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Measuring household consumption and waste in unmetered, intermittent piped water systems

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Kumpel, Emily
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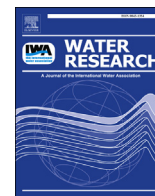**Publication Date**

DOI

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Field efficacy evaluation and post-treatment contamination risk assessment of an ultraviolet disinfection and safe storage system

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ABSTRACT

Inconsistent use of household water treatment and safe storage (HWTS) systems reduces their potential health benefits. Ultraviolet (UV) disinfection is more convenient than some existing HWTS systems, but it does not provide post-treatment residual disinfectant, which could leave drinking water vulnerable to recontamination. In this paper, using as-treated analyses, we report on the field efficacy of a UV disinfection system at improving household drinking water quality in rural Mexico. We further assess the risk of post-treatment contamination from the UV system, and develop a process-based model to better understand household risk factors for recontamination. This study was part of a larger cluster-randomized stepped wedge trial, and the results complement previously published population-level results of the intervention on diarrheal prevalence and water quality. Based on the presence of *Escherichia coli* (proportion of households with ≥ 1 *E. coli*/100 mL), we estimated a risk difference of -28.0% (95% confidence interval (CI): -33.9%, -22.1%) when comparing intervention to control households; -38.6% (CI: -48.9%, -28.2%) when comparing post- and pre-intervention results; and -37.1% (CI: -45.2%, -28.9%) when comparing UV disinfected water to alternatives within the household. We found substantial increases in post-treatment *E. coli* contamination when comparing samples from the UV system effluent (5.0%) to samples taken from the storage container (21.1%) and drinking glasses (26.0%). We found that improved household infrastructure, additional extractions from the storage container, additional time from when the storage container was filled, and increased experience of the UV system operator were associated with reductions in post-treatment contamination. Our results suggest that the UV system is efficacious at improving household water quality when used as intended. Promoting safe storage habits is essential for an effective UV system dissemination. The drinking glass appears to represent a small but significant source of recontamination that is likely to impact all HWTS systems.

1. Introduction

Household water treatment and safe storage (HWTS) is an important option for people whose drinking water sources do not meet microbiological water quality guidelines (Mintz et al., 1995; Rosa and Clasen, 2010). Several studies have found that HWTS can reduce self-reported diarrhea outcomes (Arnold and Colford Jr, 2007; Clasen et al., 2009; Fewtrell et al., 2005; Sobsey, 2002). However, it remains a major challenge for HWTS programs to achieve higher rates of adoption and consistent use (Brown and Clasen, 2012; Clasen, 2008; WHO and UNICEF, 2012a). Consistent use of existing HWTS systems has been limited by the perceived negative taste of chlorine; the dependence on the constant acquisition of chlorine and coagulation products; and the relatively long wait times for treatment via solar disinfection, boiling, and certain filtration systems (Sobsey et al., 2008). From the user's perspective, ultraviolet (UV) disinfection, where technologically feasible, may be

an attractive option because it is a fast process that does not require consumables and does not negatively impact the aesthetic characteristics of water.

Although UV disinfection is an established technology and has been effective both for centralized and point-of-use systems (Abbaszadegan et al., 1997; Colford et al., 2009; EPA, 2006; Hijnen et al., 2006), there have been only a few evaluations of its effectiveness in developing country households (Brownell et al., 2008; Gruber et al., 2014a, 2013; Reygadas et al., 2007). Water quality can degrade during household storage (Kumpel and Nelson, 2013; Levy et al., 2008; Wright et al., 2004), and thus assessing the risk and potential determinants of post-treatment contamination is particularly important for UV systems because they do not produce a residual disinfectant.

We conducted a cluster-randomized trial to evaluate an HWTS program based on a UV disinfection and safe storage system. The research objectives were to: (i) measure the field efficacy of the system in improving water quality (*Escherichia coli* levels), (ii) assess the risk of post-treatment contamination, and (iii) develop a process-based model to better understand household risk factors that drive recontamination. As part of this trial, we also measured the health and water quality impacts and the levels of adoption and consistent use achieved by the program. We have elsewhere reported the population level impacts on drinking water quality and diarrheal prevalence (Gruber et al., 2013, 2014a), and the results on adoption and consistent use (Reygadas, 2014).

2. Background

2.1. Study site

We conducted our field trial in 24 rural communities in Baja California Sur, Mexico. Participating communities ranged from 8 to 31 households, and had limited access to urban centers and basic services. Only 14% of households were connected to the electricity grid, and 81% had solar panels. The main economic activities were livestock ranching, small-scale farming, and fishing. Most households relied on springs and shallow wells for their drinking water; 20% of the study population regularly bought *garrafon*-bottled water (reusable 20-L narrow-necked containers, filled with treated water) from urban vendors. Locally-sourced water was commonly stored in wide-mouth containers (e.g., 200 L barrels, buckets, plastic water coolers, and *tinajas* — traditional clay or rock containers) (Gruber et al., 2013). Except for *garrafones*, and to some extent water coolers, water was typically extracted by dipping a cup into the storage container.

2.2. Description of the intervention

The Mesita Azul ("little blue table" in Spanish) safe water program was developed through a collaboration between the University of California, Berkeley and Fundación Cántaro Azul, a non-profit organization based in Mexico (Reygadas et al., 2009). The program consisted of an ultraviolet disinfection system (Mesita Azul), a 20-L narrow-necked container (*garrafon*) for storing treated water, and outreach activities intended to increase access to and consumption of safe water in rural households.

The Mesita Azul was designed as an easy-to-use and attractive water treatment system for low-income settings (Fig. 1). It uses a low-pressure UV lamp (254 nm) to inactivate bacteria, viruses, and protozoa, without affecting the physicochemical characteristics of water (including temperature and taste). The system operates at flow rates of up to 5 L/min, allowing households to treat their daily drinking water in less than five minutes. While in operation, the system consumes 20 W of electricity, equivalent to a small compact fluorescent lamp. For Mexico, the Mesita Azul program was coupled with a *garrafon* because it is ubiquitous and is widely perceived as a safe drinking water storage container.

