paired samples	Programme
1	Narail, Faridpur, Pirojpur	Household and community	02/2019	N=286	Baseline survey for ASWA-II (DFID, 2017)

	and Sylhet districts	tubewells with handpumps			
2	Rohingya refugee camp	Shallow and deep tubewells, taps on piped systems	01-02/2018, 10-11/2019, 01-02/2020, 01/02,2021	N=946	Quality assurance programme
3	Rohingya refugee camp	Shallow and deep tubewells with handpumps	03-12/2018	N=3161	Study of contamination pathways (Mahmud et al., 2019)
4	Matlab	Shallow and deep tubewells with handpumps	11/2019	N=125	School water quality assessment (Fischer et al., 2021)
5	Khulna	Shallow and deep tubewells, pond sand filter, taps on piped systems	01/2020, 06/2020, 03/2021	N=299	Seasonal assessment of community and school water points (Hoque et al., 2021)
6	Rohingya refugee camp	Tubewells with handpumps	01-02/2022	N=128	Bhasan Char site
7	Matlab	Tubewells with handpumps	04/2018	N=31	Water Quality Monitoring
8	7 hospitals across 7 districts	Deep tubewell supplying building piped water network	10-12/2022, 5-6/2023, 10-11/2023, 2/2024	N = 104	Hospital water quality

2.2 Contribution of household hygiene contamination pathways at the PoU

Data from MICS were accessed for 38 datasets that represent countries or areas that collected data on water quality for MICS round 6 (2021-23). Data were not population weighted so they represent the samples taken. MICS water quality sampling methodology uses PoC samples and PoU samples, for which household respondents are asked to provide a cup of water “as you would give a child to drink”. 100mL samples are analysed by membrane filtration using compact dry growth media plates and incubated with body heat (Bangladesh Bureau of Statistics and UNICEF Bangladesh, 2021; JMP/UNICEF/WHO, 2016).

For the analysis in this paper, two-tailed Pearson correlations on national rates of contamination were performed in IBM SPSS v28. The increase in contamination between PoC and PoU is widely reported, and from various contamination pathways. Here we specifically focus on contamination pathways associated with household hygiene, excluding water handling practices in the household such as storage. The analysis included only households where a) water is not contaminated at the PoC (PoC = 0 *E. coli* per 100mL) and b) PoU samples were collected from a cup that the respondent filled directly from the PoC so that there is no household storage or treatment. Using this subset, the proportion demonstrating contamination with *E. coli* at the PoU from these local hygiene-related pathways was calculated.

2.3 Contribution of contamination pathways in unchlorinated piped water systems

Dataset 8 (Table 1) was collected at hospitals, sampling from various water access points on the sites. All water originated from tubewells on site but were accessed in different ways and for different purposes. These included: tubewell samples, including from handpumps (n = 22), taps connected to piped systems for drinking water (n = 26), taps connected to piped systems for healthcare use (n = 56). Piped systems used for healthcare use included storage tanks. No disinfection was present in any of the systems. In addition to *E. coli* results, water samples were analysed for bacterial indicators linked to the presence of biofilms (Makris et al., 2014), including *Pseudomonas* are an opportunistic pathogen which is a particular concern for immunocompromised patients which can form biofilms in piped systems and is associated with hospital acquired infections (Loveday et al., 2014). Samples were taken as for other studies. The analytical methods are briefly described. Total coliforms: 100mL samples were filtered through a 0.22 µm pore size membrane filter (Millipore Corp.), which was then placed on membrane fecal coliform (MFC) agar plates. After incubation at 37°C for 18 to 24 hours, blue coloured colonies were counted (Islam et al., 2001). Total bacterial count: 100 µl samples were inoculated onto a nutrient agar plate using the drop plate technique (Hoben and Somasegaran, 1982) and incubated at 37°C for 18 to 24 hours. The total number of colonies were counted. *Pseudomonas aeruginosa*: 100 ml samples were filtered using a 0.22 µm pore size membrane filter (Millipore Corp.) and placed onto a cetrimide agar (BD Difco) plate. The plate was incubated at 35±2℃ for 18 to 48 hours. After incubation, colonies surrounded by a blue-green pigment and fluoresced under short wavelength (254 nm) ultraviolet light were presumptively identified and counted as *Pseudomonas aeruginosa* (Crone et al., 2020; Zimbro et al., 2009). This dataset was used to explore the impact of decontaminating the tap in different systems including unchlorinated piped systems. Descriptive statistics were used to assess the change in concentrations of pathogens and indicators with decontamination.

