Evaluating Four Measures of Water Quality in Clay Pots and Plastic Safe Storage Containers in Kenya

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Abstract

Household water treatment with chlorine can improve microbiological quality and reduce diarrhea. Chlorination is typically assessed using free chlorine residual (FCR), with a lower acceptable limit of 0.2 mg/L, however, accurate measurement of FCR is challenging with turbid water. To compare potential measures of adherence to treatment and water quality, we chlorinated recently-collected water in rural Kenyan households and measured total chlorine residual (TCR), FCR, oxidation reduction potential (ORP), and *E. coli* concentration over 72 h in clay and plastic containers. Results showed that 1) ORP served as a useful proxy for chlorination in plastic containers up to 24 hours; 2) most stored water samples disinfected by chlorination remained significantly less contaminated than source water for up to 72 hours, even in the absence of FCR; 3) TCR may be a useful proxy indicator of microbiologic water quality because it confirms previous chlorination and is associated with a lower risk of *E. coli*

contamination compared to untreated source water; and 4) chlorination is more effective in plastic than clay containers presumably because of lower chlorine demand in plastic.

1. Introduction

Despite substantial gains in access to improved drinking water sources worldwide since the Millennium Development Goals were developed and implemented, an estimated 663 million people still rely on unimproved water sources (UNICEF and WHO 2015). An additional estimated 1.2 billion people obtain drinking water from improved, but contaminated, water sources. Thus, an estimated 1.8 billion people lack access to safe water (Onda et al. 2013). Consumption of fecally-contaminated drinking water is a leading cause of the approximately 502,000 diarrheal deaths worldwide each year (Pruss-Ustun et al. 2014).

Chlorination is one of the most widely used, practical, and inexpensive forms of household water treatment to quickly inactivate most waterborne disease-causing bacteria and viruses (Rosa and Clasen 2010). In developing countries, liquid (e.g., sodium hypochlorite solutions) and powdered or solid (e.g., calcium hypochlorite or sodium dichloroisocyanurate) sources of free chlorine are used to disinfect household drinking water and, in a number of studies, chlorination has been shown to reduce the risk of diarrheal disease (Arnold and Colford 2007, Clasen et al. 2015).

Escherichia coli (E. coli) is used as an indicator of the microbiologic quality of water (Edberg et al. 2000). However, *E. coli* is difficult to measure in the field and other measureable water characteristics can be used as indicators of adherence to water chlorination recommendations, serving as proxies for microbiologic water quality (CDC 2014, OECD and WHO 2003, Crump et al. 2004). Following addition of chlorine to water, reactions occur that result in free chlorine species and combined chlorine species; the sum of these two is termed total chlorine. Free chlorine residual (FCR) is the most common measure used because it indicates the most effective species of chlorine for disinfection. Total chlorine residual

(TCR) is less frequently used as a water quality measure because it also detects combined chlorine species, which are much less effective for disinfection. Oxidation reduction potential (ORP) is another water chemistry parameter increasingly used in water distribution systems (Hall et al. 2007) and swimming pools (Kebabjian 1995). ORP is a measure of the tendency of oxidants (e.g., chlorine species) to be reduced and it therefore provides an indication of the disinfection capacity of the water.