The Mesita Azul was developed based on the UV Tube design principles (Brownell et al., 2008). Under standard conditions it delivers a germicidal fluence of 1224 ± 66 J/m^2 (95% confidence interval), determined from biological assays using MS2 coliphage, and following Section 6.3 of the NSF/ANSI Standard 55 as a microbiological performance test model (NSF, 2002). This dose meets the WHO's "highly protective" microbial performance target for household water treatment (WHO, 2011a) and exceeds by three times most other UV disinfection standards (DVGW, 2006; NSF, 2002; ÖNORM, 2001). The high design dose allows the system to maintain its germicidal effectiveness throughout the lamp's lifetime and for water with absorbance up to 0.1 cm^{-1}.

The Mesita Azul program, implemented by Cántaro Azul, included a needs assessment, a community presentation on safe water, enrollment of program participants, household installation of UV systems, training of household members to operate and provide basic maintenance on the UV system, training of several technicians in each community to carry out system repairs, and a follow-up visit to support technicians and households that reported any problems using the system. During the needs assessment, Cántaro Azul staff tested the water in each community for absorbance (at 254 nm), arsenic, nitrates, and total dissolved solids. The program was rolled out in communities whose drinking water was at risk of microbiological contamination and met the system's operation guidelines (absorbance at 254 nm < 0.1 cm^{-1}; most low turbidity sources meet this criterion, except when iron or manganese are present), but did not contain other tested contaminants that could not be addressed by UV treatment. To enroll in the program, households had to make a one-time payment of USD$20 (MXN$250) or commit to paying $24 (MXN$300) in installments over a six-month period. The cost of the UV system for this initial production round was approximately USD$80 (MXN$1,000).

3. Materials and methods

3.1. Study design

Our research team conducted a cluster-randomized stepped wedge trial to evaluate the Mesita Azul as it was rolled out to 444 households in the study communities (Gruber et al., 2013). The trial lasted 18 months. Cántaro Azul agreed to randomize the sequence of program rollout at the community level; this balanced covariates between control and intervention periods (Brown and Lilford, 2006; Hussey and Hughes, 2007) and created two comparable groups (Gruber et al., 2013). All communities started in the control group, and at each "step" households in four new communities crossed-over to the intervention group (Fig. 2). Cántaro Azul staff carried out key program activities (community meetings and UV system installations) during the step in which clusters crossed-over to the intervention group. Our evaluation team visited all communities to measure outcomes at baseline and during each subsequent step. By the end of step six, Cántaro Azul had rolled out the program to all 24 communities and the evaluation team had visited each cluster at least seven times.

We registered this study at ClinicalTrials.gov (NCT01637389); the Office for the Protection of Human Subjects at the University of California, Berkeley approved all research protocols (CPHS 2009-1-47); and all participating households provided informed consent.

3.2. General data and sample collection procedures

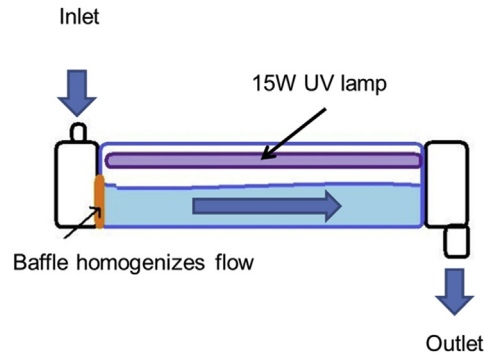In each survey visit, we collected data on the demographics,

Fig. 1. Image of the Mesita Azul and schematic of its disinfection chamber.

socioeconomic characteristics, and health status of household members. We also documented processes and conditions of all drinking water management practices used by households. During post-intervention visits, we recorded user interactions with the UV system and checked system functionality.

In each household visit, we asked respondents to identify all water access points in the home that had been used for drinking by any household member in the past seven days. When only one was reported, respondents identified an alternative point of access that they would use if their preferred access point were not available. Respondents provided us with water from each of the identified access points as though they were getting a drink (typically in a glass or a cup), from which we collected samples in 100 mL sterile containers. This approach allowed us to assess the quality of water immediately before ingestion.

3.3. Sample analysis

We used the concentration of E. coli as an indicator of fecal contamination (Tallon et al., 2005). We stored sample bottles in hermetically sealed containers inside a cooler with water and ice for up to 12 h. Samples were processed using IDEXX (Westbrook, ME, USA) Colilert 18 and Quanti-Tray 200 products. We incubated trays for 18–24 h at 36 °C (±4 °C) and determined the most probable number (MPN) of E. coli using the manufacturer's table (detection range of 1–200 MPN/100 mL).

To maintain quality control, we collected blank and duplicate samples filled from bottles with sterile water during household visits throughout the study. None of the blank samples (N = 137) tested positive for E. coli and only 4.3% of the duplicate samples (N = 46 matched pairs) had different presence–absence results. No significant difference was seen in a paired t-test between duplicate

Fig. 2. Study design: cluster randomized stepped wedge trial. Note that clusters 21–24 received a second post-intervention visit at the end of the study.

samples using MPN results.

3.4. Data analysis

We converted *E. coli* concentrations to a presence–absence binary outcome to compare the risk of contamination between different water management practices and to develop our process-based recontamination model. This allowed us to carry out more robust statistical analyses. We used presence–absence outcomes following the World Health Organization's (WHO) drinking water guidelines (*E. coli* must not be detectable in any 100 mL sample) and considering the limited evidence for increased risk of diarrhea beyond the 1 *E. coli*/100 mL cutoff (Gruber et al., 2014b; WHO, 2011b).

To complement our statistical analyses, we used a priority assessment classification based on the observed *E. coli* concentration (MPN counts/100 mL): Low Risk [0,1); Intermediate Risk [1,10); High Risk [10,100); and Very High Risk [100,+∞) (WHO, 2011b). This step allowed for more descriptive water quality results across different management practices.