3 Results

3.1 Contribution of contamination pathways from PoD and PoC

The analysis of secondary datasets at scale allowed assessment of the contribution of contamination from (a) water catchment environment and infrastructure pathways as detected at the PoD, and (b) local hygiene pathways as detected as additional contamination at PoC. The eight studies provided 5,080 paired samples of water quality in Bangladesh (Table 2). Contamination with *E. coli* from environment and infrastructure pathways (PoD method) was detected in 7.3% of samples (n=370). Contamination with *E.*

coli in samples using PoC methods was present in 14% of samples (n=712), with local hygiene pathways associated with 48.2% of detection PoC. Only one PoD-PoC pair was *E. coli* detected in a PoD sample where it hadn't been detected in the PoC sample. Across the studies, 6.7% of taps were contaminated. Figure 2 demonstrates the change in risk class between PoD and PoC methods (for those taps where *E. coli* was detected at PoC): 60% classified as low risk (*E. coli* absent) at PoD, with an additional 16.0% having a reduced risk category after cleaning (lowered *E. coli* concentration).

Table 2 Comparison of risk category from drinking water samples at PoD and PoC

	Risk class		Water as delivered (PoD)				Total
			Low	Intermediate	High	Very high	
Water as collected (PoC)	Low	N	4379	1	0	0	4380
		%	100.0	0.0	0.0	0.0	*100*
	Intermediate	N	297	123	2	0	422
		%	70.4	29.1	0.5	0.0	*100*
	High	N	40	73	58	0	171
		%	23.4	42.7	33.9	0.0	*100*
	Very high	N	6	9	32	72	119
		%	5.0	7.6	26.9	60.5	100
Total		N	4722	4722	206	92	72
		%	92.7	92.7	4.1	1.8	1.4

[insert here] Figure 2 Contribution of local hygiene contamination pathways at the tap: changes in risk category from drinking water samples at PoD and PoC, for the subset of samples with E. coli detected before decontamination.

The majority of samples are from tubewells in the Rohingya refugee camps and are not representative of Bangladesh nationally, nor are they equally representative of all water system types. Across the eight studies, the proportion of samples with no *E. coli* detected at the PoD ranged from 40.5% to 97.9%, and for PoC ranged from 25.8% to 90.4% (Table 3), with variability by system type and season described below. The contribution of water catchment environment and infrastructure pathways varied, with high contamination in studies 6 and 8. High variability between the contribution of local hygiene pathways, as indicated by the difference in the risk level between PoD and PoC sampling, was also evident: ranging from 0% to 81.8%. More complex infrastructure was associated with a greater contribution of contamination from the environment and infrastructure pathway: water from water points connected to unchlorinated piped networks was more frequently and heavily contaminated compared to water from tubewells.

Table 3. Variation in contributions of contamination from different pathways

Study	n	PoD low risk	PoC low risk	Contamination from local hygiene pathways	Contribution of local hygiene pathways to overall contamination	Contamination increased by local hygiene pathways
		PoD = 0	PoC = 0	PoC-PoD	PoC >0, PoD = 0	PoC= > 0, PoD < PoC
		%	%	%	%	%
1	286	97.9	88.5	9.4	81.8	90.9
2	946	96.2	90.4	5.8	60.4	75.8
3	3161	95.5	89.6	5.9	56.8	72.0
4	125	94.4	88.8	5.6	50.0	85.7
5	299	89.0	78.3	10.7	49.2	49.2
6	128	43.0	25.8	17.2	23.2	54.7
7	31	87.1	87.1	0.0	0.0	0.0
8	116	40.5	30.2	10.3	16.0	30.9

As seasonality has been found to significantly affect water quality (Charles et al., 2022), results were compared by sampling months for school, community and household settings (Table 4) excluding study #8. Across the studies, there were small seasonal variations in *E. coli*, with *E. coli* more commonly present in the colder, drier winter period of December to February. The contribution from water catchment environment and infrastructure pathways was greatest in the December to February period. The contribution from local hygiene pathways was greatest in the monsoon (June – August) and post-monsoon periods (September – November) periods. The contribution of local hygiene pathways was lowest in the pre-monsoon (March – May). This variability demonstrates the differing importance of these pathways seasonally.

