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Effect of handwashing on the reduction of *Escherichia coli* on children's hands in an urban slum Indonesia

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ABSTRACT

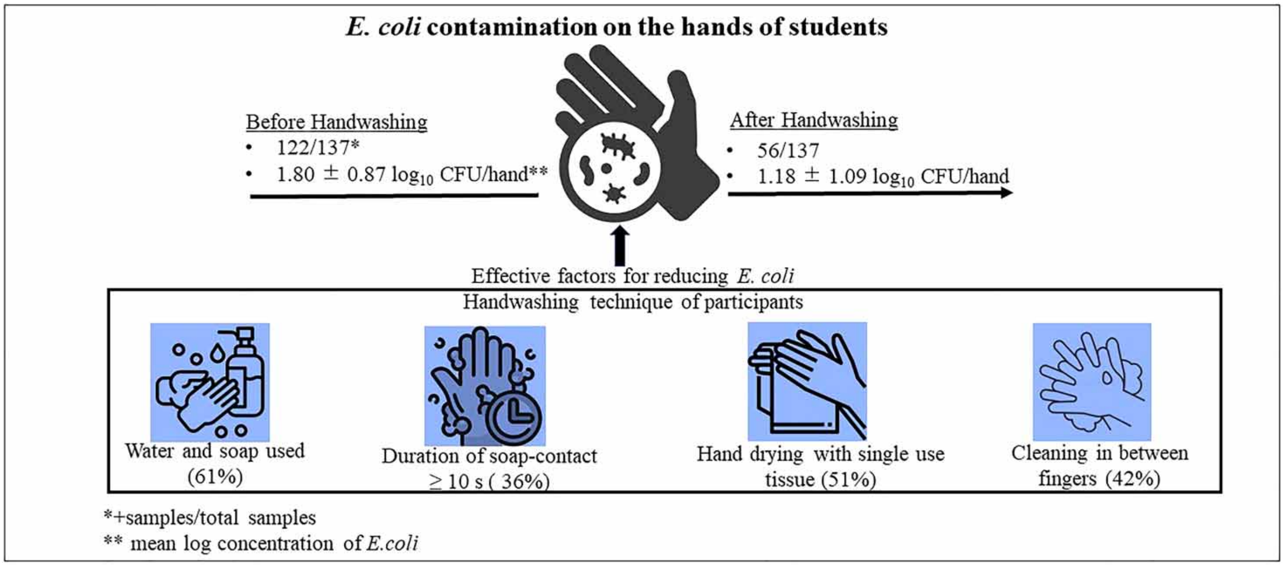
Poor hand hygiene practice has been linked to an increase in the number of infections among children in urban slums. Hands are considered an intersection for bacterial transmission, but it is unclear whether the handwashing technique affects bacteria elimination. This study investigated the effect of handwashing on the concentration of *Escherichia coli* (*E. coli*) and factors related to its reduction among children in an urban slum in Bandung, Indonesia. We observed handwashing and conducted repeated hand swabs before and after handwashing among 137 participants. The mean *E. coli* concentration on the hands decreased after handwashing, with a higher reduction in *E. coli* count among students who used soap and had soap contact for more than 10 s during handwashing. Cleaning in-between fingers, using soap, soap contact for more than 10 s, and drying hands with a single-use towel were effective factors for reducing *E. coli* concentration after handwashing ($p < 0.05$). More than half of the swab samples (59%) tested positive for *E. coli* after handwashing, indicating that the children's handwashing technique was not effective in completely removing *E. coli* from the hands. Moreover, sustained and consistent handwashing practice as a daily behavior in children would maximize the effect.

Key words: child, *E. coli*, hand hygiene, handwashing technique, urban slum

HIGHLIGHTS

- Fecal contamination was detected on the hands of students before and after handwashing.
- The children's handwashing practice falls short of the standard, which affects the level of reduction of fecal contamination.
- The short duration of soap contact with the hand (<10 s) is not enough to reduce the number of *E. coli* on the hands.
- The results imply the need to focus on specific critical points in handwashing education.

GRAPHICAL ABSTRACT



INTRODUCTION

Diarrhea is a leading cause of mortality in children worldwide, with an estimated 1,438 deaths per day (Levine *et al.* 2020). Pathogens, including *E. coli*, are often associated with diarrhea, and hands can serve as a vehicle for pathogen transmission through the fecal–oral route (Abba *et al.* 2009). Handwashing with soap (HWWS) and water has been shown to eliminate bacteria from the hands (Curtis *et al.* 2005; Burton *et al.* 2011) and reduce the risk of diarrhea by 40% (Freeman *et al.* 2014). However, the impact of hand hygiene on reducing fecal contamination remains unclear, particularly in resource-limited settings (Saboori *et al.* 2013; Aihara *et al.* 2014).

Proper handwashing remains a challenge for children in low- and middle-income countries (LMICs), and young children often demonstrate poor handwashing practice (Xuan & Hoat 2011). Several studies have found that a common problem in urban slum communities is the lack of access to soap and clean water, which can affect handwashing technique and bacterial count (Pickering *et al*. 2011; Paraduth & Biranjia-Hurdoyal 2015). Some experimental trials (laboratory and field studies) confirmed this by manipulating some steps (use of soap, length of time, or hand-drying method) to measure the effect of various methods on bacteria elimination (Burton *et al.* 2011; Kim *et al.* 2019; Gizaw *et al.* 2022). However, these studies did not reflect the natural, daily handwashing behavior of participants in their community. In addition, some of the studies were limited to clinical settings (Seid *et al.* 2022), and comprehensive assessments of the slum community are scarce.

In urban slum settings, child hygiene is often compromised (Kundu *et al.* 2018). Our recent study in Indonesia found that young children in urban slums had poor handwashing techniques, which was significantly associated with *E. coli* detection on their hands. Similarly, awareness and knowledge of HWWS was inadequate (Otsuka *et al.* 2019). Previous studies in Indonesia have primarily focused on public perception, behavioral intervention, and determinants of handwashing among people (Hirai *et al.* 2016; Karon *et al.* 2017; Dwipayanti *et al.* 2021). However, no study has examined the effect of the handwashing technique on reducing *E. coli* on children's hands in this setting, particularly during the COVID-19 pandemic.

