Drinking Water Safety: Role of Hand Hygiene, Sanitation Facility, and Water System in Semi-Urban Areas of India

Arti Kundu,¹ Woutrina A. Smith,^{1*} Danielle Harvey,² and Stefan Wuertz^{3,4,5}

¹School of Veterinary Medicine, One Health Institute, University of California, Davis, California; ²Department of Public Health Sciences, School of Medicine, University of California, Davis, California; ³Department of Civil and Environmental Engineering, University of California, Davis, California; ⁴Singapore Centre for Environmental Life Sciences Engineering, Nanyang Technological University, Singapore, Singapore; ⁵School of Civil and Environmental Engineering, Nanyang Technological University, Singapore, Singapore

Abstract. Poor drinking water quality is one of the main causes of acute diarrheal disease in developing countries. The study investigated the relationship between fecal contamination of hands, stored drinking water, and source waters in India. We further evaluated the environmental and behavioral factors associated with recontamination of water between collection and consumption. The bacterial contamination, that is, *Escherichia coli* ($\log_{10}$ most probable number per two hands), found on mothers' hands (mean = 1.11, standard deviation [SD] = 1.2, $N = 152$) was substantially higher than that on their children younger than 5 years (mean = 0.64, SD = 1.0, and $N = 152$). We found a low level of *E. coli* (< 1 per 100 mL) in the source water samples; however, *E. coli* contamination in stored drinking water was above the recommended guidelines of the World Health Organization. The study also found that *E. coli* on hands was significantly associated with *E. coli* in the stored drinking water ($P < 0.001$). Moreover, *E. coli* was positively associated with gastrointestinal symptoms (odds ratio 1.42, $P < 0.05$). In the households with elevated levels (> 100 *E. coli*/100 mL) of fecal contamination, we found that 43.5% had unimproved sanitation facilities, poor water handling practices, and higher diarrheal incidences. The water quality deterioration from the source to the point of consumption is significant. This necessitates effective interventions in collection, transport, storage, and extraction practices when hand–water contact is likely to occur. These findings support the role of hands in the contamination of stored drinking water and suggest that clean source water does not guarantee safe water at the point of consumption.

INTRODUCTION

Diarrheal disease, the third leading cause of childhood mortality in India,¹ is responsible for 13% of all deaths/year in children younger than 5 years old in India. An estimated 1.5 million deaths of children younger than 5 years old occurred in India in 2012.^{1,2} Even though “improved” water sources provide better water quality as compared with “unimproved” water sources, more than a quarter of samples from improved sources were fecally contaminated in 38% ($N = 191$) of the studies conducted in low- and middle-income countries (LMICs).³ In India, although half of all urban households have access to piped drinking water, more than 20% of urban households still obtain their water from a source located 100 m away from their households.⁴ Recent studies have shown an increased level of fecal contamination of source water and household stored drinking water leading to waterborne diseases in LMICs.^{4–6} Similarly, poor sanitation is linked to gastrointestinal diseases that can lead to stunting, malnourishment, and impaired cognitive development in children,⁷ and the World Health Organization (WHO)/United Nations International Children's Emergency Fund (UNICEF) Joint Monitoring Program estimated for 2012 that 60% of the world's open defecation occurs in India.⁷

Stored drinking water quality can be compromised in many ways depending on source types (e.g., proximity to latrines, access to animals, open defecation practices, and runoff), distribution of the water supply system (e.g., treatment processes, distribution, and handling), and water quality at the point of use by consumers (e.g., open versus wide-mouth storage containers, disinfection via boiling, chlorination, or

solar radiation). Recontamination during the storage of drinking water in the households, particularly due to poor sanitation and hygiene practices, has been linked to an increased risk of drinking water deterioration as compared with the transport of water from the source to storage in the households.^{8–11} Hands and water handling practices play an important role in drinking water contamination.^{12–14} Previously, researchers have investigated the relationships between fecal indicator bacteria (FIB) levels on hands and associated environmental and behavioral factors in source and stored drinking water.^{8,15,16} There is strong evidence that high levels of microbial contamination found on the hands of people are related to the poor quality of stored drinking water.^{14,17,18} Similarly, poor water handling practices such as the use of wide-mouth containers or inappropriate extraction practices can deteriorate the stored water quality.^{10,19,20} There is limited information available for evaluating drinking water safety related to environmental and behavioral factors (e.g., water handling practices, hand hygiene, sanitation practices, and water quality) in an improved non-networked water and sanitation system in a threshold economy like India.