Table 4. Seasonal differences in contamination (excluding study 8)

Months	Average temperature (Climatic Research Unit (CRU), n.d.)		Average precipitation (Climatic Research Unit (CRU), n.d.)	PoD low risk		PoC low risk		Contamination from local hygiene pathways	Contribution of local hygiene pathways to overall contamination		Contamination increased by local hygiene pathways	
	Min	Max		PoD=0		PoC=0		PoD-PoC	PoC >0, PoD = 0		PoC= > 0, PoD < PoC	
	°C	°C	mm	n	%	n	%	%	n	%	n	%

DJF	13.4	26.6	30	1082	90.8	995	83.5	7.3	87	44.4	129	65.8
MAM	22.1	32.8	388	1859	94.9	1767	90.2	4.7	92	48.2	117	61.3
JJA	25.7	31.7	1226	1356	94.9	1233	86.3	8.6	123	62.8	148	75.5
SON	22.4	30.9	515	378	95.0	350	87.9	7.0	28	58.3	38	79.2
Total				4675	94.0	4345	87.3	6.6	330	52.3	432	68.5

3.2 Contribution of household hygiene contamination pathways at the PoU

Data on PoC and PoU *E. coli* contamination was analysed based on MICS data from 38 datasets (Table 5) to identify the contribution of household hygiene pathways, specifically hygiene of the user when filling a drinking water vessel and any contamination of the drinking water vessel. Across these countries and regions, samples from the PoC ranged from 89.6% without *E. coli* detected (i.e. low risk) in Turks and Caicos Islands to only 5.1% in Tuvalu. At the PoU, samples without *E. coli* detected ranged from 80.0% in Kosovo to only 0.7% in Chad. To focus on the role of household hygiene pathways for contaminating water at the PoU, only households that collected the PoU water sample directly from the PoC were considered in further analysis. In the 38 datasets that were considered, the level of data that could therefore be included varied (Table 5). More than 70% of PoU samples were collected directly from the PoC in Georgia, Iraq, Kosovo, Samoa, Suriname and Tunisia. Less than a quarter of samples were collected directly from the source in 17 countries, with four countries excluded from the analysis due to insufficient data availability.

The proportion of households where no contamination was identified from household hygiene pathways (no detectable *E. coli* at the PoC that were also low risk for PoU samples collected from cups filled directly at the PoC) ranged from 29.5% in Nepal to 91.7% in Turks and Caicos Islands (Table 5). Figure 3 presents the variable impact of contamination from household hygiene pathways to drinking water at the PoU. In Nepal, 70.5% of households contaminated their drinking water through poor hygiene in collection of the water or poor hygiene of the cup. While there are correlations between the rate of contamination at PoC and PoU with national statistics on sanitation (open defecation and safely managed sanitation) and hygiene (basic access and no facility), there is no relationship between these sanitation and hygiene statistics and the number of households contaminating their water at the PoU (see supplementary material Table S2). PoU water quality was weakly correlated with the prevalence of collecting PoU samples from the source, supporting the argument that water sources on premises provide better water quality for users.