Therefore, this observational study investigated the effect of the handwashing technique on the reduction of *E. coli* on children's hands in urban slums. Our study focuses on analyzing which steps are practicable within the community's target population. We also assessed the relationship between *E. coli* reduction and several key handwashing variables, including duration of soap and water contact, handwashing steps, water used, and hand-drying methods.

MATERIALS AND METHODS

Study areas and participants

This cross-sectional study was conducted among preschool and primary school students in Kiaracondong, an urban slum in Bandung, Indonesia, between August and September 2022. Bandung is the third most populous city, experiencing rapid urbanization and overcrowding, particularly in the slums. Bandung's slum settlements still suffer from poor sanitation and water access (Tarigan *et al.* 2015). Kiaracondong is one of Bandung's most populous urban slum communities, with a high population density (22,426/km^2) and poor water and sanitation infrastructure (BPS 2022). After explaining the purpose and content of the study, 137 students provided written consent and participated. We included students from preschool, grades 2, 4, and 6 with a mean age of 8.9 $\pm$ 2.4 years old.

Data collection

Handwashing technique

The participants were asked to perform their daily handwashing practice individually in a detached space, while an observer observed the entire handwashing process. They were provided with all the needed materials, such as tap water, soap, tissue paper, and hand towels. Schools and households in developing countries (Kim *et al.* 2019), including Indonesia, provide hand towels as a common practice. The handwashing process was video recorded using two cameras (front and side views), and the evaluation was based on both recorded video and direct observation. A single observer determined the process and was supported by one research assistant who ensured the recording, use of water, and management of each student's turn. Five main points were assessed in this process: (1) handwashing steps (10 steps) (Otsuka *et al.* 2019), (2) the use of soap, (3) the volume of water, (4) the length of time (total duration, water contact, and soap contact), and (5) the hand-drying method. The volume of water used for handwashing was measured using a modified jerrycan equipped with a volume scale, while the length of time was measured using stopwatch and video observation (see Supplementary file: Appendix A).

Hand swabs and microbial test

The hand swab test was used to determine the level of *E. coli* on the children's hands before and after handwashing. The researcher performed the swab test using a swab test kit (Pro Media ST-25 PBS; Series: 64-8070-68, ELMEX, Japan) containing a wiping swab in 10 mL of phosphate buffer saline (PBS). Pre-test with four samples was performed before the school assessment. To minimize bias due to tap water contamination during handwashing, we have already tested the water, and the *E. coli* results were negative. We also implemented a cleaning protocol by using alcohol-based wet wipes to clean tap handles and soap bottles, thus preventing contamination for each child. To collect 'hand before washing' samples, a student's left or right hand was randomly selected. From 137 participants, the right hand was sampled first in 74 students, and the left hand was sampled first in 63 students. Then, students were asked to spread selected hands, and the wiping swab was rolled on the palm, backside, fingers, and in-between fingers. After that, students were requested to wash hands in the usual way. Immediately after handwashing, the opposite hand (hand that had not yet been sampled) was swabbed using the same method. The same hand was not sampled twice because the hand sampling method used could be considered similar to handwashing, which would remove *E. coli* and introduce bias into our objective of evaluating the effect of handwashing techniques on *E. coli* count after handwashing (see Supplementary file: Appendix B). After wiping, the swab was kept in PBS of the kit and delivered to the laboratory within 4 h for processing. There are some bacteria that serve as indicators to detect the presence of fecal contamination on hands. However, in this study, we focused on *E. coli* as a single indicator, as this research is a follow-up to our previous study in the same area (Otsuka *et al.* 2019). The enzyme substrate method was used to detect *E. coli*. All bacteria analysis was performed under aseptic conditions in the biosafety cabinet of a microbiology laboratory. Each sample (10 mL) was divided into two volumes (1 and 4 mL), and passed through a 47-mm-diameter 0.45-μm sterilized cellulose filter (Advantec, Tokyo) using a membrane filtration unit (vacuum pump). Approximately 10 mL of sterilized PBS buffer was added to facilitate uniform dispersion over the filter surface. Thereafter, the cellulose filter was placed on XM-G Agar growth media (XM-G; Nissui Pharmaceutical Co, Japan). The sample was incubated on the media for 20 $\pm$ 2 h at 37 °C. After incubation, the colony had turned blue and was counted. Blue colony indicates the presence of *E. coli* in the sample.

Data analysis

The results were analyzed using JMP SAS version 16 (SAS Institute, Japan) for Microsoft Windows 10. For statistical analysis, the number of colonies was measured as a colony-forming unit (CFU) and transformed to $\log_{10}$ per hand before and after handwashing. The matched pair sample *t*-test ($p < 0.05$) was used to compare the mean $\log_{10}$ concentration of *E. coli* before and after handwashing. As the maximum reliable count of *E. coli* on the filter was 300, the upper detection limit was set at 3,000 CFU/hand. For the categories of handwashing techniques, we used several key variables, such as duration of soap and water contact, handwashing steps, water used, and hand-drying methods (Aihara *et al.* 2014; Friedrich *et al.* 2017; Seid *et al.* 2022). As each parameter performed might influence the effect, multivariate stepwise logistic regression was used to investigate the association and interconnection between handwashing method/steps and its effect on *E. coli* reduction. The reduction of *E. coli* was categorized into two groups: reduction and no reduction before and after handwashing. The 'no reduction' criterion indicates that the *E. coli* concentration remained the same or even increased after handwashing. Eligible dependent factors for *E. coli* reduction were handwashing steps 2, 4, 9, and 10, duration of soap contact and water contact, and hand-drying. These factors were computed using the stepwise forward method to determine those that were significantly associated with *E. coli* reduction. The *p*-value threshold for entry and removal from the model was 0.25 and 0.1, respectively.