Therefore, it is imperative to evaluate different water sources, hand hygiene, water storage practices, and sanitation facilities in the peri-urban households for effective intervention implementation. Importantly, it will constitute a starting point in characterizing the study population in this research where relatively little is known about local perceptions and cultural barriers for using sanitation and hygiene practices. Given the knowledge gap related to the role of hands in fecal–oral disease transmission routes in semi-urban areas of India, the research objectives were aimed to 1) quantify the fecal contamination on the hands of the respondents by measuring *Escherichia coli* levels, 2) compare the difference in the microbiological drinking water quality between the point of consumption and source in improved non-networked water systems, and 3) characterize the households with high levels

* Address correspondence to Woutrina A. Smith, School of Veterinary Medicine, One Health Institute, 1089 Veterinary Medicine Dr., University of California, Davis, CA 95616. E-mail: wasmith@ucdavis.edu

of fecal contamination by surveying the households for socioeconomic and demographic characteristics, water handling management, hygiene, and sanitation practices. We hypothesized that characterizing water sources, sanitation practices, and hygiene behavior can ameliorate the stored drinking water quality at the point of use in households and, thereby, improve associated health benefits.

MATERIALS AND METHODS

Sampling site and approach. The study area, National Capital Territory (NCT), Delhi, is the second largest populated city in the world with 25 million inhabitants in 2014, a number that is projected to rise swiftly to 36 million in 2030 (United Nations Department of Economics and Social Affairs, 2014). The capital city gets its drinking water from various sources such as the Yamuna River, Bhakra Dam storage, Upper Ganga Canal, and groundwater, but the supplies are inadequate to satisfy the demands of the increasing population. The piped networked water supply system has poor water quality and quantity because of intermittent water supply and low pressures in the piped water systems.²¹

Before the household recruitments, the study area was evaluated by trained staff in collaboration with a local non-governmental organization to recruit the households with the study guidelines and eligibility criteria (i.e., children aged < 5 years). An additional consideration was the willingness of the households to participate in the study. Based on the survey, the 152 households that are at the interface of the rural and urban transition zone (peri-urban) of the northwestern part of NCT (Puth Khurd, Shahbad Daulat Pur, and Sector-27 Rohini) were identified such that each household had at least one child younger than 5 years old. Informed consent for the collection of hand rinse samples and stored drinking water samples was obtained during the household visit from the primary respondent (usually primary caretaker for the children and the person responsible for the household water management practices). Data collection started in the participating households between the months of September 2013 and November 2013. The study protocol was approved by the Institutional Review Board of the University of California, Davis (UCD), and the Institute Ethics Committee of All India Institute of Medical Sciences (AIIMS IRB no. IEC/NP-528/2013RP-04/2013) in India in 2013 (UCD IRB no. 493207-1).

Household interviews and sample collection. A trained field worker conducted the household interviews in the local language, that is, Hindi under the supervision of an experienced health educator. The female head of the family was invited to participate in the study. The interviews in the form of a 30-minute questionnaire were carried out at each household and evaluated the following characteristics of each household: demographic, socioeconomic, different water types, water handling practices, water quality, sanitation, and hygiene. Observations regarding the presence of flies in the households, the availability of handwashing facilities (bucket, tumbler, washbasins, etc.) in the toilets, the presence of soap in the toilet facilities, and the presence of animals in the households were noted down. The participants were requested to collect the drinking water sample as they usually do and pour it into a 69-oz. sterile bag (VWR, Radnor, PA).

In addition, information about the last time that respondents washed their hands was recorded. Before handwashing

procedures, the enumerator asked the mothers to show both hands and collected information related to visible dirt inside the fingernails, on their palms, and on finger pads of the hands. After that, the mothers and their children younger than 5 years of age were asked to place their hands one at a time into a sterile 69-oz. sample bag (VWR) containing 350 mL of sterile deionized water. Each hand was massaged for 15 seconds in the sample collection bag, after which a surveyor massaged the hands for an additional 15 seconds through the sample collection bag. In the end, the respondents were provided a clean paper towel to dry their hands. Researchers previously published the handwashing procedure.¹⁸ In addition, field blanks were run every week to check for microbial contamination of collected hand rinse samples where the field blank sample was exposed to the same field conditions as the sample processed in the field and later on assessed for microbial contamination in the laboratory as a quality assurance/quality control measure.

Health outcomes regarding the presence of highly credible gastrointestinal symptoms (HCGS) and significant respiratory symptoms (SRS) were reported for the last 2 days before data collection. The respondents were asked to report anybody in the households with given symptoms for HCGS: two or more of the following symptoms or one of the following symptoms with fever: stomach pain, three or more bowel movements in 24 hours, watery or loose stools, blood in the stool, or vomiting. The SRS were two or more of the following symptoms or one of the symptoms of a fever: coughing, congestion or a runny nose, or difficulty in breathing.¹⁴ Respondents were asked if they sought medical treatment for the illnesses.