The World Health Organization (WHO) recommends that FCR in treated water should not fall below 0.2 mg/L (WHO 2011). For treating water in the home, WHO recommends dosing clear water (<10 Nephelometric Turbidity Units [NTU] turbidity) at 2 mg/L FCR and turbid water (>10 NTU) at 4 mg/L FCR in order to maintain a FCR of 0.2 mg/L for 24 h after treatment (WHO 2011, Lantagne et al. 2010). Many studies of household water chlorination rely on a combination of self-reported use of chlorine and FCR field tests that utilize N,N-diethIy-p-phenylenediamine (DPD) to confirm water treatment. In these studies, discrepancies between reported and confirmed chlorination have been common (Blanton et al. 2010, DuBois et al. 2006, Gupta et al. 2007, Luby et al. 2008). Potential causes of these discrepancies include: 1) reliance on water sources with a high content of organic material that rapidly consumes chlorine (i.e., exerts chlorine demand) (Lantagne 2008); 2) use of clay pots, which are culturally preferred because they lower water temperature through evaporative cooling, but can exert chlorine demand (Null and Lantagne 2012, Ogutu et al. 2001); 3) use of wide-mouthed storage containers which facilitate insertion of hands or other objects that could add organic material and decrease FCR (Wright et al. 2004); 4) storage of water for periods exceeding 24 hours, a common practice in regions in which water is scarce or water sources are located far from homes, during which time FCR naturally decays (Lantagne 2008, Briere et al. 2012, Colindres et al. 2008) and; 5) courtesy, or social desirability, bias, in which interviewees provide responses to water treatment questions that they believe interviewers expect, resulting in over-reporting of water treatment (Briere et al. 2012, Luoto et al. 2011).

71 The “real world” problems of turbidity, proper dosing, type of storage container used, time of
72 storage, and reliance on self-reported water treatment complicate the ability of household water
73 chlorination program staff to evaluate: 1) whether water has been treated and 2) the effectiveness of
74 treatment. Simple methods that are feasible for field use are needed to confirm whether, in the absence
75 of detectable FCR, water was chlorinated and whether this treatment improved water quality. To
76 address these problems, we conducted a household-based study in western Kenya in which we analyzed
77 four measures of water quality at five time points in both clay pots, the most commonly used water
78 storage container (ranging from 62-92% of households) (Blanton et al. 2010, Garrett et al. 2008, O'Reilly
79 et al. 2008, Parker et al. 2006), and plastic safe storage containers. In particular, we attempted to
80 determine whether ORP offered advantages over TCR and FCR as confirmatory measures of chlorination,
81 using *E. coli* concentration as the “gold standard” of disinfection effectiveness.
82
83 2. Materials and Methods
84 2.1. Study design
85 To assess changes in water quality over time in a real-world setting and to compare four measures
86 of water quality in two types of water storage containers, we conducted a controlled crossover trial of 2
87 randomly selected groups of households in western Kenya from August 27-October 19, 2012. In one
88 group (Group A), water was chlorinated and stored in clay pots typically used for drinking water storage;
89 in the other group (Group B), water was chlorinated and stored in a plastic safe storage container (Fig.
90 1). Over the following 72 h, water quality tests were performed for both groups. After a two-week
91 washout period, the container types were switched between the groups, and the process described
92 above was performed (Fig. 2).
93
94 2.2. Study population

We selected a convenience sample of six rural villages in Kisumu County that relied on variety of community drinking water sources and household water storage. Households with the following characteristics were eligible to participate: had ≥one child <5 years old; collected and transported drinking water in 10 liter (L) or 20 L containers (jerry cans or buckets); stored drinking water in a ≥15 L ceramic pots (range 15-30 L) in the home; and were willing to use a plastic safe storage container to store drinking water for half of the study period and their own ceramic pot for the other half of the study. Households that did not store drinking water in ceramic pots with ≥15 L capacity were excluded because of the likelihood that stored water would not last for more than one day.

2.3. Enrollment

In each of the 6 study villages, we obtained a list of all households with at least one child <5 years old from the village chief, or conducted a brief census to obtain the list of households. We then used a random numbers table to select a sample of households with children <5 years old in each of the six communities. A total of 60 households were initially enrolled in the study. At the time of enrollment, respondents in households were interviewed about demographic characteristics, and water, sanitation, and hygiene practices. Electronic questionnaires were verbally administered in Dholuo, the local language, by trained Kenyan field research assistants.

2.4. Intervention

The 60 households were randomly allocated to two groups– Groups A (30 households) and B (30 households) (Fig. 2). Group A households were asked to use their clay pots during the first half of the study while Group B households were provided a new, 60-L plastic safe storage container with a lid, tap, and stand.