RESULTS

E. coli on the hands and handwashing

The 137 preschool and primary school students who participated in the study were aged 4–12 years and were in grades 2, 4, and 6. More than half of the participants (52%) were females. To investigate fecal contamination, we evaluated the *E. coli* count in 137 paired pre- and post-handwash participants. *E. coli* was detected in 122 (89%) samples before handwashing, which decreased to 81 (59%) after handwashing (Table 1). Except for preschoolers, there was a significant difference in the mean concentration of *E. coli* by grade and gender, before and after handwashing ($p < 0.05$). Second graders had the highest (1.69 ± 1.01 $\log_{10}$ CFU/hand), followed by preschoolers (1.51 ± 1.08 $\log_{10}$ CFU/hand). Overall, handwashing decreased *the E. coli* count by 34%, with the lowest concentration found in the highest grade (grade 4). There was also a reduction in the number of colonies of *E. coli*, with most of them having *E. coli* concentrations below 100 CFU/hand. Interestingly, in some students, we also observed that *E. coli* levels remained the same or even increased after handwashing. These findings support our hypothesis that children's handwashing technique influences the presence and concentration of *E. coli* after handwashing.

Handwashing techniques and *E. coli* reduction

We measured the mean *E. coli* concentration before and after handwashing by the method practiced (Table 2). Table 2 displays the $\log_{10}$ CFU of *E. coli* before and after handwashing, categorized according to the techniques used by the participants. The average time spent by participants for the entire handwashing process was 18.5 ± 9.3 s. A greater reduction in *E. coli* was found when using water and soap (handwashing duration ≥ 20 s), soap contact (soap contact during handwashing duration $\geq$ 10 s), 4–5 steps, $\geq$500 mL amount of water, and single-use tissue for the hand-drying method. Handwashing with $\leq$3 steps did not significantly influence *E. coli* reduction. Although we provided tissue paper, some students used hand towels to dry their hands, resulting in no significant reduction after handwashing. Table 2 also shows the correlation between handwashing technique and *E. coli* reduction. The reduction of *E. coli* on children's hands was significantly associated with soap use, duration of soap contact, number of steps, and hand-drying method ($p < 0.05$).

Handwashing step and *E. coli* reduction

Table 3 shows the association between handwashing step and *E. coli* reduction. More than 90% of the participants wet their hands and rubbed them palm to palm and about two-thirds used soap during handwashing. Nevertheless, less than 25% completed scrubbing of the back of their fingers, thumbs, and fingertips, indicating poor knowledge of this aspect of handwashing. Soap use during handwashing significantly reduced *E. coli* concentration on children's hands, compared to water only ($p < 0.05$). Among those who scrubbed some parts of the hand, cleaning in-between fingers had a significant correlation with *E. coli* after handwashing. Although some students skipped the final two steps in handwashing, we found a significant correlation between rinsing with water and drying hands with a towel or tissue paper and *E. coli* reduction.

Factor associated with *E. coli* reduction

The results of the regression analysis model predicting the handwashing technique and *E. coli* reduction are presented in Table 4. This regression model represents the function of the handwashing technique performed and reduction of *E. coli*, which might be influenced by other techniques or steps. The odds of *E. coli* reduction were three times and four times higher when handwashing included the use of soap and water (AOR: 2.81, 95% CI: 1.62–11.8) and fingers interlaced cleaning

Table 1 | Overall mean and number of samples (%) in each category of *E. coli* pre- and post-handwashing ($n = 137$)

Category	Sample number tested (n)	Pre-handwashing						Post-handwashing						
					E. coli concentration (CFU/hand)*						*E. coli* concentration (CFU/hand)*			
		Positive No. (%)	ND (%)	Mean $\log_{10}$ concentration (SD)	$0 < x < 100$	$100 < x < 1,000$	$1,000 < x < 3,000$	Positive No. (%)	ND (%)	Mean $\log_{10}$ concentration (SD)	$0 < x < 100$	$100 < x < 1,000$	$1,000 < x < 3,000$	p-value
Grade														
Preschool	25 (18)	19 (76)	6 (24)	1.64 (1.05)	5 (20)	13 (52)	1 (4)	18 (72)	7 (28)	1.51 (1.08)	8 (32)	8 (32)	2 (8)	NS
Grade 2	37 (27)	37 (100)	0 (0)	2.34 (0.58)	10 (27)	22 (59)	5 (14)	30 (81)	7 (19)	1.69 (1.01)	11 (30)	17 (46)	2 (5)	<0.05
Grade 4	37 (27)	31 (84)	6 (16)	1.51 (0.82)	19 (51)	12 (32)	0 (0)	14 (38)	23 (62)	0.80 (1.12)	4 (11)	9 (24)	1 (3)	<0.05
Grade 6	38 (28)	35 (92)	3 (8)	1.67 (0.82)	22 (58)	10 (26)	3 (8)	19 (50)	19 (50)	0.83 (0.91)	14 (37)	5 (13)	0 (0)	<0.05
Gender														
Male	66 (48)	58 (88)	7 (11)	1.81 (0.83)	30 (45)	31 (47)	3 (5)	39 (59)	29 (41)	1.51 (1.08)	20 (30)	19 (29)	3 (5)	<0.05
Female	71 (52)	64 (90)	8 (11)	1.79 (0.92)	26 (37)	26 (37)	6 (8)	42 (59)	27 (41)	1.21 (1.11)	17 (24)	20 (28)	2 (3)	<0.05
Total	137	122 (89)	15 (11)	1.80 (0.87)	56 (41)	57 (42)	9 (7)	81 (59)	56 (41)	1.18 (1.09)	37 (27)	39 (28)	5 (4)	<0.05

ND, not detected, SD, standard deviation.

*Mean log10 concn (SD) represented the concentration of *E. coli* by each category. Differences between pre-post handwashing were analyzed using matched pairs t-test P<0.05.