We selected the types of outcomes, comparison tests, and model variables *a priori* based on the type of data being analyzed. All data analyses were conducted using Stata 12 (Stata Corp, College Station, TX, USA).

3.5. Field efficacy evaluation

3.5.1. Controlled comparison tests

Household drinking water quality is influenced by multiple factors internal and external to the Mesita Azul system, including: water source characteristics, seasonality of environmental conditions, water handling processes, hygiene and sanitation conditions, a household's awareness of the relationship between water and health, and operation and performance of the UV system. To isolate and evaluate the field efficacy of the Mesita Azul, we used an as-treated analysis, in which we defined treated households as those that used the Mesita Azul correctly, as promoted by the safe water program. Compliance with the Mesita Azul program was defined as having UV-treated water (based on self-report) safely stored in a *garrafon* (based on visual observation) during an unannounced evaluation visit. As reported by Gruber et al. (2013), 51% of household observations in intervention periods complied with the Mesita Azul. In contrast with previous analyses (Gruber et al., 2014a, 2013) that combine multiple treatment strategies into one safe water compliance index, we disaggregated "compliance" of treatment strategies to isolate the efficacy of the Mesita Azul. To address biases that can result from as-treated analyses (Friedman et al., 1998), we developed a robust assessment based on three types of controlled comparisons. For all comparisons, we used samples collected exclusively from drinking glasses.

3.5.1.1. Intervention vs. control. We compared drinking water quality between complying households in intervention periods and households in control periods that would later acquire a UV system. The stepped wedge design allowed us to identify future compliers in control periods based on observed behavior after crossover to the intervention periods (Gruber et al., 2014a). Comparing the compliers in the intervention group to the entire control group could have introduced a bias because of non-compliers in the control group. We computed risk differences and 95% confidence intervals (CI) using a chi-square test (χ^2).

3.5.1.2. Intervention vs. pre-intervention. We compared drinking water quality post- and pre-intervention. We restricted this analysis to complying households during the step at which the intervention was introduced, and compared water quality to those same households one step prior to the intervention. We included only households that had matched data available from both steps. The seasonal variation of water quality could have introduced a time bias in this comparison. We calculated risk differences and 95% confidence intervals using the McNemar test for paired data, which does not require independent observations (McNemar, 1947).

3.5.1.3. Intervention vs. alternative. We compared the quality of drinking water treated with the UV system and stored in a *garrafon* to that of drinking water from an alternative access point inside the household. We selected the alternative access point by asking the respondent from where she would drink if she did not have UV-treated water available. By collecting two matched samples from the same location at the same time we could control for seasonal effects. However, alternative water sources might have been managed differently once the household had access to UV-treated water stored in a *garrafon*. We used the McNemar test to calculate risk differences and 95% confidence intervals.

3.5.2. Comparison with other treatment and storage alternatives

We compared the presence of *E. coli* in drinking water treated with the UV system and stored in *garrafones* to other treatment and storage practices. These alternatives were: purchasing *garrafon*-bottled water, in-home chlorination, boiling, and storing UV-treated water in containers other than *garrafones*. In these comparisons we pooled samples collected throughout the study from both intervention and control groups for each alternative water management practice.

3.5.3. Safe drinking water reliability framework

According to Quantitative Microbiological Risk Assessment model estimates, even sporadic consumption of contaminated water can attenuate the health benefits of potable water interventions (Brown and Clasen, 2012; Enger et al., 2013; Hunter et al., 2009). To consistently drink safe water, people need to consume water from access points that are reliably safe. We created a framework to assess reliable access to safe drinking water, and used it to compare UV disinfection and safe storage with non-UV access points and with *garrafon*-bottled water. For a given water management practice, we pooled samples collected at different points in time for each household and computed the proportion of samples that had non-detectable levels of *E. coli*. We used only samples from drinking glasses in households that had at least three samples throughout the study from the same water practice. We categorized the reliability of a water practice by the proportion of samples with non-detects (*E. coli* was absent) for each household: Always Safe [100.0%, 100.00%]; Mostly Safe (100.0%, 66.6%); Often Contaminated (66.6%, 33.4%); Mostly Contaminated [33.4%, 0.0%); and Always Contaminated [0.0%, 0.0%]. We used these categories to create graphs that showed the percentage of households per level of reliability for each water management practice.

3.6. Post-treatment contamination risk assessment

3.6.1. Water quality at the outlet, storage container, and drinking glass

During baseline (before UV systems had been implemented), we collected matched samples directly from a small non-random subset of storage containers with drinking water and from glasses filled from the same containers. We asked respondents to pour water from a storage container into a 100 mL sterile recipient exactly as they would fill a glass for drinking (e.g., opening a spigot, using a pump, dipping into the container, or pouring from the top). We then collected a matched sample from the glass (see Section 3.2 above) and used McNemar tests to calculate risk differences and

We assessed water quality at different phases of UV treatment and safe storage, by aggregating samples collected from the outlet of the UV system (during the second post-intervention visit, once users were accustomed to operating their system), directly from *garrafones* with UV-treated water (when there was a second *garrafon* available; not matched with drinking glass samples), and glasses filled from *garrafones* with UV-treated water (in complying households throughout intervention periods). We also carried out a controlled test during the second post-intervention visit to reduce biases that could arise from aggregating samples from different households at different points in time. For this analysis, we first collected a sample from a drinking glass filled with UV-treated water in a *garrafon*. Then we asked the interviewee to fill a *garrafon* using the UV system, and we collected the first 100 mL extracted from the outlet. These comparisons showed the impacts of storage and the use of a glass, independent of each other, on drinking water quality.

3.6.2. Process-based recontamination model

Recontamination levels are modulated by processes (treatment, storage, washing of the storage container, and extraction from the storage container) and conditions (household and human hygiene) associated with a given water management practice (see Fig. S1 in the Supplemental Information (SI)). These processes and conditions are directly related to in-home water management. Unlike explanatory variables that are less viable for water programs to act upon, such as income, education, gender, or age of household members, the results of a process-based recontamination model could be used to improve the Mesita Azul program or to develop other effective HWTS programs.