Table 5. Contamination of water quality at PoC & PoU based on MICS6 data

Country/area	Proportion of samples low risk (without *E. coli* detected)				PoU samples from cups filled directly at the PoC		No contamination identified from household hygiene pathways	
	PoC=0		PoU=0					
	n	%	n	%	n	%	n	%
Algeria	2641	84.3	2210	70.4	1518	48.4	1108	84
Bangladesh	3750	61.8	1101	18.1	345	5.7	75	33.9

Central African Republic*	323	30.8	131	12.5	64	6.1	8	61.5
Chad*	275	13.1	15	0.7	180	8.3	0	0
Democratic Republic of the Congo	1103	40.3	842	30.7	457	16.7	107	75.9
Dominican Republic	1126	44	581	22.7	507	19.8	137	55.7
Fiji	622	57.1	528	48.5	1035	95	483	79.7
Gambia	1021	57.9	489	27.7	64	3.6	35	71.4
Georgia	1590	65.5	1376	56.6	1804	74.3	1137	88.3
Ghana	1695	53.6	767	24.3	1011	32	527	72.9
Guinea-Bissau	657	36.8	333	18.7	102	5.7	18	47.4
Guyana	586	40.5	403	27.9	919	63.6	230	56.1
Honduras	2121	52.7	1672	41.5	2432	60.4	1196	73
Iraq	3866	57.8	3327	49.8	5365	80.2	2279	76.4
Kiribati	68	11.5	48	8.1	320	54.3	10	30.3
Kosovo	899	82.5	872	80	999	91.7	754	91.4
Laos	570	17.3	506	15.4	839	25.5	55	39.9
Lesotho	918	68.6	638	47.6	44	3.3	30	83.3
Madagascar	502	15.4	313	9.6	148	4.5	51	89.5
Malawi	1209	38.7	261	8.4	186	6	44	55.7
Mongolia	2106	81.1	2073	79.8	344	13.2	266	91.1
Nepal	524	21.4	310	12.7	585	24	64	29.5
Pakistan Baluchistan	379	14	201	704	734	27.2	40	52.6
Pakistan Khyber Pakhtunkhwa	537	16.3	176	5.3	508	15.4	26	39.4
Pakistan Punjab	4455	64.9	2859	41.7	3627	52.9	1446	61.4
Pakistan Sindh	979	41.2	405	17.1	597	25.1	114	40.1
Samoa	458	69.3	395	59.8	553	83.7	334	85
Sao Tome and Principe*	236	69	200	58.5	21	6.1	12	85.7
Sierra Leone	179	10.2	58	3.3	290	16.6	25	59.5

State of Palestine	1483	81.5	1265	69.5	1168	64.2	781	83.5
Suriname	888	54.8	563	34.8	1301	80.4	381	49.8
Togo	376	34.6	126	11.6	118	10.8	53	80.3
Tonga	153	28.2	115	21.2	361	66.5	54	57.4
Tunisia	2127	79.8	1916	71.9	2454	92.1	1700	85.3
Turks & Caicos Islands	232	89.6	205	79.2	137	52.9	111	91.7
Tuvalu*	8	5.1	17	10.8	78	49.7	0	0
Viet Nam	1670	50.5	1807	54.6	884	26.7	355	71.9
Zimbabwe	926	45.3	428	20.9	230	11.3	135	77.6
* Countries had fewer than 30 samples at PoU collected direct from a clean source. These results are not included in further analysis.								

[insert here] Figure 3. Contribution of household hygiene contamination pathways: proportion of households with water quality contaminated between the PoC and PoU

3.3 Contribution of contamination pathways in unchlorinated piped water systems

The results above for PoD focused primarily on smaller infrastructure such as tubewells where results almost uniformly show reductions in contamination with *E. coli* after decontamination (PoC > PoD). However, in unchlorinated piped systems in hospitals, significant increases in contamination were observed after decontaminating taps, either by flaming or wiping with ethanol. In these systems, water quality from the source (or nearby tubewell) was typically low risk for *E. coli* at the point of delivery (81% for deep tubewell sources n=16; 77% for handpumps, n=9). Contamination at the taps used for healthcare was common with only 54% of taps free from *E. coli* contamination at the PoD and 50% at PoC. *Pseudomonas aeruginosa* were detected in water from 96% (n=54/56) healthcare use taps at PoC. Contamination was visible when wiping inside taps. Contamination in taps used for drinking water was slightly less common as these included taps on filtration systems. In these settings, contamination often increased after cleaning and flushing the taps (Figure 4), particularly for *Pseudomonas aeruginosa* which are associated with biofilm formation.