Table 2 | Difference concentration of *E. coli* pre- and post-handwashing by the technique, and relationship with the reduction of *E. coli* ($n = 137$)

No.	Method of handwashing	Total $n = 137$	Mean $\log_{10}$ concentration (SD)*		Mean $\log_{10}$ Δ pre–post-handwashing	*p*-value	Reduction of *E. coli***		*p*-value
			Pre-handwashing	Post-handwashing			Yes	No	
1	Water and soap used								
	Water only	54 (39)	1.77 (0.99)	1.38 (1.17)	0.39	<0.05	26 (19)	28 (20)	< 0.05
	Water and soap	83 (61)	1.82 (0.78)	1.05 (1.02)	0.77	<0.05	67 (49)	16 (12)	
2	Total duration of handwashing								
	<20 s	81 (59)	1.89 (0.87)	1.39 (1.11)	0.49	<0.05	50 (37)	31 (23)	NS
	≥20 s	56 (41)	1.67 (0.87)	0.87 (0.99)	0.80	<0.05	43 (31)	13 (10)	
3	Duration of soap contact								
	<10 s	88 (64)	1.81 (0.96)	1.36 (1.12)	0.44	<0.05	50 (37)	38 (28)	< 0.05
	≥10 s	49 (36)	1.80 (0.69)	0.85 (0.98)	0.94	<0.05	43 (31)	6 (4)	
4	Duration of water contact								
	<10 s	106 (77)	1.87 (0.90)	1.25 (1.11)	0.61	<0.05	69 (50)	37 (27)	NS
	≥10 s	31 (23)	1.58 (0.74)	0.92 (0.99)	0.65	<0.05	24 (18)	7 (5)	
5	Number of steps								
	≤3 steps	34 (25)	1.63 (1.03)	1.49 (1.08)	0.15	NS	14 (10)	20 (15)	< 0.05
	4–5 steps	46 (34)	1.87 (0.81)	0.97 (1.11)	0.90	<0.05	36 (26)	10 (7)	
	6–8 steps	57 (42)	1.85 (0.81)	1.13 (1.03)	0.71	<0.05	44 (32)	13 (10)	
6	Amount of water used								
	<500 mL	58 (42)	1.98 (0.80)	1.48 (1.09)	0.49	<0.05	36 (26)	22 (16)	NS
	≥500 mL	79 (58)	1.67 (0.90)	0.95 (1.04)	0.71	<0.05	57 (42)	22 (16)	
7	Hand-drying method								
	Single-use tissue	70 (51)	1.91 (0.83)	1.14 (1.08)	0.76	<0.05	56 (41)	14 (10)	< 0.05
	Uniform/clothes	63 (46)	1.67 (0.92)	1.20 (1.11)	0.47	<0.05	36 (26)	27 (20)	
	Cloth towel	4 (3)	2.06 (0.67)	1.56 (1.13)	0.50	NS	1 (1)	3 (2)	

*Mean $\log_{10}$ concentration (SD) of *E. coli* represented the concentration of *E. coli* by each category. Differences between pre- and post-handwashing were analyzed by the matched pair sample *t*-test.

**Correlation between the method of handwashing practiced and the reduction of *E. coli* was analyzed by the Chi-square test.

(AOR: 4.45, 95% CI: 1.54–12.8), respectively. Additionally, scrubbing the hand with soap for more than 10 s was five times (AOR: 5.07, 95% CI: 1.34–19.1) more likely to reduce *E. coli*. Using multi-use towels or clothes for hand-drying had a protective effect in reducing *E. coli* (AOR: 0.03, 95% CI: 0.01–0.47).

DISCUSSION

A high level of fecal contamination on hands has been observed in an urban slum, which has been linked to poor handwashing technique (Otsuka *et al.* 2019). Contamination may come from the household and environment, such as physical contact with contaminated water or materials (Daneshmand *et al.* 2018). Although several studies have investigated *E. coli* in children's hands (Aihara *et al.* 2014; Paraduth & Biranjia-Hurdoyal 2015; Ogba *et al.* 2018), limited research has documented this during the COVID-19 pandemic. The pandemic may have influenced handwashing behavior, as people increased their frequency of handwashing to prevent the spread of the infection (Dwipayanti *et al.* 2021). Frequent handwashing during the pandemic was associated with a reduction in infections (Seid *et al.* 2022). Our study found that 89% of the children had *E. coli* on their hands. This was lower than the 98.7% previously documented in the same population (Otsuka *et al.* 2019). However, the level of contamination in this study was higher than in previous studies. For example, a systematic review revealed that the mean *E. coli* prevalence on hands in LMICs was 69% (Cantrell *et al.* 2023). Although different

Table 3 | Relationship between handwashing step practiced and reduction of *E. coli* ($n = 137$)

Handwashing step	Total ($n = 137$) n (%)	Reduction of *E. coli* Yes n (%)	No n (%)	*p*-value*
Wet hands with water	136 (99)	92 (68)	44 (32)	0.378
Apply enough soap to cover all hand surfaces	83 (61)	67 (49)	16 (12)	**0.001**
Rub hands palm to palm	129 (94)	86 (63)	43 (31)	0.185
Right palm over left dorsum with interlaced fingers and vice versa	57 (42)	47 (34)	10 (7)	**0.002**
Palm to palm with fingers interlaced	59 (43)	41 (69)	18 (13)	0.726
Backs of fingers to opposing palms with fingers interlaced	2 (0)	1 (0.7)	1 (0.7)	0.597
Rotational rubbing of left thumb clasped in right palm and vice versa	28 (20)	20 (15)	8 (6)	0.689
Rotational rubbing, backwards and forwards with clasped fingers of right hand in left palm and vice versa	28 (20)	21 (15)	7 (5)	0.357
Rinse hands with water	92 (67)	72 (53)	20 (15)	**0.001**
Dry hands thoroughly with a towel/tissue	74 (54)	57 (42)	17 (12)	**0.013**
Interaction of hand-rubbing				
Palm × between fingers × fingertips	18 (13)	13 (10)	5 (3)	0.672
Back × between fingers × fingertips	21 (15)	17 (12)	4 (3)	0.163
Palm × Back × between fingers × fingertips	13 (10)	10 (8)	3 (2)	0.463

*Correlation between handwashing step practiced and reduction of *E. coli* was analyzed by the Chi-square test, $p < 0.05$.
Bold values represent the significant variables ($p < 0.05$).

measurements and counting methods were used in a previous study (Cantrell *et al.* 2023), the findings from our present study indicate that there is still a high risk of contamination among young children in urban slums in Indonesia.