Water testing. The drinking water and source water samples were collected into a sterile 69-oz. sample bag containing sodium thiosulfate to neutralize the effect of any chlorine in the water. All samples were stored on ice in a cooler and transported to the laboratory for analysis of *E. coli* within 6 hours after collection. To assess the physical characteristics of the collected water and hand rinse samples, turbidity was measured using a 2100-Q portable turbidimeter (HACH Company, Loveland, CO) and protocols compliant with USEPA method 180.1. The bacterial indicator (*E. coli*) was estimated using the most probable number (MPN)-based IDEXX Colilert® Quanti-Tray® 2000 defined substrate method (IDEXX, Westbrook, ME); concentrations were expressed as MPN/100 mL. The assay was performed as per the manufacturer's instructions. Briefly, on receipt of samples in the laboratory, each sample was massaged for 60 seconds in the sample collection bag; then 100 mL of each sample was removed in a sterile bag (HACH Company) and reagents were added. The samples were gently mixed to dissolve the media and the contents were transferred into sterile Quanti-Tray 2000 trays. Then, each Quanti-Tray was heat-sealed and incubated at $35 \pm 0.5^\circ\text{C}$ for 24 hours. Blanks were run daily in the laboratory using autoclaved bottled drinking water and always tested negative as expected.

Statistical methods. Data for *E. coli* were $\log_{10}$ -transformed for statistical analysis. Statistical tests for comparing results among groups used paired *t* tests, analysis of variance (ANOVA), Wilcoxon rank sum test, and Pearson's correlation coefficient. Multivariable linear and logistic regression models were built in a forward stepping manner using SPSS software (SPSS Inc., Chicago, IL) to investigate the association between stored drinking water quality, *E. coli* levels on respondents' hands, and health outcomes. The multiple stepwise regression method

was selected because it is a semi-automated process of building a model by successively adding or removing predictor variables that have the highest simple correlation with the outcome. It has several advantages as it is less prone to overfitting the data and the user can know more about explanatory variables by observing the order in which variables are added or deleted in the model building. P values < 0.05 were considered statistically significant. For the inclusion of concentration data above and below measurable limits, 1 MPN/100 mL was assigned to values below the lower detection limit of < 1 MPN/100 mL and 2,419.6 MPN/100 mL was assigned for values that were above the detection limit of 2,419.6 MPN/100 mL, respectively. The FIB concentration estimates were analyzed using SPSS software (SPSS Inc.). A paired t test was used to compare the difference between *E. coli* concentrations on mothers and children younger than 5 years old; the paired t test was used to see the difference in mean *E. coli* levels in source water and stored drinking water. A one-way ANOVA was conducted to see the difference in *E. coli* levels in more than two groups, for example, *E. coli* levels on very dirty hands, somewhat dirty hands, or clean hands; the Wilcoxon rank sum test served as a nonparametric test to examine the differences between variables; and Pearson's (r_p) correlation coefficient was used to see the linear correlation between two continuous variables, for example, *E. coli* in stored drinking water and *E. coli* on respondents' hands.

RESULTS

Household baseline characteristics and data collection.

The potential role of hands in the contamination of stored drinking water along the point of supply/source to the point of consumption exposure pathway was evaluated (Figure 1). To

accomplish the study objectives, 152 households ($N = 500$ samples) were surveyed in three different peri-urban areas, and in more than half (62%) of the households, the average family living in the house consisted of five or more individuals, with 25% of these households reporting at least one infant (< 1 year old) in the house. The average monthly income per household was 12,273 Indian national rupees ($\sim \$204$). Of the households visited, 14% of the households owned domestic animals ($N = 21$), and most of these households with animals had livestock, that is, cattle in the houses (standard deviation [SD] = 3.2, range 1–12).

Regarding water consumption per household, the female caretaker of the family reported an average of 2 liters of drinking water consumption per person per day (SD = 1.13, range 0.53–8.5), which is an important input parameter in future health risk assessment studies. The three study regions featured a mixture of networked and non-networked drinking water sources. The water supply in these areas came from two major sources: hand pumps that provided a 24-hour water supply and municipal taps which supplied water for 1–2 hours at variable intervals of time ranging from alternate days to sometimes once in 3–4 days. At the time of the interview, only 45% of households reported using an improved toilet facility, that is, a septic tank or a pit with improved lining.²² By contrast, 51% of respondents reported using a shared sanitation facility and 4% of people reported openly defecating in the environment, considered “unimproved sanitation” according to the WHO/UNICEF Joint Monitoring Program definition.²²

Presence of *E. coli* on hands. The microbial fecal contamination on the hands of respondents was measured using *E. coli*, with the mean $\log_{10}$ concentration of *E. coli* found on the hands of respondents as 1.13 (SD = 1.1, $N = 152$) $\log_{10}$ MPN per two hands (Table 1). The microbial fecal contamination,

FIGURE 1. Conceptual framework of the study. The quality of stored drinking water is related to water sources, sanitation, hand hygiene, and water management practices (collection, transport, storage, and extraction) in the given households. The *Escherichia coli* contamination in stored drinking water is significantly correlated with *E. coli* contamination present on respondents' hands. This figure appears in color at www.ajtmh.org.