[insert here] Figure 4. Change in concentration of organism in samples before and after cleaning and flushing taps for healthcare use in unchlorinated, piped hospital water systems (top), and in all taps and spouts sampled in hospitals (bottom)

4 Discussion

The results demonstrate the influence of different contamination pathways on water quality sample results. Understanding the relative contributions of different contamination pathways influences the interpretation of different sample types and has important implications for drinking water safety management. Here, we focus on operational

understanding of water systems, however, the interpretation also relates to public health and regulator understandings of health risks and appropriate interventions.

PoD samples characterise the water as delivered. It should not be interpreted as a 'source' sample because it includes the impact of the engineered system (Bartram and Hunter, 2015), although it is often difficult to sample source water quality without using infrastructure, especially in groundwater systems. For drinking water managers, PoD results can be clearly interpreted as demonstrating the water quality that the water system (source, treatment, distribution) is capable of providing. As such PoD samples can help identify where interventions are required in the source, treatment, or distribution system, including at the handpump or spring. PoD samples will not further differentiate between source, treatment or distribution contamination pathways, so more distributed sampling is recommended to identify vulnerabilities in complex systems such as piped networks (World Health Organization, 2024).

PoC samples identify the user's risk of exposure to faecal contamination via drinking water associated with where they collect their water from. Our results demonstrate that local hygiene at the tap is a significant contamination pathway. This has implications for human health. PoC samples are assumed to represent the quality of water that is available or is collected (e.g. Rufener et al., 2010), however, there are limitations to this assumption. Contamination on the tap will likely have high temporal variability due to both the nature of the contamination source from people and animals accessing the tap, and the potential for rapid decreases with die-off of bacteria due to heat and drying of the tap or washing of the tap during use. The authors' fieldwork observations of user behaviour at the tap have indicated that people (and animals) regularly touch water supply outlets, with different collection vessels, priming methods, cleaning practices and mobility impacting the potential for transference of contamination onto the outlet and therefore into the water collected. For drinking water managers, results from PoC samples cannot differentiate between the need to clean the tap, and the need for interventions to the water system.

PoU samples identify contamination in drinking water as consumed by a child, which includes contamination originating from the water system and local hygiene, from household water management, and from household hygiene including contamination on the cup and on hand hygiene. Our analysis demonstrates that the contribution from household hygiene varies, with household hygiene contributing contamination in up to 70% of samples. For drinking water managers, understanding faecal contamination at the PoU does not aid operational decisions due to the multiple contamination pathways involved, however, in-household sampling approaches remain important to understand chlorine residuals and contamination from the infrastructure, such as lead.

Consistent reporting of sampling methods is essential to enable appropriate interpretation of results. In the literature, PoU sampling methods are generally clearly communicated. However, there is often poor description of sample collection and processing (Sikder et al., 2021), with sampling methodologies to differentiate PoD and PoC methods often unclear. These two methods are not differentiated further in systematic reviews of drinking water quality (e.g. Bain et al., 2014; Kostyla et al., 2015; Sikder et al., 2021; Wright et al., 2004), with each using a combination of papers that use either PoD or PoC or do not report the

sampling method clearly. This poor reporting can be found in current papers, for example, Murei et al. (2024) report on tap water quality samples and draw conclusions about relationships with sanitary inspections with no details of the sampling method. Our results (Figure 2) demonstrate that around 60% of sources that are contaminated with *E. coli* at the PoC are free from contamination at the PoD. This large difference makes it imperative for research and practice to explicitly communicate the sampling methods used to enable useful interpretation and comparison of results. Sampling guidance should be updated to clearly differentiate the types of samples, purposes and how to report them.

Cleaning the tap or spout is not just a sampling practice but should be a standard part of hygiene education programmes. Contamination of communal water points from poor hygiene presents a risk of disease transmission, between households and also between vulnerable individuals at shared taps at schools and hospitals. However, there is a gap in the guidance and programme documents on cleaning at the point of water distribution. Literature addresses hygiene education, with a focus on the household, or infrastructure operation and maintenance, but cleaning the tap is overlooked. For example, WaterAid's Technology Notes (WaterAid, n.d.) provide guidance on hygiene education and water source development, but do not mention cleaning handpumps. In guidance for small-community supplies (WHO, 1985) it is advised that "improved water sources should be used hygienically" but there is no reference to cleaning. Early drafts of current sanitary inspection guidance (World Health Organization, 2024) propagated that approach, with hygiene only focused on household practices. This was revised to include cleaning as a daily action for management of water systems based on discussion of the preliminary results presented in this paper.