119 2.5. Phase 1

120 Households were contacted in advance and requested to fill their water collection containers (in
121 most cases, 20-L jerry cans) using water from their usual drinking water source on the morning of the
122 first home visit and to keep it in the transport containers. During the first home visit, investigators
123 collected Time 0 ("pre-dose") water samples by pouring water directly from the transport containers
124 into test vials and sample bottles.

125 To assess water quality, three water quality and treatment measures were performed using
126 portable field meters in the home. Water samples collected into 10-mL glass vials were tested for TCR
127 (mg/L) and FCR (mg/L) (Hach® Pocket Colorimeter™ II, Loveland, CO, USA); water samples collected into
128 50-mL polypropylene conical tubes were tested for ORP (mV) (Oakton® Waterproof ORPTestr® 10,
129 Vernon Hills, IL, USA). Additionally, a 100-mL sample was collected in a WhirlPak™ bag containing
130 sodium thiosulfate, stored on ice, and transported to the laboratory within 4-6 hours of collection for *E.*
131 *coli* quantification (CFU/100 mL) using membrane filtration (0.45 µM, 47 mm filters) with m-ColiBlue24®
132 media (Hach®, Loveland, CO, USA). In some cases, because of exceedingly slow filtration rates of water
133 samples due to high turbidity, we limited the volume of filtrate to 20 or 50 mL of sample and multiplied
134 positive results by the appropriate proportion factor; samples with no growth were reported as non-
135 detectable for *E. coli*.

136 In addition, because physicochemical parameters can influence chlorine residuals and other
137 water quality measures, we also tested samples collected in 50-mL polypropylene conical tubes for the
138 four following physicochemical parameters: turbidity (NTU) (Hach® 2100Q Portable Turbidimeter,
139 Loveland, CO, USA), temperature (°C), electrical conductivity (µs/cm), and total dissolved solids (mg/L)
140 (Oakton® Waterproof Multiparameter PCS Tester 35).

141 In the presence of the head of household, investigators then treated each water transport vessel
142 with the proper dose of WaterGuard™, a familiar, locally available water treatment product containing

1.25% sodium hypochlorite solution. The dose was based on turbidity and the volume of water in the jerry can; water with turbidity <10 NTU was dosed with a single 3 mL dose of WaterGuard per 20 L and water with turbidity >10 NTU was dosed with a double dose of 6 mL of WaterGuard per 20 L. Treated water was then poured into either the household's empty ceramic pot (Group A) or the new plastic safe storage container (Group B).

After 30 min, water samples were collected and tested, as described above. Because the size and weight of clay storage pots precluded pouring water samples, each head of household was asked to wash a cup or ladle that was normally used to obtain water so that water samples could be collected; water samples from the improved plastic storage containers were taken directly from the tap. Heads of households were asked not to retreat the water or refill the container unless it was completely emptied out.

The household was revisited at 24, 48, and 72 h for a short-follow-up interview about water addition or treatment since the previous visit, followed by collection and testing of water samples, as described above. If respondents reported that water or additional disinfectant had been added to the storage container since the previous visit, they were excluded from the remainder of this phase of the study.

2.6. Phase 2 (cross-over)

The crossover period of the project began after a 2-week washout period. Households were again contacted in advance and requested to fill their transport containers using water from their usual drinking water source on the morning of the first home visit and to keep it in the transport containers. Households in Group A were provided with a plastic safe storage container with a lid, tap, and stand; Group B households were asked to resume using their ceramic pots for water storage. Water treatment and testing proceeded in a fashion identical to phase 1. At the conclusion of phase 2, all households

were allowed to keep the plastic safe storage containers, stands, and a bottle of WaterGuard for participation in the study.