We developed a logistic regression model of *E. coli* contamination with explanatory variables that represent processes and conditions associated with managing water treated with the UV system and stored in *garrafones* (Table 1). We used the presence or absence of *E. coli* (in 100 mL) as the outcome variable. We controlled for time (fixed effect for evaluation step) and used a robust estimator of variance to compute the contamination odds ratios and their 95% confidence intervals for each explanatory variable. We limited the model to samples collected, directly or via a drinking glass, from *garrafones* with UV-treated water. Table 1 shows our hypothesized effects on contamination for each process and condition, the respective explanatory variables, operational definitions, and types of data collected. All variables were selected prior to analyzing the data. We excluded a washing variable (time since container was last washed) due to its relatively high correlation coefficient (>0.25) with a storage variable (time since container was last filled).

4. Results

4.1. Field efficacy evaluation

4.1.1. Controlled comparison tests

We found that treating water with the UV system and storing it in *garrafones* resulted in significant improvements in drinking water quality (Fig. 3). We calculated the risk difference for each comparison group based on the proportion of samples with *E. coli* $\geq$ 1 MPN/100 mL.

4.1.1.1. Intervention vs. control. We recorded 449 intervention household observations (Fig. 3). Out of the recorded 947 control household observations, 542 met the criteria for this comparison (restricting analysis to households that would later acquire a UV system). We found a risk difference of -28.0% (CI: -33.9%, -22.1%; χ^2) when comparing the presence of *E. coli* in samples collected

from glasses filled from *garrafones* with UV-treated water (Mesita Azul program compliers: 29.4%; N = 449) to control households that would become compliers after crossing-over to the intervention group (57.4%; N = 542). Control samples were collected from glasses filled from preferred access points: 79% no treatment, 20% *garrafon*-bottled water, <1% boiling, and <1% chlorination.

4.1.1.2. Intervention vs. pre-intervention. We identified 140 household observations for this comparison (Fig. 3). We found a risk difference of -38.6% (CI: -48.9%, -28.2%; McNemar) between samples matched by household and collected from drinking glasses of UV-treated water in *garrafones* during the step at which the intervention was implemented (24.3%; N = 140), and samples collected from glasses filled from preferred access points during the step prior to crossing-over to the intervention (62.9%; N = 140). We found no significant water quality trends across the study in samples collected from intervention and control households.

4.1.1.3. Intervention vs. alternative. Out of the 449 intervention household observations, 224 met the criteria for this comparison (Fig. 3). We found a risk difference of -37.1% (CI: -45.2%, -28.9%; McNemar) between samples matched by household, comparing samples collected from glasses filled from *garrafones* with UV-treated water (25.9%; N = 224) to samples collected during the same visit from glasses filled from alternative access points (62.9%; N = 224).

We classified samples into four risk categories based on MPN *E. coli*/100 mL (Low Risk [0,1); Intermediate Risk [1,10); High Risk [10,100); and Very High Risk [100,+∞)) to further explore these results. Post-intervention water quality improvements were mostly driven by lower frequencies of the High and Very High Risk categories across all three comparisons (Fig. 3).

4.1.2. Comparison with other treatment and storage alternatives

We compared the fraction of *E. coli* positive samples collected from households that complied with the Mesita Azul program to samples collected from households reporting the use of other treatment alternatives. We collected all samples from drinking glasses. We found no difference (risk difference = 1.9%; CI: -3.5%, 7.4%; χ^2) in water quality between samples from access points that complied with the Mesita Azul program (25.9%; N = 624) and samples from purchased *garrafon*-bottled water (24.0%; N = 387). We observed a non-statistically significant risk difference of -9.9% (CI: -25.4%, 5.5%; χ^2) between samples that complied with the Mesita Azul program (26.0%; N = 624) and boiled or chlorinated samples (35.9%; N = 39). In contrast to these treatment alternatives, samples collected from un-treated (Non-disinfected) access points used for drinking were more likely to test positive for *E. coli* (63.7%; N = 1781) (Fig. 4).

To minimize the risk of post-treatment contamination, program staff strongly encouraged people to store UV-treated water only in *garrafones*. However, 40% of households stored UV-treated water in other containers (*tinajas*, buckets, and plastic water coolers) at least once during the study. We found a risk difference of -21.8% (CI: -29.2%, -14.3%; χ^2) between UV-treated samples stored in *garrafones* (26.0%; N = 624) and stored in alternative containers (47.7%; N = 220).

4.1.3. Safe drinking water reliability framework

We assessed the reliability of water quality for the most prevalent water management practices observed during our study. For households that had at least three samples, collected at different times, from glasses filled from *garrafones* with UV-treated water, we found that 37% met the Always Safe category, 3% the Always Contaminated, and the remaining 60% had both *E. coli* positive and

Table 1
Origin and structure of explanatory variables used in Mesita Azul recontamination model.