TABLE 1
Bacterial levels in samples collected from hand rinse samples, stored drinking water, and drinking water sources

Sample type	N*	<i>Escherichia coli</i> prevalence (%)	Mean† (SD)‡	Median (log ₁₀)	Range (log ₁₀) (min, max)
Mother	152	68	1.10 (1.20)	0.70	(0, 3.38)
Child aged younger than 5 years	152	53	0.60 (1.00)	0.0	(0, 3.38)
Household stored water from					
Municipal tap water	56	57	0.63 (0.95)	0.0	(0, 3.38)
Filtered water	3	67	2.08 (1.82)	2.86	(0, 3.38)
Bottled water	2	100	2.73 (0.50)	2.73	(2.38, 3.08)
Hand pump	76	69	0.87 (0.96)	0.79	(0, 3.38)
Vendor	4	75	1.15 (1.07)	1.02	(0, 2.56)
Tanker	5	80	0.76 (0.72)	0.99	(0, 1.54)
Combination of either sources	6	50	1.15 (1.28)	1.00	(0, 2.69)
Source water from					
Municipal tap water	7	0	0	0	< dl§
Bottled water	5	0	0	0	< dl
Hand pump	7	0	0	0	< dl
Vendor	3	33	0.15 (0.38)	–	(0, 0.61)
Total samples collected for analysis	478	–	–	–	–

MPN = most probable number; SD = standard deviation.

* N is the total number of samples.

† Mean of the log₁₀-transformed concentrations.

‡ SD is the SD of the log₁₀-transformed.

§ < dl is the lowest detection limit, that is, 0 log₁₀ MPN/100 mL and > dl is the highest detection limit, that is, 3.38 log₁₀ MPN/100 mL.

in log₁₀ MPN per two hands, found on the hands of mothers (mean = 1.11, SD = 1.2, and N = 152) was substantially higher than that of their children aged younger than 5 years (mean = 0.64, SD = 1.0, and N = 152). In addition, there was a significant difference in *E. coli* levels found on the hands of mothers and children aged younger than 5 years ($P < 0.01$) (Figure 2). Moreover, when we compared the mean *E. coli* concentrations on the hands of mothers and children with mean *E. coli* levels of household stored drinking water, we found a significant correlation between the log₁₀ mean *E. coli* on the hands of participants and the log₁₀ mean *E. coli* in household stored drinking water ($r_p = 0.29$, $P < 0.001$, and $N = 152$).

Exposure pathways of contamination in the households.

Overall, households reported collecting stored drinking water from six types of sources: the majority of respondents (51%) had reported public hand pumps as the primary drinking water source, whereas only 37% of households had reported municipal tap water as their primary drinking water source. Other drinking water sources reported by households included tankers (3%) and vendors (2%). Only 2% of households had used “filter water” and 1% of respondents had reported “bottled water” as their primary drinking water source. Also, 4% of the households were using more than one water source (Table 1). According to the WHO/UNICEF Joint Monitoring Program²² definitions of “improved” drinking water sources, 88 (88%) households in the present study collected their water from “improved” drinking water sources.

There was a significant difference in the quality of stored drinking water collected from different sources, that is, municipal piped water, hand pump, vendor, and bottled water. At the time of site visit, 81% of respondents reported that they never treat their drinking water, whereas 11% of households reported sometimes treating their drinking water. Also, 8% of respondents reported always treating stored drinking water before drinking. There were no significant differences in *E. coli* concentrations (log₁₀) between samples that were reported to be “always” ($N = 12$, 7.9%, mean = 0.81, and SD = 1.2), “sometimes” ($N = 17$, 11.2%, mean = 1.05, and SD = 1.1), or “never” ($N = 123$, 80.9%, mean = 0.82, and SD = 0.9) treated before consumption of drinking water (ANOVA, $P > 0.05$).

In the study, 57% of respondents reported storing drinking water in closed containers, 30% of respondents reported storing drinking water in open containers, and 13% reported using a mixture of close and open storing system. When comparing the household characteristics of stored drinking water with the quality of stored water, there was no statistical difference in the quality of drinking water stored in an open container ($N = 45$, 29.6%, mean = 0.76, and SD = 1.1), closed container ($N = 86$, 56.6%, mean = 0.92, and SD = 1.1), or a mixture of both ($N = 21$, 13.8%, mean = 0.52, and SD = 0.6) (ANOVA, $P > 0.05$). A majority of respondents reported the use of earthen pots as the kind of container used for storing drinking water (47%, $N = 72$) under the “others” category, whereas 26% ($N = 39$) reported plastic buckets for storing drinking water, 6% ($N = 9$) used metallic drums, 4% ($N = 6$) used jerry cans, and 47% ($N = 21$) of households reported using more than one storage container. The kind of container used for storing water was not associated with *E. coli* levels in the stored waters (ANOVA, $P > 0.05$).