2.7. Data analysis

Interview data were entered into personal digital assistants (PDAs) and uploaded into an Access (Microsoft, Redmond, WA, USA) database. Chemical and microbial data were collected on hardcopy forms, entered into an Excel database, and analyzed with SAS® 9.3 software (Cary, NC, USA). TCR, FCR, turbidity, and *E. coli* concentration had skewed distributions and were categorized according to the following metrics. For descriptive purposes, *E. coli* was categorized according to WHO risk thresholds as non-detectable or 1-10, 11-100, or >100 CFU/100 mL (WHO 1997). Since WHO guidelines and public health interventions are aimed at complete removal of *E. coli*, we further categorized data into a dichotomous presence/absence for modeling. FCR was categorized as <0.2 or ≥0.2 mg/L, as this is the minimum recommended concentration by the WHO Guidelines for Drinking Water Quality for infrastructure treated water (WHO 2011). TCR was similarly categorized as <0.2 or ≥0.2 mg/L based on previous research that utilized this strategy to assess chlorine treatment efficacy and storage time in ceramic pots (Null and Lantagne 2012). Water samples were categorized as turbid when turbidity measures were ≥10 NTU, in reference to chlorine dosing recommendation for turbid water. The primary outcomes of interest were TCR, FCR, ORP, and *E. coli*.

To investigate water quality differences in clay pots and plastic safe storage containers across the five time intervals, two-way within-subjects random effects models were constructed; logistic regression models for the outcomes TCR, FCR, and *E. coli* and linear regression for ORP. Interaction terms for storage container and time interval were significant for all four primary outcomes (TCR, FCR, ORP, and *E. coli*). For results stratified by water storage container type, we present estimates from separate repeated measures models for binary outcomes using generalized estimating equations (GEE)

and an autoregressive correlation structure. Odds Ratios (OR) and 95% Confidence Intervals (CI) computed from robust standard error estimates are reported. ORP mean differences are computed from random effects linear regression models and Tukey adjusted p-values are reported. All models adjusted for turbidity.

2.8. Ethical considerations

The study protocol was approved by the Ethical Review Committee of the Kenya Medical Research Institute (protocol 2324) and the Institutional Review Board of the Centers for Disease Control and Prevention (protocol 6313). Written informed consent was obtained from all participants. Data were maintained in an encrypted file in a password-protected computer. Personal identifiers were destroyed after all data were collected.

3. Results

3.1. Demographic characteristics and baseline water, sanitation, and hygiene practices

A total of 60 respondents were enrolled in the study. Five households were excluded from the study because respondents weren't available for one or more of the intervention phases; ultimately 25 respondents remained in Group A and 30 respondents comprised Group B. The median age across participating respondents was 27 (range 17-55) and all were women. Fewer than half (n=23) had less than a complete primary school education and only one, in Group B, had electricity. The majority (85%) of study households used improved water sources and 60% of respondents reported that they treated water stored in their homes. Of 32 households that reported treating their water, 24 (75%) reported using WaterGuard; 2 (6%) reporting using other chlorine-based products, 5 (16%) reported boiling, and 12 (38%) reported using a cloth to filter water. Fewer than half (47%) of households had an improved