Water management processes and conditions	Potential effects on contamination level	Explanatory Variables	Operational definitions	Types of collected data
Washing	Using untreated source water could introduce contamination to container.	Type of water used for washing storage container.	Used disinfected water last time they washed container?	Reported; binary
	Using cleaning supplies could reduce contamination.	Type of cleaning supplies used for washing storage container.	Used bleach or soap last time they washed container?	Reported; binary
Treatment	Working system should reduce contamination.	Operational status of the system.	Does the UV system work at time of visit?	Observed; binary
	Operating the system correctly should reduce contamination.	Ability of operator to use system.	Is the operator an expert? (Knows the operation steps in perfect order and carries them out with confidence.)	Observed; binary
Storage	Storage time could modulate contamination via environmental exposure and bacterial growth or decay.	Length of time that water has been in storage container since it was last filled.	Time since container was last filled.	Reported; continuous (Time unit = 1 day; range from 0 to 11.)
	Exposure to the environment during storage could increase contamination.	Type of exposure of stored water to the environment.	Is container covered with proper lid at time of visit?	Observed; binary
Extraction	Extractions from storage container could increase contamination.	Number of water extractions since storage container was last filled.	Number of extractions (in multiples of 10) since container was last filled. (Calculated based on remaining volume, assuming container was filled to top.)	Observed; continuous (Each extraction = 400 mL; range from 0 to 50.)
	The contamination risk during the extraction process could vary across different extraction methods.	Type of mechanism used to extract water from storage container.	Extraction with pump? Extraction through spigot? (In contrast with tilting container and pouring water directly from it.)	Observed; categorical
	Pouring extracted water into drinking glass could increase contamination.	Point at which sample is collected during the extraction process.	Is sample collected from drinking glass? (As opposed to directly from extraction mechanism.)	Observed; binary
Hygiene	The overall infrastructure of the house could modulate contamination risks.	Type of household infrastructure.	Does household have concrete floors?	Observed; binary
	The hygiene of the kitchen could modulate contamination risks.	Level of kitchen hygiene.	Are the kitchen hygiene conditions good or very good? (Evaluated based on presence of flies, trash, and exposed food.)	Observed; binary
	The hygiene of people in the house could modulate contamination.	Type of facilities available for hand washing.	Is there a water access point used mainly for hand washing? (Dedicated hand washing station.)	Observed; binary

negative samples (N = 97 households; 45% = three samples; 31% = four samples; 24% = five samples) (Fig. 5A). In contrast, for households with at least three samples from drinking glasses filled

from non-UV access points, 13% met the Always Safe category, 22% the Always Contaminated, and the remaining 65% had both positive and negative samples (N = 171 households; 31% = three samples;
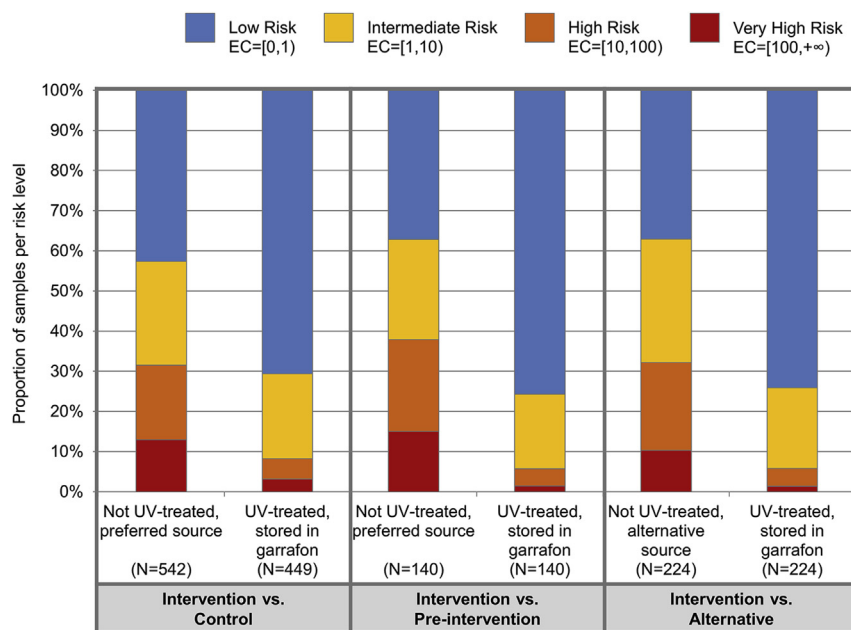

Fig. 3. *E. coli* (EC) risk levels (based on MPN/100 mL) for the controlled comparison tests. Sample units (N) represent the number of household observations throughout the study. All samples were collected from drinking glasses.
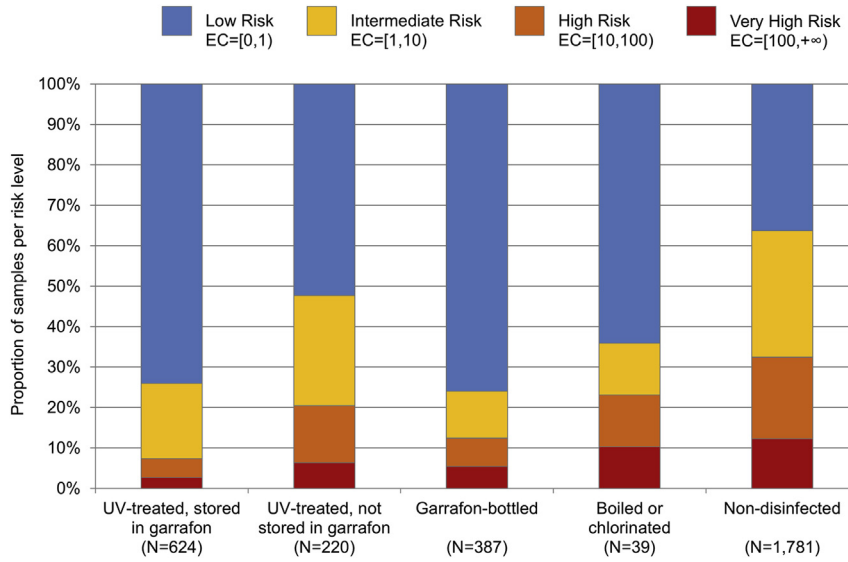
Fig. 4. Fraction of *E. coli* (EC) samples in each risk level for water collected in drinking glasses from different water access points. Sample units (N) represent the number of household observations throughout the study.

32% = four samples; 37% = five samples) (Fig. 5B). The analysis of non-UV samples collected during control periods only among households that later adopted the UV system led to similar results: 18% met the Always Safe category, 28% the Always Contaminated, and the remaining 54% had both positive and negative samples (N = 132 households). The reliability of the UV-treated water stored in *garrafones* and that of commercially purchased *garrafon*-bottled water was equivalent (Fig. S2).