The concentration of *E. coli* on hands has been measured as 0.6–3.5 $\log_{10}$ CFU/hand (Pickering *et al.* 2011; Friedrich *et al.* 2017; Kundu *et al.* 2018). This is consistent with our study which found that the average *E. coli* concentration by grade was 1.5–2.3 $\log_{10}$ CFU/hand, with higher contamination observed among participants in the lower grades. However, a previous study conducted in an urban slum setting in India found a lower concentration of 0.64 $\log_{10}$ CFU/per two hands in children under 5 years (Kundu *et al.* 2018). This higher concentration of *E. coli* contamination observed in our study could be attributed to some children spending more time engaging in outdoor activities, which exposes them to more germs and dirt. Some children in this community were involved in garbage sorting to help their parents who worked as garbage collectors (Sai *et al.* 2020).

Except for preschoolers, we found a statistically significant reduction in bacteria after handwashing. Lower graders performed fewer handwashing steps, which could explain the low effectiveness in bacteria reduction. Furthermore, performing three or fewer steps did not result in significant *E. coli* reduction when compared to four steps or more. We found that in some students, the *E. coli* concentration remained the same or even increased after handwashing. Similar observations have been made by other authors in which young children had limited knowledge of HWWS (Xuan & Hoat 2011) and performed handwashing with a low volume of water and without soap, which could have limited the reduction in *E. coli* on their hands after handwashing (Aihara *et al.* 2014; Agestika *et al.* 2019). This underscores the importance of the age of children as an essential factor in bacteria reduction of handwashing techniques.

Despite the availability of soap, 39% of the students did not use soap during handwashing. A similar trend was found among children in LMICs, with estimates showing that only 27–39% of children wash their hands with soap (Saboori *et al.* 2013; Otsuka *et al.* 2019). A possible explanation for this is the lack of effectiveness of handwashing promotion or prevalent social practices at home. In urban slum communities, some families cannot provide handwashing soap because it is an additional cost. Moreover, some respondents describe using soap as challenging because it is a new practice, and they believe washing with water is enough to make hands physically clean. Experimental trials have shown that HWWS is more effective in removing bacteria than using water alone (Burton *et al.* 2011; Amin *et al.* 2014). Similarly, our findings showed that the use of soap was twice as effective as using water alone in eliminating bacteria. HWWS for 20 s has been highlighted as an effective method for removing bacteria from the hands (WHO 2009). Wetting the hands with water and scrubbing with soap

Table 4 | Factors associated with the reduction of *E. coli*

Characteristic	AOR*	*p*-value	CI
Handwashing step			
(2) Apply enough soap to cover all hand surfaces			
No	ref	–	–
Yes	2.81	**0.048**	1.62–11.8
(4) Right palm over left dorsum with interlaced			
No	ref	–	–
Yes	4.45	**0.006**	1.54–12.8
(9) Rinse hands with water			
No	ref	–	–
Yes	3.28	0.081	0.86–12.5
(10) Dry hands thoroughly with a towel/tissue			
No	ref	–	–
Yes	0.09	0.091	0.01–1.46
Number of steps			
<5 steps	ref	–	–
5 steps or more	0.216	0.069	0.04–1.12
Duration of soap contact			
<10 s	ref	–	–
≥ 10 s	5.07	**0.017**	1.34–19.1
Duration of water contact			
<10 s	ref	–	–
≥ 10 s	0.41	0.166	0.11–1.44
Hand-drying method			
Single-use tissue	ref	–	–
Reusable (hand towels/clothes)	0.03	**0.012**	0.01–0.47

CI, confidence interval; ref, reference value.
*Multivariable model adjusted for handwashing steps 2, 4, 9, and 10, number of steps, duration of soap contact and water contact, and hand-drying. $R^2 = 0.242$.
Bold values represent the significant variables ($p < 0.05$).

creates a lather that traps and eliminates these bacteria (Burton *et al.* 2011). Although some students performed handwashing for more than 20 s, only 6% of them scrubbed their hands with soap for that entire duration, while others did so briefly. It is important to note that scrubbing the hands with soap for more than 10 s significantly reduces *E. coli* concentration. This finding adds to our understanding that using soap for a short duration is insufficient to remove bacteria. Therefore, future handwashing promotion campaigns that target children should emphasize the use of soap and the appropriate duration of soap contact with the hands.

Previous studies have demonstrated the presence of diverse bacteria on different parts of the hands, such as fingertips (Julian *et al.* 2015), palms (Fierer *et al.* 2008), and in-between fingers (Rosenthal *et al.* 2014). Hence, scrubbing specific parts of the hands is essential for effective bacteria removal during handwashing (Julian *et al.* 2015). Similarly, we found a significant correlation between *E. coli* reduction and scrubbing in-between fingers during handwashing. Hand scrubbing during handwashing is an important action that physically destroys germs. However, we observed that more than half (58%) of the students did not wash in-between their fingers during handwashing (step 4). This confirms a previous finding before the pandemic that only 55% performed step 4 (Otsuka *et al.* 2019), indicating that handwashing promotion during the pandemic did not have a significant effect on accomplishing this step. Students often consider the palm as the dirtiest part of the hand (Ray *et al.* 2011), and they may overlook other parts, including in-between fingers, during handwashing. Our findings highlight the importance of focusing on specific parts during handwashing promotion campaigns for children.