214 sanitation facility. Soap was present in 93% of households and 56% of respondents were able to
215 demonstrate proper handwashing technique.
216
217 3.2. Water testing: clay pots
218 Water sources used for dosing experiments in clay pots included rain (40%), surface water
219 (24%), springs (18%), piped networks (16%), and ground water (2%). The median turbidity of water
220 samples in clay pots at Time 0 was 37 NTU (range 0-300 NTU) (Table 1). Turbidity measures did not vary
221 widely over the 72 h testing period. Median TCR and FCR values at Time 0 were 0.1 mg/L; over three-
222 fourths of samples were <0.2 mg/L for both TCR and FCR. The median ORP was 393 mV (range 196-597
223 mV). At Time 0, 83% of water samples were contaminated with *E. coli*. Water had a median pH 6.8, 25°C
224 temperature, electrical conductivity of 106 µs/cm, and 78 mg/L total dissolved solids; these median
225 measures did not vary widely over the 72 h testing period.
226 Thirty minutes after chlorination (Time 0.5 hr), median TCR and FCR levels increased to 1.2 and
227 0.9 mg/L, respectively (Table 1). Median ORP increased to 541 mV (range 392-757 mV), with 93% of
228 samples increasing by >10% of the time 0 value. *E. coli* were non-detectable in 83% of samples. By 24 h,
229 FCR was <0.2 mg/L in 61% of samples and TCR was <0.2 mg/L in 31% of samples. Approximately 40% of
230 samples had ORP values 10% of the time 0 value. *E. coli* were non-detectable in 74% of samples. At 48 h,
231 51% of TCR and 67% of FCR values were <0.2 mg/L and the median ORP measurement decreased to
232 slightly lower than the time 0 value. The percentage of samples with non-detectable *E. coli* decreased to
233 48%. By 72 h, median TCR was 0.2 mg/L and FCR was 0.1 mg/L; 35% of samples had no detectable *E. coli*.
234 Compared to Time 0 values and adjusted for turbidity, water treated with the recommended
235 amount of chlorine and stored in clay pots was significantly less likely to contain *E. coli* for up to 48 h
236 (Table 2). Although FCR levels were significantly more likely to be >0.2 mg/L at 30 min than at Time 0, by
237 24 h FCR was significantly more likely to have fallen below the threshold of 0.2 mg/L. However, TCR

levels were less likely to have fallen below the 0.2 mg/L threshold over the entire 72 h time period than at baseline. At 30 min and 24 h post treatment, ORP was significantly higher than at Time 0. By 48 h, ORP values were not significantly different than at Time 0.

3.3. Water testing: plastic safe storage containers

Source water used for dosing experiments in plastic safe storage containers include rain (44%), surface water (22%), springs (18%), piped networks (13%), and ground water (4%). The median turbidity of water samples in plastic containers at time 0 was 28 NTU (range 0-300 NTU) (Table 3). Turbidity measures did not vary widely over the 72 h testing period. At Time 0, median TCR and FCR were 0.1 mg/L and <0.1 mg/L, respectively, with over three-fourths of samples <0.2 mg/L for both TCR and FCR. The median ORP was 387 mV (range 252-556 mV). At Time 0, 87% of water samples were contaminated with *E. coli*. Water had a median pH 7.2, 24°C temperature, electrical conductivity of 104 µs/cm, and 69 mg/L total dissolved solids; these median measures did not vary widely over the 72 h testing period.

Thirty minutes after chlorination, median TCR and FCR levels increased to 1.3 and 0.8 mg/L, respectively (Table 3). Median ORP increased to 541 mV (range 392-747 mV), with 89% of samples increasing by >10% of the Time 0 value. *E. coli* were non-detectable in 91% of samples. By 24 h, median FCR decreased to 0.3 mg/L, median TCR was 0.7 mg/L, and 15% of water samples had fallen to within 10% of the time 0 ORP value. *E. coli* remained non-detectable in 85% of samples. At 48 h, 17% of TCR and 33% of FCR values were <0.2 mg/L and the median ORP measurement remained higher than the Time 0 value. *E. coli* were non-detectable in 90% of samples. By 72 h, median TCR was 0.6 mg/L and FCR was 0.3 mg/L, the median ORP value was higher than at Time 0, and 80% of samples had no detectable *E. coli*.

When compared with Time 0 values and adjusted for turbidity, water treated with the recommended amount of chlorine and stored in a plastic safe storage containers was significantly less

likely to contain *E. coli* across all time points, indicating a protective effect for up to 72 h (Table 4). Both FCR and TCR levels were significantly less likely to be <0.2 mg/L than the Time 0 values over the entire 72 h time period. Up through 24 h, mean ORP was significantly higher than Time 0 ORP values, however, by 48 h ORP values were not significantly different than Time 0 values.