4.2. Post-treatment contamination risk assessment

4.2.1. Water quality at the outlet, storage container, and drinking glass

We found a statistically significant risk difference of −9.4% (CI: −18.0%, −0.9%; McNemar) in the contamination of matched

samples collected during baseline (before the UV intervention) directly from containers (30.2% positive for *E. coli*; N = 106) and from glasses of water from the same containers (39.6%; N = 106). In this subset of baseline samples, most containers had been filled with treated water (67.0%) and had safe-storage characteristics (82.1%). A higher proportion of samples in the Intermediate Risk category accounted for most of the additional contamination in the drinking glasses. We observed a similar but not significant effect (risk difference = −7.9%; CI: −19.4%, 3.6%; McNemar) in a smaller number of paired samples collected directly (21.1% positive for *E. coli*; N = 76) and through a drinking glass (28.9%; N = 76) from *garrafones* with UV-treated water during intervention periods.

Aggregating data water throughout the study, we found *E. coli* in 5.0% of samples (N = 161) collected directly from the outlet of the UV system; 21.1% (N = 76) from *garrafones* of UV-treated water; and
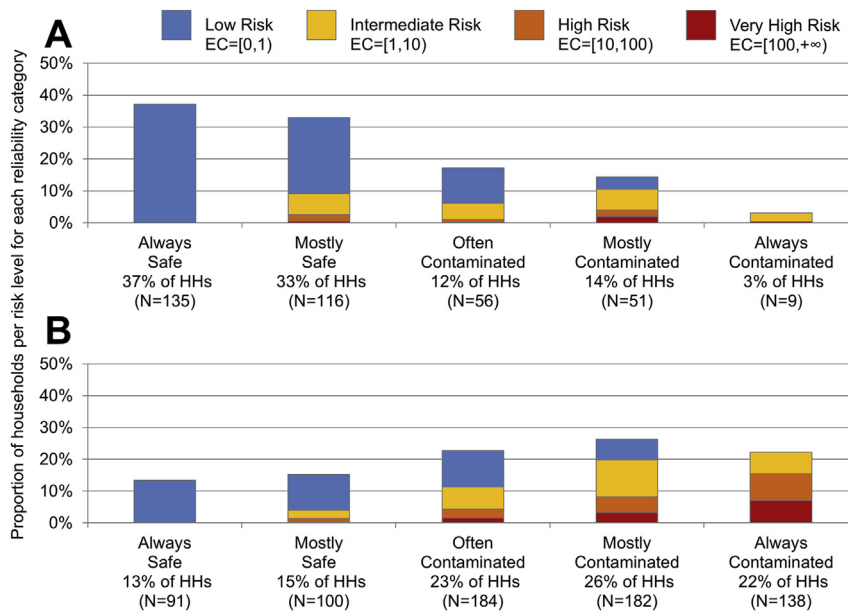

Fig. 5. Water quality reliability for: (A) UV-treated and safely stored water and (B) Preferred access points in control periods. Percentages in the columns represent the proportion of households (HHs) in each reliability category. Sample units (N) represent the number of household observations throughout the study. All samples were collected from drinking glasses.

26.0% (N = 624) from drinking glasses filled from *garrafones* of UV-treated water (Fig. 6). During the second post-intervention visit, we found an increased risk of *E. coli* contamination (risk difference = 16.1%; CI: 8.2%, 24.0%; McNemar) between matched samples from a glass (19.5%; N = 118) and from the outlet of the Mesita Azul (3.4%; N = 118).

4.2.2. Process-based recontamination model

We present the results of our process-based recontamination model in Table 2. None of the Washing process variables had statistically significant associations with the presence of *E. coli* in water. Both Treatment variables resulted in significant reductions in contamination. Once UV-treated water was stored in a *garrafon*, the duration of storage had a significant protective effect on contamination: each additional day since the container had last been filled reduced the odds of contamination by 19% (observations ranged from 0 to 11 storage days; Table S1). Having the storage container covered appeared to have a protective effect on contamination, but was not significant; however, only 2% of the containers were not covered. Of the Extraction variables, each additional 10 servings (1 serving = 400 mL) from the *garrafon* reduced the odds of contamination by 16%, but the extraction mechanism was not significantly associated with contamination. Samples from drinking glasses had increased odds of contamination compared to samples collected directly from the containers. Among Hygiene variables, households with concrete floors had 64% lower odds of contamination. Kitchen hygiene and access to a dedicated hand washing station were not significantly associated with recontamination.

5. Discussion

Through this field efficacy study we measured the impact of the Mesita Azul system on the microbiological quality of drinking water (presence of *E. coli*) among households that complied with the treatment and storage instructions (as observed at the time of a survey visit). Complementing our previous evaluation of the Mesita Azul program as a whole (Gruber et al., 2014a, 2013), these as-treated analyses allowed us to estimate the current maximum potential efficacy of the system. We performed a series of comparison tests to minimize any selection biases from any single as-treated analysis and to develop a more robust impact assessment (Friedman et al., 1998). We built on the efficacy results to develop a process-based recontamination model of household risk factors for *E. coli* contamination.

5.1. Controlled comparisons

Compliance with the Mesita Azul system significantly reduced the presence of *E. coli* in drinking water. The risk differences in our comparison tests were −28.0% (Intervention vs. Control), −38.6% (Intervention vs. Pre-intervention), and −37.1% (Intervention vs. Alternative). These values are greater than our previous effectiveness evaluations, which showed risk differences of −19% (intention-to-treat analysis) (Gruber et al., 2013) and of −22% (complier average causal effects for any method) (Gruber et al., 2014a). The observed additional benefits of compliance with the Mesita Azul, independent of other treatment alternatives, justify investments to increase program adoption and consistent use of the system.

A comparison of differences in *E. coli* concentrations revealed that water quality improvements in UV-treated and safely stored water were mostly driven by reducing the number of households with water in the High and Very High Risk categories. This could be caused by a higher concentration of *E. coli* in source water (which would affect the quality of samples collected from untreated access points) or by differences in recontamination mechanisms between the *garrafon* and other storage containers (leading to higher concentrations in the latter).