Our study also revealed that the method of hand-drying has contrasting effects on *E. coli* reduction after handwashing. The largest reduction was found in the hands of students who used single-use tissue. This parallels a previous study that showed that single-use or paper towels could dry hands while causing less contamination in the washroom environment (Huang *et al.* 2012). Drying hands with single-use paper towels resulted in a larger reduction than air-drying (Jensen *et al.* 2015). By contrast, using reusable hand-drying methods such as cloth towels or clothes had a much smaller reduction effect and was considered a limiting factor in reducing *E. coli*. This may corroborate our finding that there was no reduction in *E. coli* levels for some students after handwashing. Clothes may not dry the hand completely, leaving it slightly moist. Residual water on the other hand may increase the risk of contamination because bacterial transmission is more effective in wet conditions (Patrick *et al.* 1997), so undried hands increase the possibility of recovery of *E. coli* (Ansari *et al.* 1991). Cloth towels have the lowest reduction in the number of fecal bacteria when drying hands compared to warm air and single-use paper (Ansari *et al.* 1991). A possible reason for this is that sharing hand towels with users who practice poor handwashing techniques increases the chance of the transfer of bacteria from poorly washed hands to the towel. Many studies have focused on hand-drying methods by comparing paper towels and air-drying. However, most urban slums lack access to these methods due to financial constraints. Therefore, it is crucial to emphasize the hand-drying method when promoting handwashing. Private reusable cloth towels may be an effective alternative when a school or household cannot provide single-use paper.

On average, we found that handwashing had a minimal effect on bacteria count (34%). This finding is consistent with other studies which reported that a single handwashing is insufficient; sustained and consistent hand hygiene behavior is required to reduce bacteria significantly (Aihara *et al.* 2014). Furthermore, the complexity of *E. coli* through environmental contamination in urban slum areas cannot be underestimated. Microbial contamination of water used to wash hands in schools or households poses a higher risk of hand contamination (Daneshmand *et al.* 2018; Berhanu *et al.* 2021). Some households and schools in Bandung's urban slum use groundwater for activities, such as handwashing, cooking, or bathing (Otsuka *et al.* 2018). Groundwater in some areas of Bandung is moderately to heavily polluted, which can lead to cross-contamination with humans (Hasanawi *et al.* 2022). A previous study indicated that high levels of *E. coli* have been detected in various types of water in peri-urban areas (Asada *et al.* 2022). If the water is potentially unsafe, it will potentially become a contaminant for hands. This necessitates further research to investigate the pathway of *E. coli* contamination in school and household environments in order to track and identify the sources.

This study has some limitations. This study employed a purposive sampling of two schools with relatively small sample size and the same level of sanitation in the study area. Hence, the findings may not be generalizable to all children in LMIC urban slums. In addition, there was no observation at the household level, which would represent students' daily practice in a natural setting. The presence of observers and the facilities provided during the handwashing observation may have biased the participants to modify their handwashing technique. To minimize this bias, we conducted the evaluation in a detached space and consistently reminded participants to wash their hands as they normally would. Nonetheless, this study enabled us to assess the importance of handwashing steps in reducing bacteria. Regarding the method of *E. coli* detection, we used an enzymatic method that cannot specifically recognize the type of *E. coli*. *E. coli* has many types of strains, some of which can cause severe health hazard, such as diarrhea. Lastly, *E. coli* is not the sole indicator for fecal contamination on hands; there are other bacteria that may serve as indicators, which should be considered for future studies.

CONCLUSION

This study highlights the correlation between poor handwashing practices among young children living in an urban slum and fecal contamination on their hands. There may be a misconception that the primary purpose of handwashing is simply to wet hands and clean palms, while other parts of the hands are overlooked. The study recommends that the focus on handwashing techniques for children should be extended to include scrubbing specific parts of the hands, such as between fingers and under fingernails, where fecal contamination can be found. Additionally, this study found that the low percentage of students who use soap and single-use tissue is likely due to the limited availability of facilities or prevalent social practice at home. To improve the removal of *E. coli* contamination from hands and prevent cross-contamination, it is recommended that schools provide soap and single-use tissue paper. To improve fecal contamination removal on the hands, it is recommended that schools provide soaps and single-use tissue paper. Proper handwashing is critical in preventing the spread of pathogens, and education on proper handwashing should start early for young children. Handwashing intervention should be tailored to the needs of LMICs, where resources may be limited. Further research that incorporates a comprehensive assessment of handwashing behavior and environmental contamination is needed to understand why handwashing does not effectively reduce fecal contamination in some students.

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DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

CONFLICT OF INTEREST

The authors declare there is no conflict.

REFERENCES

Abba, K., Sinfield, R., Hart, C. A. & Garner, P. 2009 Pathogens associated with persistent diarrhea in children in low and middle-income countries: Systematic review. *BMC Infectious Diseases* **9**, 88. doi:10.1186/1471-2334-9-88.

Agestika, L., Otsuka, Y., Widyarani., Sintawardani, N. & Yamauchi, T. 2019 Handwashing skills, hand bacteria reduction, and nutrition status of elementary school children in an urban slum Indonesia. *Sanitation Value Chain* **3** (1), 13–32.

Aihara, Y., Sakamoto, I., Kondo, N., Shrestha, S. & Kazama, F. 2014 Handwashing and microbial contamination on the palms of preschool children in Kathmandu, Nepal. *Journal of International Health* **29** (2), 69–74. doi:10.1011197/jaih/29/69.

Amin, N., Pickering, A. J., Ram, P. K., Unicomb, L., Najnin, N., Hokaira, N., Ashraf, S., Abedin, J., Islam, M. S. & Luby, S. P. 2014 Microbiological evaluation of the efficacy of soapy water to clean hands: A randomized, non-inferiority field trial. *American Journal of Tropical Medicine and Hygiene* **91** (2), 415–423. doi:10.4269/ajtmh.13-0475.

Ansari, S. A., Springthorpe, V., Sattar, S. A., Tostowaryk, W. & Wells, G. A. 1991 Comparison of cloth, paper, and warm air drying in eliminating viruses and bacteria from washed hands. *American Journal of Infection Control* **19** (5), 243–249.