In this study, the turbidity of household drinking water samples ranged from 0 to 19 nephelometric turbidity unit (NTU), with a mean of 1.96 NTU (SD = 2.45). The levels of turbidity in source drinking water samples were higher than those of household drinking water and ranged from 1.14 to 8.04 NTU, with a mean of 3.51 NTU (SD = 2.63). In terms of microbial water quality, the log₁₀ mean concentration of *E. coli* in stored drinking water was 0.85 (SD = 1.0, $N = 152$) log₁₀ MPN/100 mL. By contrast, the log₁₀ mean concentration of *E. coli* in hand pump samples, municipal piped water, and bottled water was below the detection limit of 0 MPN/100 mL. The log₁₀ mean of *E. coli* in vended water sources was 0.31 log₁₀ MPN/100 mL ($N = 3$). The relationship of *E. coli* in stored household drinking water (log₁₀ MPN/100 mL) with household characteristics, water sources, hand contamination levels, and water handling practices was modeled using a regression model (Table 2) and showed that the mean bacterial levels found on the hands of respondents were the only significant variable associated with the *E. coli* levels found in the household stored drinking water.

Water, sanitation, and hand hygiene characteristics. The hand hygiene behaviors, that is, self-reported handwashing

FIGURE 2. Concentrations of *Escherichia coli* in three different samples collected from each household: 1) hand rinse samples of mothers (non-detects = 60), 2) hand rinse samples of children aged younger than 5 years (non-detects = 78), and 3) stored drinking water (non-detects = 59).

behavior, the presence of soap in the toilet facility, and visible dirt on the hands, were compared with the *E. coli* concentrations found on the respondent's hands, with no statistical difference observed between self-reported hand behavior and mean levels of *E. coli* on hands in households ($t = 0.356$, degrees of freedom [df] = 150, and $P > 0.05$). Participants had no significant difference in *E. coli* levels between those households with observed soap and those without soap ($t = 0.692$, df = 145, $P > 0.05$). At the time of site visit, it was found that 79% of households had no soap present in the toilet facility. *Musca domestica*, that is, common houseflies that are carriers of enteric pathogens,²³ were commonly present in the households (83%).

Respondents were asked how many times they wash their hands with soap on a given day: 18% of respondents reported that they "always" use soap to wash their hands, 80% reported "occasionally," and 2% reported "rarely." Of the households visited, 88% of mothers had dirt observed underneath their fingernails. Visual inspection of hands and

finger pads showed that 26% of respondents' hands were visually "very dirty," 59% of them were "somewhat dirty," and 15% met the "clean" criteria. Also, no significant difference was found between mean levels of *E. coli* on hands in households and observed dirt in palms, finger pads, and underneath the fingernails.

Dirt underneath the fingernails was classified into two categories as "very dirty" and "clean," whereas observed dirt on hands was classified into three categories as "very dirty," "somewhat dirty," and "clean." Surprisingly, mean *E. coli* levels were comparatively higher in washes from "clean" hands (mean = 1.37, $N = 22$, and SD = 1.1) than "somewhat dirty" hands (mean = 1.05, $N = 91$, and SD = 1.1). Also, "very dirty" hands (mean = 1.09, $N = 39$, and SD = 1.1) had slightly higher *E. coli* levels than "somewhat dirty" hands (log₁₀ MPN/two hands) (Figure 3). Similarly, hands with no visible dirt underneath their fingernails (mean = 1.89, $N = 18$, and SD = 1.3) had higher *E. coli* levels than fingernails with observed dirt

TABLE 2

Relationship of *Escherichia coli* in stored household drinking water (log₁₀ MPN/100 mL) with household characteristics using multiple linear regression model (*E. coli* drinking water model [$N = 152$]; Adj. $R^2 = 0.12$)

Variables	Beta	SE*	P value	95% CI
Constant	-1.941	1.489	0.20	(-4.88 to 1.00)
Container covered†	0.015	0.182	0.93	(-0.34 to 0.37)
Treated before drinking†	0.048	0.273	0.86	(-0.49 to 0.58)
Hand pump vs. tap water†	-0.315	0.169	0.07	(-0.64 to 0.02)
Average monthly income‡	0.261	0.151	0.09	(-0.03 to 0.56)
Presence of flies†	0.194	0.252	0.44	(-0.31 to 0.69)
Presence of animals†	0.025	0.232	0.91	(-0.43 to 0.48)
<i>E. coli</i> on hands§	0.284	0.071	< 0.0001*	(0.14-0.43)

MPN = most probable number.

* Significant $P < 0.05$. Standard error (SE) of beta.

† Binary variables (0 and 1).

‡ Indian rupees per household.

§ Mean log₁₀ MPN/two hands.

Hand rinse samples collected from mothers with different levels of visible dirt on their hands ($N = 152$, Error bars = 95% CI)

FIGURE 3. Levels of *Escherichia coli* in mothers' hand rinse samples with visible dirt level from very dirty (mean = 1.09, $N = 39$, and standard deviation [SD] = 1.1) to somewhat dirty (mean = 1.05, $N = 91$, and SD = 1.1) and clean hands (mean = 1.37, $N = 22$, and SD = 1.1).

underneath them (mean = 1.0, $N = 134$, and SD = 1.1). Also, *E. coli* concentrations found in child hand rinse samples showed a significant positive association with turbidity using the Spearman rank correlation test, whereas no significant relationship was obtained between mothers' *E. coli* levels and turbidity.