Despite significant reductions in *E. coli* after compliance with the intervention, 24.3–29.4% of samples collected from drinking glasses of UV-treated water stored in a *garrafon* had detectable levels of *E. coli*. In comparison, a study in Ecuador (Levy et al., 2014) found that 48.8–61.3% of samples collected from storage containers with chlorinated water had detectable levels of *E. coli*. Chlorinated water should have lower contamination levels due to its residual disinfection capacity, but there may have been higher prevalence of *E. coli* in the source water or higher risks of recontamination in the Ecuador study. Furthermore, Levy et al. (2014) collected samples directly from containers, whereas we collected samples from drinking glasses. Had we collected samples directly from *garrafones*
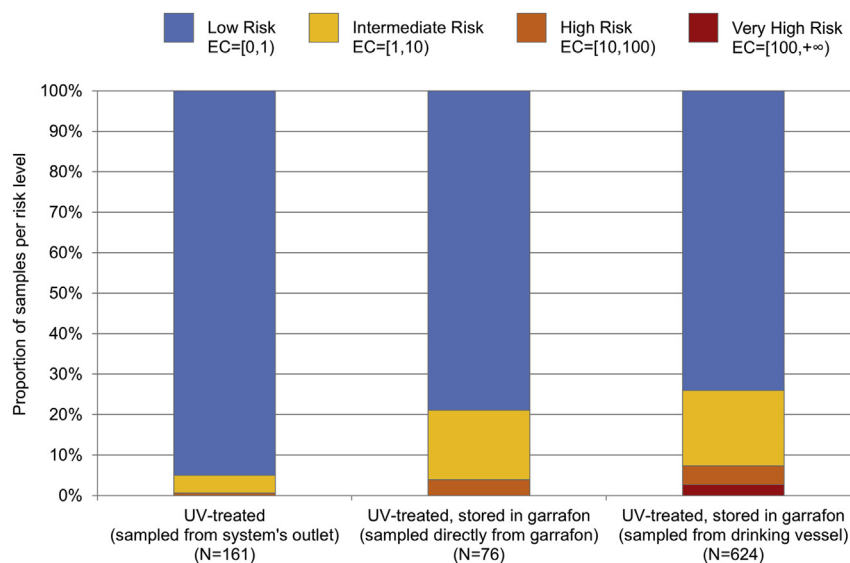

Fig. 6. Fraction of *E. coli* samples in each risk category for water collected from the outlet of the UV system, from storage containers of UV-treated water, and from drinking glasses filled from these storage containers. Sample units (N) represent the number of household observations throughout the study.

Table 2
Results from process-based recontamination model. Variables that had statistically significant association with the presence of *E. coli* in water are in bold and with an asterisk next to the odds ratio (*).

Water management processes and conditions	Independent variables	% of positive observations (N = 619)	Odds ratios (OR)	Confidence intervals (95%)	
Washing	Used disinfected water last time they washed container?	18%	1.26	0.78	2.03
	Used bleach or soap last time they washed container?	62%	1.32	0.88	1.98
Treatment	**Does the UV system work at time of visit?**	**97%**	**0.26***	**0.10**	**0.68**
	Is the operator an expert?	**29%**	**0.61***	**0.37**	**1.00**
Storage	**Time since container was last filled.**	**>3 days = 32%**	**0.81***	**0.70**	**0.93**
	Is container covered with proper lid at time of visit?	98%	0.53	0.15	1.93
Extraction	**Number of extractions (in multiples of 10) since container was last filled.**	**≥25 ext.=62%**	**0.84***	**0.72**	**0.98**
	Extraction with pump vs. tilting container?	50%	0.88	0.56	1.37
	Extraction with spigot vs. tilting container?	43%	1.43	0.65	3.11
	Is sample collected from drinking glass?	**85%**	**1.91***	**1.02**	**3.57**
Hygiene	**Does household have concrete floors?**	**88%**	**0.36***	**0.20**	**0.65**
	Are the kitchen hygiene conditions good or very good?	86%	0.86	0.49	1.50
	Is there a water access point used mainly for hand washing?	20%	1.38	0.82	2.33

we would have observed an even lower fraction of contaminated samples in UV-treated water. These results from the UV system are encouraging. However, because of our as-treated analysis, they are generalizable only to households that would comply with the Mesita Azul in similar contexts.

5.2. Comparison with other treatment and storage alternatives

The Mesita Azul system allowed complying households to produce drinking water of equivalent quality to that of purchased *garrafon*-bottled water, suggesting that transferring the treatment responsibility from commercial bottling facilities in urban areas to individuals in rural households did not lead to an increase in *E. coli* contamination of drinking water.

Household chlorination and boiling were rare in the study area, and, in most cases, water treated by these methods was unsafely stored. This could explain the (non-statistically significant) higher proportion of contaminated samples when comparing chlorinated or boiled water to UV-treated water stored in *garrafones*. Storing UV-treated water in *garrafones* significantly reduced *E. coli* levels compared to other commonly used containers (*tinajas*, buckets, and plastic coolers). Our results imply that water treatment programs should strongly emphasize the recontamination risks of storing water in containers that are not covered, have a wide opening, or require dipping a cup for extraction (Mintz et al., 1995; Trevett et al., 2005). Going forward, the Mesita Azul program should incorporate more evidence-based behavior change strategies (Figueroa and Kincaid, 2010) to promote safe storage habits.

5.3. Water quality reliability

Consuming safe water consistently depends on the reliability of water quality at each access point used for drinking. Even sporadic consumption of contaminated water can limit the health benefits of water treatment systems (e.g. Brown and Clasen, 2012). Based on *E. coli* levels, compliance with the Mesita Azul program gave users more reliable access to safe drinking water than they had had with water management practices observed during control periods in our study. UV-treated and safely stored water, and purchased *garrafon*-bottled water, were equivalent in reliability.