Further guidance might consider recommending handwashing before water collection to reduce contamination of water points. Our results demonstrate that flaming can remove the contamination, but more research is needed to understand how frequently and what materials are needed to reduce the risk of disease spread. Anecdotal evidence from partners in Uganda suggests washing with just water can remove contamination, which is a practice that is appropriate for the individual user. Understanding the drivers of contamination, how quickly water collection outlets are contaminated after cleaning and the efficacy of different cleaning methods could inform the design of interventions that can improve water safety for users. Our analysis demonstrates that improving guidance on cleaning taps could substantially increase access to safer drinking water: In Bangladesh, we estimate an additional 35 million people would be considered to have access to safe drinking water (i.e. without *E. coli*) if the water collection outlets were cleaned, with a further 11 million people having access to cleaner drinking water with lower concentrations of *E. coli*. This would lower the proportion of people nationally without safe water from 40% to 19%.

Piped drinking water access has increased substantially in recent years, with an additional 1.8 billion people estimated to have access to piped water since 2000 (UNICEF/WHO JMP, 2023). However, the quality of that water is not always free from faecal contamination, with chlorination not routinely practiced, creating new challenges in managing water safety. the results from the unchlorinated piped systems in hospitals that are presented in this paper show that tap cleaning methods such as flaming (for metal fittings) and using ethanol wipes

(for plastic fittings) increased the concentrations of bacteria in drinking water samples, particularly for bacteria that are associated with biofilms, even after flushing (Figure 4). For *Pseudomonas aeruginosa* over 70% of samples demonstrated increased concentrations after cleaning the tap. It was not possible to do repeated samples to measure how concentrations vary over time post-cleaning, and further PoD sampling was stopped to avoid increasing risks for patients at the facilities. Similar results have been described by Wang et al. (2012) with an increase in bacteria liberated into the water from disrupted biofilm post-cleaning. Further research is needed to understand what sampling methods are appropriate in these settings. And with limited knowledge of the impact of decontamination on the scale and duration of release of such pathogens such as *Pseudomonas* and *Legionella*, decontamination should be attempted with caution in distribution systems serving vulnerable populations e.g. elderly, the immune-compromised patients in hospitals.

5 Conclusion

We demonstrated the substantial contamination of drinking water from local hygiene conditions at the point of collection. Local hygiene sources, such as from people and animals touching the tap, contributed to contamination of drinking water in 76% of water samples where *E. coli* was detected, including being the sole source of contamination in an average of 60% of samples. This impact of hygiene is also notable in samples from within the household, at the PoU, where hand- and household-hygiene is the primary source of contamination in some contexts, but with high variation by context. These pathways of contamination related to hygiene have public health implications for drinking water users, with a variety of interventions to address them. Recognising the contributions of these pathways, and embedding this in sampling programmes, can improve understanding of the contextual and seasonal drivers of contamination and support advances towards safer drinking water.

One key intervention that could improve drinking water quality is cleaning the tap or spout, especially before collecting drinking water. Behaviour interventions and hygiene have focused on water management in the household and overlooked behavioural aspects at the point of collection. It is important to note the limitations in the evidence, however, with the potential for unintended consequences where cleaning may impact on biofilms resulting in release of pathogens. There is increasing application of complex water supply infrastructure in piped water systems without chlorination or other treatments to minimise biofilm formation. More research is needed to refine guidance on the frequency and methods of cleaning, and implications for systems with substantial biofilm growth. However, this does not negate the immediate benefit to be gained from expanding communication and hygiene education to encourage cleaning of the tap.