Health-related outcomes (HCGS and SRS). In this study, 30% of the participants reported that they regularly got sick from drinking water, 48% reported sometimes getting sick from drinking water, and 22% of respondents mentioned never recalling getting sick from drinking water. The respondents stated the following reasons for getting sick from drinking water: poor quality, yellow in color, salty taste, dirt in the water, and low-pressure water. Moreover, 25% of respondents reported at least one household member with HCGS within the last 2 days. Also, SRS was even more prevalent in the community as compared with HCGS, that is, 39% of the households reported at least one household member with SRS in the previous 2 days. Furthermore, 66% of respondents reported seeking medical treatment regarding seeing a doctor for the self-reported health outcomes and 80% of respondents who received the medical treatment stated they used antibiotics for treating the illness. Binary logistic regression models were used to predict the presence of HCGS and SRS as a function of *E. coli* on the hands of household members, the presence of an animal in the household, average monthly income, single home versus multifamily home, and whether an infant was present in the household as shown in Table 3. Fecal indicator bacteria (*E. coli*) on the hands of respondents were significantly associated with HCGS. There was no significant association between the explanatory variables and SRS.

DISCUSSION

Hands play an important role in the contamination of stored household drinking water in the three semi-urban areas of the NCT, India. With low or no detectable *E. coli* in source waters, but high levels on hands and in stored drinking water, study findings suggest that interventions should target exposure routes of diarrheal pathogen transmission at the household level in a threshold country. The study found significantly higher loading of *E. coli* on the hands of mothers than the hands of children. Several activities such as preparing food, cleaning up a child's feces, visiting a toilet, bathing, etc., can significantly increase the fecal load on mothers' hands.¹⁷ Another study in low-income settings conducted by Pickering et al.¹⁴ demonstrated that hand contamination of the respondents was positively correlated with fecal contamination in the stored drinking water within the households. It is notable that visibly clean hands can still have higher levels of *E. coli* as compared with visibly dirty hands, and this may help explain why interventions should focus on good hygiene practices regardless of hand appearance.

The main results of the study relate to the general characteristics of the study population in terms of water storage and handling practices, sanitation facilities, and hygiene behaviors. Our study identified the households ($N = 23$) with elevated levels of *E. coli* (≥ 100 *E. coli*/100 mL) in the stored drinking water, with half of these households (43.5%) reporting episodes of HCGS and SRS. The primary health outcomes were chosen in terms of episodes of HCGS and SRS because of their use in prior trials and high sensitivity (any of several symptoms constitute an episode).^{14,24} There were two important characteristics of the households with high levels of

TABLE 3

Binary logistic regression models assessing the relationship between *Escherichia coli* on hands and in household stored drinking water and health outcomes

Highly credible gastrointestinal symptoms model ($N = 152$; Cox & Snell $R^2 = 0.04$)					Significant respiratory symptoms model ($N = 152$; Cox & Snell $R^2 = 0.03$)				
Variables	Beta	SE*	P	Odds ratio (95% CI)	Variables	Beta	SE	P	Odds ratio (95% CI)
<i>E. coli</i> on the hands of respondents†	0.37	0.17	0.02	1.45 (1.01–2.00)	<i>E. coli</i> on the hands of respondents†	0.02	0.15	0.89	1.02 (0.75–1.37)
Presence of animals‡	0.02	0.55	0.97	1.02 (0.35–3.02)	Presence of animals‡	0.82	0.49	0.09	2.26 (0.86–5.90)
Average monthly income§	–0.38	0.36	0.32	0.70 (0.35–1.40)	Average monthly income§	0.21	0.32	0.51	1.24 (0.66–2.31)
Single home‡	0.53	0.44	0.23	1.70 (0.71–4.06)	Single home‡	–0.01	0.39	0.99	0.99 (0.46–2.17)
Infant‡	–0.05	0.44	0.90	0.95 (0.39–2.26)	Infant‡	0.21	0.39	0.59	1.23 (0.57–2.64)
Constant	1.74	3.21	0.58	N/A	Constant	–2.56	2.88	0.37	N/A

MPN = most probable number; N/A = not applicable.

* Standard error (SE) of beta.

† Mean log₁₀ MPN/two hands.

‡ Binary variables (0 and 1).

§ In Indian rupees per household.