We observed that 74% of UV-treated and safely stored water samples aggregated throughout the study fell in the Low Risk category (<1 *E. coli*/100 mL). Only 37% of households, however, met the Always Safe condition (i.e., all samples collected in each household were Low Risk). Many common sampling strategies for assessing the effectiveness of safe water programs, such as cross-

sectional or aggregated data, do not enable assessment of reliability. Although the WHO highlights the importance of monitoring HWTS programs (WHO and UNICEF, 2012a) and provides recommendations on sampling strategies (WHO, 2011b; WHO and UNICEF, 2012b), we found no specific suggestions on how to assess their field reliability. We encourage monitoring programs to incorporate a reliability assessment similar to that used in our study.

5.4. Post-treatment contamination risk assessment

We observed a small percentage of samples contaminated with *E. coli* at the outlet of the UV system, and a significant increase in contamination of UV-treated water after being stored in narrow-necked containers and served in drinking glasses. Considering the high germicidal dose delivered by the UV chamber of the Mesita Azul and the low absorbance of source water documented in study communities, we believe that *E. coli* in treated water was reduced to below the detection limit when users operated the system correctly. We hypothesize that most of the contaminated samples collected directly from the outlet of the system (3.4–5.0%) were due to non-working systems, improper operation, or contamination of the outlet itself. These hypotheses are supported by the results of our recontamination model, in which we found statistically significant associations between the presence of *E. coli* in drinking glasses and the state of the system or the ability of the operator. While 97% of the systems were working properly in our study, only 29% of the operators could perform the treatment steps in perfect order and with confidence when observed. We note that correct and consistent use of HWTS is challenging in general (Brown et al., 2009; Brown and Clasen, 2012), and recommend that, going forward, the Mesita Azul program consider simplifying its operational requirements and strengthening its operator training strategy.

Once UV-treated water had been stored, additional storage time and number of extractions resulted in a statistically significant protective effect on drinking water quality. An attenuation of *E. coli* in stored water with time was also reported by Levy et al. (2008), but we observed it in recontaminated water with much lower concentrations of *E. coli*. Bacterial die off and limited growth inside the *garrafon* could explain the observed negative correlation with storage time. Settling of bacteria associated with particles could explain the decline in *E. coli* concentration with increasing extractions, as pumps and spigots extract water from the bottom of the *garrafon*. Based on our results, disinfected water stored in a *garrafon* can be kept for at least 7 days (the number of observations became small and the variability of our results increased for storage times higher than this)

without posing an additional contamination risk (Table S1).

We found no significant association between *garrafon* washing processes and the presence of *E. coli* in drinking water. This was a surprising finding considering that fewer than 20% of households reported washing *garrafones* with disinfected water and approximately 60% of untreated water was contaminated. It may be that mixing the small volume of untreated water left over from the washing process with the large volume of disinfected water used to fill the *garrafon* resulted in a high dilution rate. We recognize that no association in our study does not mean that it is safe to wash containers with untreated water, given that *E. coli* is only one indicator of contamination, that some pathogens have low infectious doses, and that some untreated water could be highly contaminated.

We did not find significant associations between recontamination of drinking water and kitchen hygiene or the presence of a dedicated hand washing station. But household infrastructure conditions, particularly the presence of concrete floors, were significantly associated with lower water contamination levels. We expect that concrete floors, as opposed to dirt floors, allow people to better maintain household hygiene, which in turn reduces the risk of post-treatment contamination. Cattaneo et al. (2009) observed that a government program to implement concrete floors in urban Mexico improved child health outcomes, but cautioned against extrapolating these results to rural areas due to the likely transmission of pathogens through unsafe water. More research is needed to investigate potential synergistic effects between concrete floors, water, and health in a rural context, where concrete floors tend to cover a smaller share of the total household floor space.

We found evidence of contamination introduced at the drinking glass through direct comparisons and also via a statistically significant association in the recontamination model. Contamination at the drinking glass could come from water previously served in the glass or used to wash it, contact with soil or dirt, settling of dust into the glass, or contact with fomites. Contamination at the drinking glass affects most water management strategies, and no interventions (that we know of) have addressed this issue directly in rural settings. Due to the short contact time between serving and drinking, residual chlorine in water is unlikely to address recontamination that occurs at the glass, except possibly in subsequent servings. Washing glasses with soap, rinsing them with disinfected water, and improving the hygiene of areas where drinking glasses are kept (Oswald et al., 2007) are all likely to reduce recontamination at the drinking glass. This finding underscores the importance of collecting samples as close as possible to the point of ingestion when evaluating safe drinking water programs.

Although the process-based recontamination model (by design) does not allow us to derive causal inferences, the results were useful for generating broader hypotheses of the recontamination pathways for household water management. The model can be easily adapted for HWTS systems beyond the Mesita Azul. We recommend the incorporation of process-based models in trials that seek to evaluate the impact of HWTS programs.

6. Conclusions

- The Mesita Azul program allowed complying households (those that had UV-treated water stored in narrow-necked containers available during evaluation visits) to significantly reduce the presence of *E. coli* in drinking water.
- *E. coli* concentrations were similar in water collected from drinking glasses from *garrafones* (reusable 20-L narrow-necked containers) of UV-treated water and from purchased *garrafon*-bottled water. Thus, the UV system enabled isolated rural households to access drinking water of equivalent quality to that available via water bought from bottling facilities in urban areas.
- Storage of UV-treated water increased contamination, compared to Mesita Azul effluent. Contamination was significantly higher in containers that were not covered, had a wide opening, or required dipping a cup to extract water than in *garrafones*.
- For UV-treated water already stored in *garrafones*, the duration of storage and number of extractions were negatively correlated with the presence of *E. coli*.
- The presence of concrete floors was negatively correlated with recontamination of treated water.
- The use of a drinking glass further increased contamination, a finding that affects most drinking water management strategies.

Author contributions

Conceived and designed the experiments: FR JG KN IR. Performed the experiments: FR JG. Analyzed the data: FR. Contributed reagents/materials/analysis tools: FR JG KN. Wrote the paper: FR JG KN IR.

Acknowledgments

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.watres.2015.08.013.

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