3.4. Comparison of clay pots and plastic safe storage containers

Using two-way random effects models and adjusting for turbidity, we assessed differences in water storage containers and time points for TCR, FCR, and *E. coli*. Water container type was a statistically significant effect modifier for time interval, thus we present results stratified by either water container or time interval. There were statistically significant differences between clay pots and plastic safe storage containers for TCR, FCR, and *E. coli* at 48 and 72 h. Despite no differences in water quality measures between storage containers at pre-treatment, 30 mins and 24 h, the odds of having a positive *E. coli* result were greater in clay pots compared to plastic containers at 48 (p=0.0002) and 72 h (p=0.0004). The odds of having TCR <0.2 mg/L were significantly greater in clay pots than plastic containers at 24 (p=0.0199), 48 (p=0.0023), and 72 h (p=0.0061); likewise, the odds of having FCR <0.2 mg/L were significantly greater in clay pots than plastic containers at 24 (p=0.0370), 48 (p=0.0014), and 72 h (p=0.0245) (Table 5).

If TCR or FCR was ≥0.2 mg/L in stored water, regardless of container type or time, there was a decreased likelihood that *E. coli* was present. This association was stronger for TCR ≥0.2 in plastic containers (OR 0.08, 95% confidence interval [CI] 0.04-0.16) than in clay pots (OR 0.44, 95% CI 0.27-0.75); likewise, this association was stronger for FCR ≥0.2 in plastic containers (OR 0.25, 95% CI 0.14-0.44) than in clay pots (OR 0.43, 95% CI 0.26-0.69).

4. Discussion

To our knowledge, this is the first study in which a controlled chlorination experiment at the household level tested four water quality measures, including ORP, for a period of up to 72 hours. This evaluation yielded several key findings. First, ORP served as a reasonable proxy for chlorination in plastic containers up to 24 hours, but was not a good proxy after 24 hours as ORP decreased to near pre-treatment levels. ORP was also not a good proxy in clay pots because the level was not significantly different at 24 hours than pre-treatment. The ease of ORP measurement using a probe and without a need for reagents offers the advantage of convenience, while the main disadvantage is the initial investment in the ORP meter. Second, chlorinating various types of source waters at recommended doses resulted in a statistically significant increase in the percentage of stored water samples that had no detectable *E. coli* for up to 72 hours, even as FCR fell below the recommended minimum concentration of 0.2 mg/L and ORP decreased to pre-treatment levels. Third, as expected, TCR persisted above 0.2 mg/L over a longer period than FCR. There was a statistically significant association between TCR values ≥0.2 mg/L and non-detectable *E. coli* in stored water, which presents the possibility of TCR serving as a useful proxy measure of water quality.

This evaluation also demonstrated that chlorination at the recommended dose was more effective at eliminating detectable *E. coli* for up to 72 hours in plastic safe storage containers than in traditional clay pots, even when adjusting for source water turbidity. This finding most likely occurred because FCR was significantly more likely to persist at higher concentrations over time in plastic versus clay containers. These findings are expected, consistent with other studies (Ogutu et al. 2001, Quick et al. 1996), and plausible because clay pots often have organic materials on the surface that exert chlorine demand and facilitate biofilm growth (Murphy et al. 2009). In addition, clay pots have wide mouths, which permit the insertion of hands or other objects potentially increasing chlorine demand and the risk of recontamination. In this study, by testing water stored in clay pots that had been in use in households rather than new clay pots, chlorine demand in the pots may have been greater and likely to decrease

FCR levels at a faster rate than in new pots, thereby possibly biasing results toward the null. However, at least one study has shown no difference in chlorine behavior between new and used clay pots (Ogutu et al. 2001). In addition, the evaluation of water storage in used clay pots more accurately represents actual household circumstances. One caveat to this finding is that we used new plastic safe storage containers that initially would have been free of biofilm, so their performance might decline over longer periods of use as biofilm formed on the inner surface (UNICEF and WHO 2015, Arnold and Colford 2007, Jagals et al. 2003). Further study is needed to evaluate this possibility.