Asada, Y., Chua, M. L., Tsurumi, M., Yamauchi, T., Nyambe, I. & Harada, H. 2022 Detection of *Escherichia coli*, rotavirus, and *Cryptosporidium* spp. from drinking water, kitchenware, and flies in a periurban community of Lusaka. Zambia. *Journal of Water and Health* **20** (7), 1027. doi:10.2166/wh.2022.276.

Badan Pusat Statistik Kota Bandung [BPS-Statistics of Bandung Municipality] 2022 *Kota Bandung Dalam Angka 2022. (Bandung in Figures 2022)*. Badan Pusat Statistik Kota Bandung, Bandung.

Berhanu, L., Mereta, S. T., Gume, B., Kassa, T., Berihun, G., Dadi, L. S., Suleman, S., Tegegne, D., Gataneh, A. & Bedru, H. 2021 Effect of microbial quality of washing water on hand hygiene status of food handlers in Jimma town: Implication for food hygiene and safety. *Journal of Multidisciplinary Healthcare* **14**, 1129–1134. doi:10.2147/JMDH.S306359.

Burton, M., Cobb, E., Donachie, P., Judah, G., Curtis, V. & Schmidt, W. P. 2011 The effect of handwashing with water or soap on bacterial contamination of hands. *International Journal of Environmental Research and Public Health* **8**, 97–104. doi:10.3390/ijerph8010097.

Cantrell, M. E., Sylvestre, E., Wharton, H. C., Scheidegger, R., Curchod, L., Gute, D. M., Griffiths, J., Julian, T. R. & Pickering, A. J. 2023 Hands are frequently contaminated with fecal bacteria and enteric pathogens globally: A systematic review and meta-analysis. *ACS Environmental Au*. doi:10.1021/acsenvironau.2c00039.

Curtis, V., Scott, B. & Cardosi, J. 2005 *The Handwash Handbook: A Guide for Developing a Hygiene Promotion Program to Increase Handwashing with Soap*. The World Bank Group, Washington, DC, USA, pp. 67–68.

Daneshmand, T. N., Friedrich, M. N. D., Gacher, M., Montealegre, M. C., Mlambo, L. S., Nhiwatiwa, T., Mosler, H. J. & Julian, T. R. 2018 *Escherichia coli* contamination across multiple environmental compartments (soil, hands, drinking water, and handwashing water) in urban Harare: Correlations and risk factors. *American Journal of Tropical Medicine and Hygiene* **98** (3), 803–813. doi:10.4269/ajtmh.17-0521.

Dwipayanti, N. M. U., Lubis, D. S. & Harjana, N. P. A. 2021 Public perception and hand hygiene behavior during COVID-19 pandemic in Indonesia. *Frontiers in Public Health* **9**, 621800. doi:10.3389/fpubh.2021.621800.

Fierer, N., Hamady, M., Lauber, C. L. & Knight, R. 2008 The influence of sex, handedness, and washing on the diversity of hand surface bacteria. *PNAS* **105**, 17994–17999. doi:10.1073pnas.0807920105.

Freeman, M. C., Stocks, M. E., Cumming, O., Jeandron, A., Higgins, J. P. T., Wolf, J., Pruss-Ustun, A., Bonjour, S., Hunter, P. R., Fewtrell, L. & Curtis, V. 2014 Hygiene and health: Systematic review of handwashing practices worldwide and update of health effects. *Tropical Medicine & International Health* **19**, 906–916. doi:10.1111/tmi.12339.

Friedrich, M. N. D., Julian, T. R., Kappler, A., Nhiwatiwa, T. & Mosler, H. J. 2017 Handwashing, but how? Microbial effectiveness of existing handwashing practices in high-density suburbs of Harare, Zimbabwe. *American Journal of Infection Control* **45**, 228–233. doi:10.1016/j.ajic.2016.06.035.

Gizaw, Z., Yalew, A. W., Bitew, B. D., Lee, J. & Bisesi, M. 2022 Effects of local handwashing agents on microbial contamination of the hands in a rural setting in Northwest Ethiopia: A cluster randomized controlled trial. *BMJ Open* **12**, e056411. doi:10.1136/bmjopen-2021-056411.

Hasanawi, A., Salami, I. R. S. & Thufailah, N. A. 2022 Spatial analysis of health risk due to contamination groundwater resources (a study in Bandung District, Indonesia). *IOP Conference Series: Earth and Environmental Science* **1109**, 012074.

Hirai, M., Graham, J. P., Mattson, K. D., Kelsey, A., Mukherji, S. & Cronin, A. A. 2016 Exploring determinants of handwashing with soap in Indonesia: A quantitative analysis. *International Journal of Environmental Research and Public Health* **13**, 868. doi:10.3390/ijerph13090868.

Huang, C., Ma, W. & Stack, S. 2012 The hygienic efficacy of different hand-drying methods: A review of the evidence. *Mayo Clinic Proceedings* **87** (8), 791–798. doi:10.1016/j.mayocp.2012.02019.

Jensen, D. A., Danyluk, M. D., Harris, L. J. & Schaffner, D. W. 2015 Quantifying the effect of hand wash duration, soap use, ground beef debris, and drying methods on the removal of *Enterobacter aerogenes* on hands. *Journal of Food Protection* **78** (4), 685–690. doi:10.4315/0362-028X.JFP-14-245.

Julian, T. R., Islam, M. A., Pickering, A. J., Roy, S., Fuhrmeister, E. R., Ercumen, A., Harris, A., Bishai, J. & Schwab, K. 2015 Genotypic and phenotypic characterization of *Escherichia coli* isolates from feces, hands, and soils in rural Bangladesh via the Colilert Quanti-Tray System. *Applied and Environmental Microbiology* **2015** (81), 1735–1743. doi:10.1128/AEM.03214-14.

Karon, A. J., Cronin, A. A., Cronk, R. & Hendrawan, R. 2017 Improving water, sanitation, and hygiene in schools in Indonesia: A cross-sectional assessment on sustaining infrastructural and behavioural interventions. *International Journal of Hygiene and Environmental Health* **220** (3), 539–550. doi:10.1016/j.ijheh.2017.92.001.