E. coli: first, only 26% of the households reported drinking water from improved water source such as municipal tap water. Instead of one main source, the locals were dependent on multiple types of water sources such as hand pumps or vendors serving in the area. Second, in terms of water management practices, 65% reported that they had never treated their water before drinking and 69% of these households reported either using earthen pots or wide-mouthed jars for drinking water storage containers as compared with narrow-mouthed containers because it is easier to scoop the water directly from the container than pouring from the container. In northern India, earthen pots are common to store drinking water especially during the summer season because 1) water evaporates quickly through the pores leading to cooling of stored water and 2) the earthen pots are economical. Overall, the household management practices show that unsafe handling can contaminate stored drinking water at the point of use. In terms of sanitation facilities and handwashing practices, 43.5% of the households were using “unimproved” sanitation. In terms of hygiene practices, only 13% reported “always” washing their hands and a majority of the households did not have soap present in the toilets. Future studies may consider investigating water handling practices and their relationship to sanitation practices, and hygiene behavior in Delhi, India.

Our study showed that stored drinking water management practices and hand hygiene behavior poorly correlated with *E. coli* levels on hands. The bacterial contamination regarding *E. coli* levels found on the hands of mothers and children was associated with *E. coli* found in stored drinking water. Visible dirt found on the hands of mothers did not significantly correlate with *E. coli* levels. There was a poor correlation between self-reported handwashing behavior and *E. coli* concentration on the respondent's hands. The lack of correlation between the handwashing behavior and *E. coli* levels can be due to the following reasons: the trained field worker did not directly observe the handwashing practices, instead she verbally asked the respondents about hand hygiene practices, for example, how many times they wash their hands or if they use soap for washing their hands. These answers may not truly represent the actual hygiene practices as it is personal information and may be a confounding factor in our analysis. Second, the lack of robust dataset may be responsible for the relative lack of statistical significance in the dataset. Navab-Daneshmand et al.¹² reported the small sample size limitation

(97 households) while evaluating *E. coli* correlations and risk factors in multiple matrices, including soil, hands, drinking water, and handwashing water in a highly densely populated area. By contrast, a strong correlation between visible dirt on hands and FIB levels was previously reported by Pickering et al.¹⁴ Those authors evaluated the correlations between visible dirt and observed handwashing behavior using structured observation.¹⁴ In conclusion, these results imply that self-reported handwashing behavior data are unreliable and, hence, future work should incorporate an unobtrusive direct observation of hand hygiene practices by a trained observer, still considered the gold standard for evaluating compliance by the WHO.

Presence of gastrointestinal and respiratory symptoms.

A notable finding of the study was that the occurrence of gastrointestinal symptoms (HCGS) was significantly associated with the levels of *E. coli* found on the hands of respondents. For every 1-log₁₀ increase in the mean levels of *E. coli* found on the hands of respondents, a household was 1.42 times (confidence interval: 1.01–2.00) more likely to report a household member with gastrointestinal symptoms. The different variables such as the presence of animals ($N = 21$) in the household, the presence of an infant (< 12 months old) ($N = 38$) in the household, average monthly income, and whether families lived in a joint/extended family house ($N = 97$) (versus a single family home [$N = 55$]) were not associated with the health outcomes. By contrast, *E. coli* levels found on the hands of respondents were not significantly associated with SRS.

The presence of HCGS reported in this study was higher than that previously reported for Delhi (8%) during the 2 weeks before the survey from December 2005 to April 2006 National Health Family Survey-3. Our study was conducted in the wet monsoon season (September–November), and it is possible that seasonality affected the outcome. Furthermore, data related to the total number of diarrheal cases in Puth Khurd village were collected from the main general hospital, and found that the diarrheal cases were highest in the month of October 2012 with 2,803 cases. The presence of respiratory symptoms reported by respondents was slightly higher than that of gastrointestinal symptoms in this study. Twenty-five percent of respondents had reported that at least one household member with the primary health outcomes were chosen in terms of episodes of HCGS within the last 2 days, whereas 39% of the households reported at least one household member with respiratory symptoms in the last

2 days. Lower presence rates for self-reported HCGS (3%) as compared with SRS (17%) was also reported in Tanzania, and the main difference between HCGS and SRS presence rates was due to the presence of symptoms in SRS (e.g., running nose, and coughing) visible to an enumerator, whereas HCGS symptoms were not visible.¹⁷

Does the provision of providing safe drinking water at source extend to safe drinking water at the point of use?

An important finding of the present study was that the household stored drinking water was significantly more contaminated than the water collected directly from the community sources. In the present study, we found that 65% of the households had bacterial contamination in the stored drinking water above the recommended standard guidelines of 0 *E. coli*/100 mL of sample per the WHO.²⁵ In contrast to the stored drinking water, we found *E. coli* levels of less than one per 100 mL in source water samples. On average, stored drinking water had 0.85 log₁₀ MPN more *E. coli* per 100 mL than the source from which it was collected. These findings are also supported by previous research studies where researchers reported that high-quality drinking water sources did not ensure clean water at the point of use.^{26–28} Harris et al.¹⁶ confirmed that stored drinking water was contaminated regarding FIBs immediately after filling the storage containers at the source and extraction utensils used at home post collection. However, drinking water quality has been impacted by various other factors such as storage types, the height of the collection utensils, covering of the container, materials of the containers, handling practices, etc. However, Trevett et al.⁹ examined different factors associated with stored drinking water quality in rural Honduras. The study suggested that only source water quality was the significant factor in determining household water quality rather than other factors such as type of materials, covering of the container, and material of the container, which did not cause any significant difference to the stored water quality.⁹ Jensen et al.²⁹ reported that interventions aimed at improving domestic domain contamination mattered only when the public domain transmission was clean. By contrast, the present study suggests that provision of safely improved water sources does not necessarily extend to safe water quality at the point of use in low-income settings. Overall, understanding the relative importance of methods of collection, transport, storage, and extraction is needed, especially where hand–water contact is highly likely to happen.