ORP proved to be a poor proxy of drinking water disinfection after 24 hours because, although ORP is increasingly used to monitor disinfection capacity of water in distribution systems and swimming pools, a higher concentration of chlorine is often used in those systems (i.e., FCR 1-3 mg/L) than in stored drinking water, resulting in higher post-treatment OPR values. When treating water for human consumption, palatability is an important consideration, and chlorine concentrations that would results in elevated ORP for greater than 24 hours, such as those used in treatment facilities or swimming pools, would be unpalatable in drinking water stored in household containers. For ORP to meet its potential as a field measurement of effective household water treatment over periods <24 hours guidelines would need to be developed for interpretation of measures.

The practical importance of the above findings can be appreciated when considering other studies of chlorination in which reported rates of chlorine use were high but measured FCR in water samples were low (Colindres et al. 2007, Lantagne and Clasen 2012, Harshfield et al. 2012, Mong et al. 2001). In those studies, it was not possible to determine whether the high reported rates were a result of social desirability or courtesy bias in which water treatment was not actually performed, or a result of a poor indicator (i.e., FCR) for turbid water treated with hypochlorite, for water treated with hypochlorite >24 hours before testing, or both. Findings of this study suggest that, because TCR persists longer than FCR in stored water, it may serve as a better proxy measure for adherence to recommended

334 treatment with sodium hypochlorite. Additionally, the statistically significant association between TCR
335 ≥0.2 mg/L and non-detectable *E. coli* in stored water suggests that TCR may also serve as a rough,
336 though imperfect, proxy measure for water quality. While not completely free of *E. coli* contamination,
337 water remained improved up to 72 hours as compared with its pre-dose quality. Recent research found
338 a positive association between the risk of child diarrhea and increasing *E. coli* concentration in drinking
339 water; the dose-response relationship observed suggested that even modest improvements in water
340 quality can provide a health benefit (Luby et al. 2015). However, TCR would be a less reliable proxy
341 measure of water quality in populations that use clay pots for water storage, particularly if the water
342 were stored over a period of several days before being replenished. Populations that prefer clay pots
343 because of evaporative cooling of stored water would likely be difficult to motivate to switch to plastic
344 water storage containers. In this case, chlorination promotion campaigns would need to take into
345 account the properties of clay pots, particularly those with wide mouths that permit the introduction of
346 hands or other objects, and recommend daily treatment of stored water.
347 This study had several important limitations. First, we cannot be certain that households did not
348 chlorinate water before our first visit or between visits over the 72 hour study period, even though we
349 requested that they not do so. If water had been chlorinated between visits, or non-chlorinated water
350 had been added to containers, then our data would not provide an accurate representation of the
351 behavior of chlorine, ORP, or *E. coli* over time. The steady decrease of TCR and FCR that we observed
352 over time during both study periods suggest that the population adhered to our request. Second, TCR
353 and FCR were detected in some source waters (primarily surface, rain, and spring water); this finding
354 might be related to false positive results related to DPD interference from chemicals present in water
355 and warrants further research. Third, during both study periods, there was attrition in the number of
356 households at each visit as participants used up the water that had been placed in their containers
357 before Time 0 (pre-dosing), which decreased the precision of our findings. Fourth, because clay pots are

cumbersome and heavy, we were not able to directly sample stored water but instead relied on the use of a ladle or cup. While we observed household members washing these collection vessels before sampling, we cannot be certain of the effect they had on water quality. Finally, this study was conducted in a limited geographical area and is not representative of the larger Kenyan population, or other populations. Although the findings were consistent with known behavior of residual chlorine in stored water and *E. coli* exposed to chorine, further study in other populations would help determine how broadly applicable our findings are.

5. Conclusions

- ORP may be a useful proxy to confirm chlorination for periods up to 24 hours in plastic containers, but further study is needed to verify its utility.
- Most stored water samples disinfected by chlorination remained significantly less contaminated than source water for up to 72 hours, even in the absence of FCR.
- TCR may be a useful proxy indicator of microbiologic water quality because it confirms previous chlorination and is associated with a lower risk of *E. coli* contamination compared to untreated source water.
- Chlorination is more effective in plastic than clay containers presumably because of lower chlorine demand in plastic.

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