A key part of recognising the contribution of hygiene pathways is to improve report. To date, there are inconsistencies in reporting drinking water sampling methods across the literature. We call on authors to ensure transparency by reporting if they use decontamination techniques as part of water sampling methods and urge caution in drawing comparisons across studies using different or unclear methods.

Data statement

The MICS datasets analysed during the current study are available from the MICS website, https://mics.unicef.org/

The icddr,b datasets in Table 1, used and analysed during the current study, are available from the corresponding author on reasonable request.

Author Contribution Statement

K.C. and D.J. conceptualised the study. K.C., S.N. and L.O contributed to the study design and conceived the analysis. Z.M., L.O. and K.C. collected and curated the data. K.C. performed the analyses. K.C. and S.N. undertook visualisation of the published work. K.C. and D.J. supervised the study. K.C. wrote the initial draft of the manuscript. K.C., S.N., L.O, Z.M. and D.J. contributed to reviewing and editing the manuscript.

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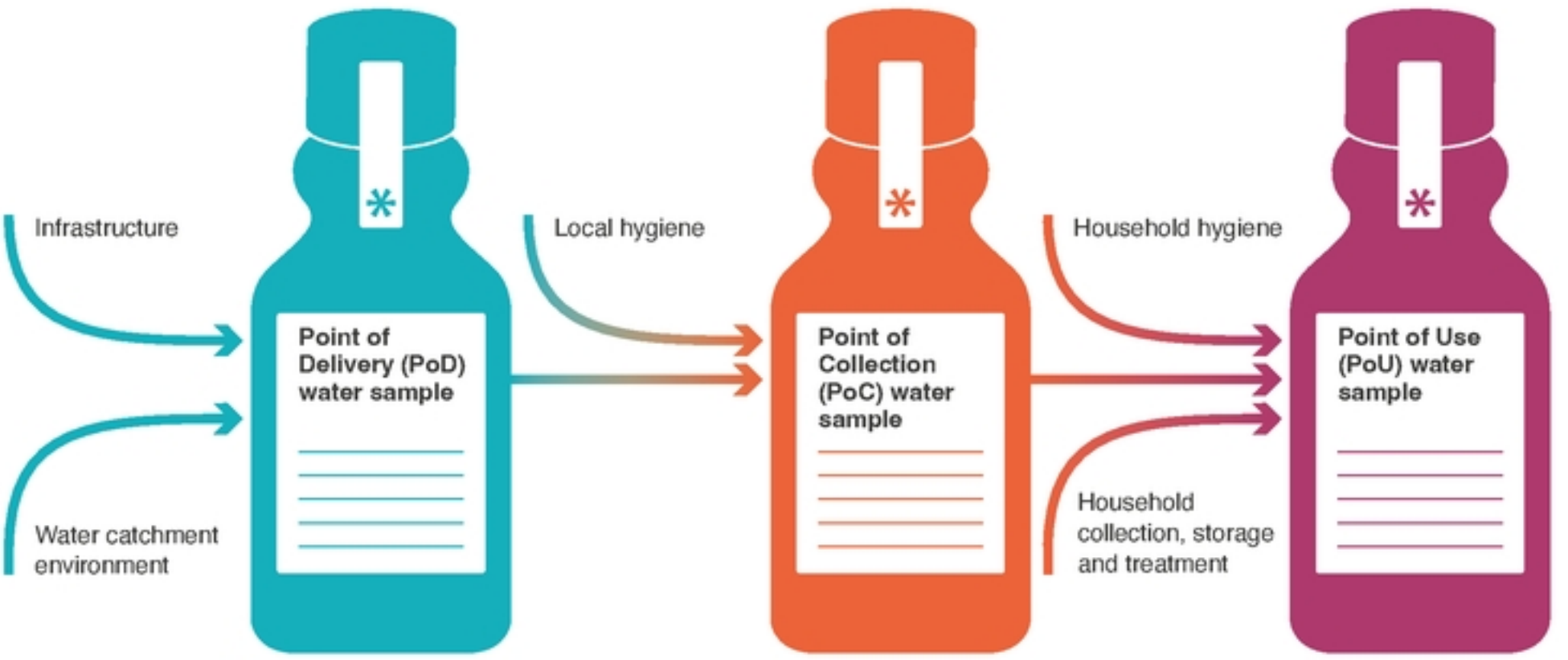