Kim, S., Brown, A. C., Murphy, J., Oremo, J., Owour, M., Ouda, R., Person, B. & Quick, R. 2019 Evaluation of the impact of antimicrobial hand towels on hand contamination with *Escherichia coli* among mothers in Kisumu County, Kenya, 2011-2012. *Water Research* **157**, 564–571. doi:10.1016/j.watres.2019.03.085.

Kundu, A., Smith, W. A., Harvey, D. & Wuertz, S. 2018 Drinking water safety: Role and hand hygiene, sanitation facility and water system in semi urban areas of India. *American Journal of Tropical Medicine and Hygiene* **99** (4), 889–898. doi:10.4269/ajtmh.16-0819.

Levine, M. M., Nasrin, D., Acacio, S., Bassat, Q., Powel, H., Tennant, S, M., Sow, S, O., Sur, D., Zaidi, A. K. M., Faruque, A., Hossain, M. J., Alonso, P. L., Breiman, R. F., O'reilly, C. E., Mintz, E. D., Omore, R., Ochieng, J. B., Oundo, J. O., Tamboura, B., Sonogo, D., Onwuchekwa, U., Manna, B., Ramamurthy, T., Kanungo, S., Ahmed, S., Qureshi, S., Quadri, F., Hossain, A., Das, S.K., Antonio, M., Saha, D., Mandomando, I., Blackwelder, W. C., Faraq, T., Wu, Y., Houpt, E. R., Verweiij, J. J., Sommerfelt, H., Nataro, J. P., Robbins-Browne, R. M. & Kotloff, K. L. 2020 Diarrhoeal disease and subsequent risk of death in infants and children residing in low-income and middle-income countries: Analysis of the GEMS case-control study and 12-month GEMS-1A follow-on study. *The Lancet Global Health* **8**, e204–e214. doi:10.1016/S2214-109X(19)30541-8.

Ogba, O. M., Asukwo, P. E. & Bassey, I. B. O. 2018 Assessment of bacterial carriage on the hands of primary school children in Calabar municipality, Nigeria. *Biomedical Dermatology* **2**, 6. https://doi.org/10.1186/s41702-017-0017-0.

Otsuka, Y., Ushijima, K., Ikemi, M., Nilawati, Sintawardani, N. & Yamauchi, T. 2018 Mapping of water, sanitation, hygiene and child health in an urban slum of Indonesia. *Sanitation Value Chain* **2** (1), 27–37.

Otsuka, Y., Agestika, L., Harada, H., Sriwuryandari, L., Sintawardani, N. & Yamauchi, T. 2019 Comprehensive assessment of handwashing and faecal contamination among elementary school children in an urban slum of Indonesia. *Tropical Medicine & International Health* **24** (8), 954–961. doi:10.1111/tmi.13279.

Paraduth, S. K. & Biranjia-Hurdoyal, S. D. 2015 Hygiene practice and faecal contamination of the hands of children attending primary school in Mauritius. *International Health* **7**, 280–284. doi:10.1093/inthealth/ihu080.

Patrick, D., Findon, G. & Miller, T. 1997 Residual moisture determines the level of touch-contact-associated bacterial transfer following hand washing. *Epidemiology and Infection* **1119** (3), 319–325. doi:10.1017/S0950268897008261.

Pickering, A. J., Julian, T. R., Mamuya, S., Boehm, A. B. & Davis, J. 2011 Bacterial hand contamination among Tanzanian mothers varies temporally and following household activities. *Tropical Medicine & International Health* **16**, 233–239. doi:10.1111/j.1365-3156.2010.02677.

Ray, S. K., Amarchand, R., Srikanth, J. & Majumdar, K. K. 2011 A study on prevalence of bacteria in the hands of children and their perception on hand washing in two schools of Bangalore and Kolkata. *Indian Journal of Public Health* **55** (4), 2011. doi.10.4103/0019-557X.92408.

Rosenthal, M., Aiello, A., Larson, E., Chenoweth, C. & Foxman, B. 2014 Healthcare workers' hand microbiome may mediate carriage of hospital pathogens. *Pathogens* **2014** (3), 1–13. doi:10.3390/pathogens3010001.

Saboori, S., Greene, L. E., Moe, C. L., Freeman, M. C., Caruso, B. A., Akoko, D. & Rheingans, R. D. 2013 Impact of regular soap provision to primary schools on hand washing and *E. coli* hand contamination among pupils in Nyanza Province, Kenya: A cluster-randomized trial. *American Journal of Tropical Medicine and Hygiene* **89** (4), 698–708. doi:10.4269/ajtmh.12-0387.

Sai, A., Al furqan, R., Ushijima, K., Hamidah, U., Ikemi, M., Widyarani., Sintawardani, N. & Yamauchi, T. 2020 Personal hygiene, dignity, and economic diversity among garbage workers in an urban slum of Indonesia. *Sanitation Value Chain*. doi:10.34416/svc/00019.

Seid, M., Yohanes, T., Goshu, Y., Jemal, K. & Siraj, M. 2022 The effect of compliance to hand hygiene during COVID-19 on intestinal parasitic infection and intensity of soil transmitted helminthes, among patients attending general hospital, southern Ethiopia: Observational study. *PLoS ONE* **17** (6), e0270378. doi:10.1371/journal.pone.0270378.

Tarigan, A. K. M., Sagala, S., Samsura, D. A. A., Fiisabilillah, D. F., Simarmata, H. A. & Nababan, M. 2015 Bandung city, Indonesia. *The International Journal of Urban Policy and Planning Cities* **50**, 100–110. doi:10.1016/j.cities.2015.09.005.

World Health Organization 2009 *Guidelines on Hand Hygiene in Health Care: First Global Patient Safety Challenge Clean Care Is Safer Care*. WHO, Geneva, Switzerland.

Xuan, L. T. T. & Hoat, L. N. 2011 Handwashing among schoolchildren in an ethnically diverse population in northern rural Vietnam. *Global Health Action* **6** (1), 18869. doi:10.3402/gha.v6i0.18869.

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