The elevated levels of *E. coli* in hand rinse samples of mothers and children as well as in stored drinking water indicated the presence of fecal contamination. However, previous studies have shown that *E. coli* originated from both human and animal sources.^{30,31} Future work should include microbial source tracking techniques to identify and quantify the sources of fecal contamination, especially in resource-poor countries. Recent research^{13,32,33} has demonstrated that human and animal fecal contamination detected on the hands of mothers and children were highly correlated with fecal contamination detected in stored drinking water in the low-income country setting. We also could not rule out the possibility of bacterial contamination from soils to hands and in water. Higher concentrations of *E. coli* from soil samples in the house floors as compared with soil samples collected from the latrine floor was observed in Tanzania and the study hypothesized that soil plays an important role in diarrheal illness transmission in low-income countries.¹⁸

Household stored drinking water management practices and hand hygiene.

Treatment of stored water before drinking did not change *E. coli* levels in treated versus untreated water. There are two main possible reasons for the nonsignificant change in the *E. coli* levels in stored water from treated versus non-treated water: first, the households had recall bias when they reported the treatment of drinking water before drinking and second, there could be the possibility of recontamination in the stored drinking water after storing in the households. There was no significant difference in water quality in terms of *E. coli* in boiled versus not-boiled water in Peru.³⁴ Similar levels of bacterial contamination in treated and untreated water were reported in Tanzania.¹⁴ There were no significant differences observed in *E. coli* levels when the drinking water was treated at the point of storages in houses with contaminated storage samples and houses with uncontaminated storage samples in urban slums in Hyderabad, India.¹¹ Meta-analyses showed that household interventions were effective in reducing diarrheal disease burden.³⁵ However, the possible inflated health benefits of household water treatment interventions could be because of various biases (e.g., responder and observer bias, and publication bias).³⁶

Study limitations and future work. A limitation of the study was that diarrheal health outcomes, management practices, and hygiene behavior used in the models were self-reported by the respondents, and may introduce bias and inaccuracy in the estimates as reported by other researchers.^{14,17} Moreover, the data were collected during the monsoon season and, hence, the effect of seasonal variation of *E. coli* levels in stored water and on hands was not known during winter and summer seasons. The findings of the study were based on assumptions of *E. coli* as an indicator characterizing human and animal feces. The WHO recommends *E. coli* because of its better performance regarding pathogen presence, especially in tropical regions and a stronger association with diarrheal risk.^{37–39} However, *E. coli* may be autochthonous in the present study region, and there are very limited studies showing the association between indicators and pathogens, especially in Asia.⁴⁰ Therefore, future studies should include additional molecular fecal indicators such as human-specific *Bacteroidales* to link fecal contamination to health outcomes in low-income countries. The study was limited in terms of obtaining data from 152 selected households, and future studies should include large sample size to obtain the statistical power to observe the significant relationships, but the present study will provide an initial assessment of the presence of indicator bacteria on the hands, source water, and stored drinking water.

An effective point-of-use water treatment can help in reducing waterborne illnesses in developing countries. Our study has shown that hands may play a vital role in household stored drinking water contamination and, hence, future work should look into the relative contribution of additional exposure transmission routes within household environments. In conclusion, the results of this study will help elucidate the relationships between microbial water quality, hand hygiene, and health outcomes at household levels in a peri-urban setting.

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Authors' addresses: Arti Kundu, Department of Veterinary Medicine and Epidemiology, University of California, Davis, CA, E-mail: akundu@ucdavis.edu. Woutrina A. Smith, Veterinary Medicine and Epidemiology Department, School of Veterinary Medicine, University of California at Davis, CA, E-mail: wasmith@ucdavis.edu. Danielle Harvey, Department of Public Health Sciences, School of Medicine, University of California, Davis, CA, E-mail: djharvey@ucdavis.edu. Stefan Wuertz, Department of Civil and Environmental Engineering, University of California, Davis, CA, Singapore Centre for Environmental Life Sciences Engineering, Nanyang Technological University, Singapore, Singapore, and School of Civil and Environmental Engineering, Nanyang Technological University, Singapore, Singapore, E-mail: swuertz@ucdavis.edu.

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