Figure 1

Figure 2

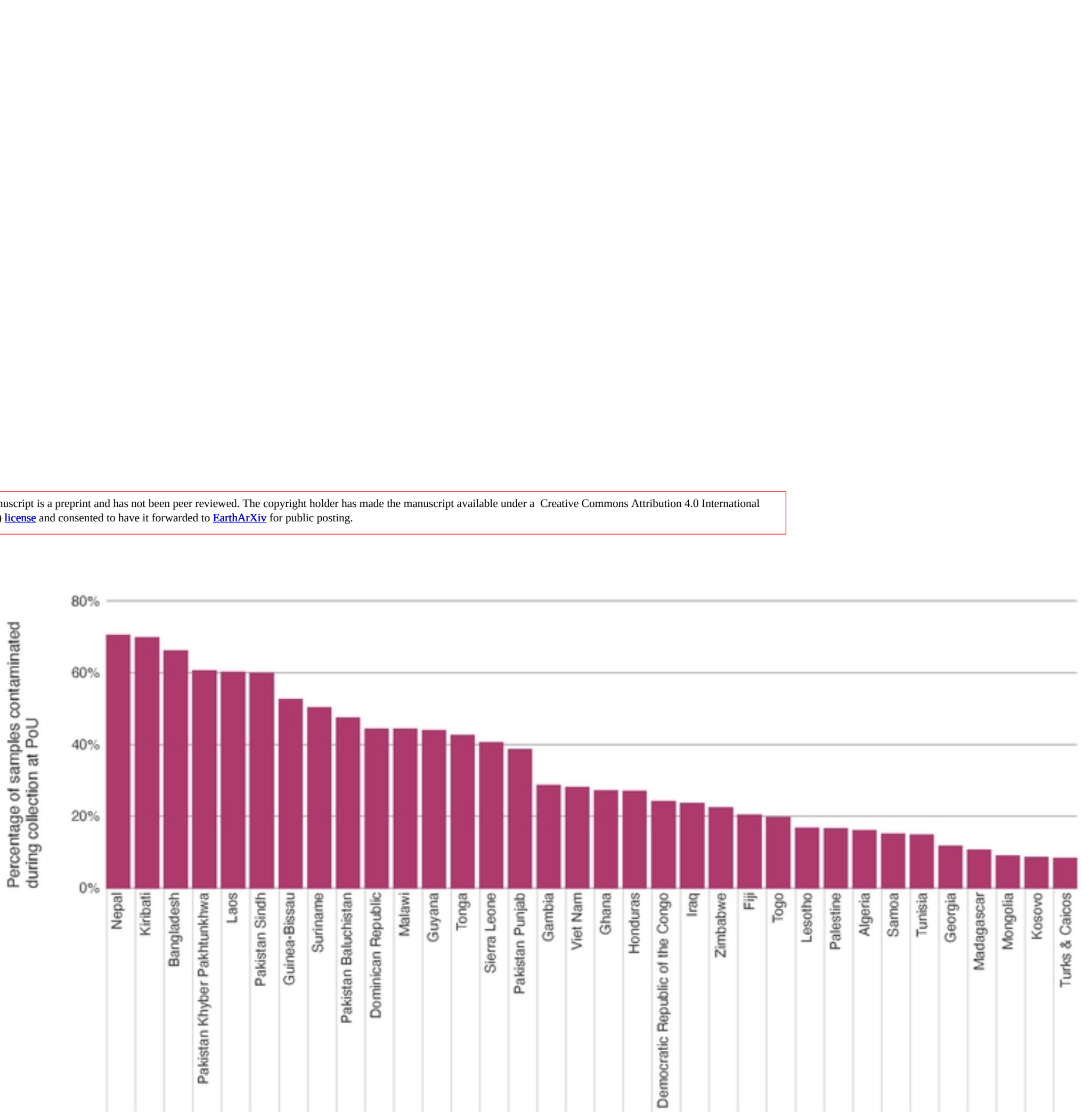

Figure 3

Figure 4