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# Handwashing Practice of Food Establishment Customers, Microbial Quality of Handwashing Water, and Associated Factors in Ginjo Kebele, Jimma Town, Southwest Ethiopia

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**ABSTRACT:** Handwashing plays a major role in preventing the spread of infection and, if poorly implemented, leads to different illnesses. However, the quality of water meant for handwashing and handwashing practice and hand hygiene conditions of food establishment customers are overlooked issues, unlike drinking water and hygienic conditions and practice of food handlers. This study aimed to assess the microbial quality of water used for handwashing and hygienic practice and the status of customers of food establishments and associated factors in Ginjo Kebele, Jimma town, Southwest, Ethiopia. A cross-sectional study design was employed in July 2022. Forty water and 40 hand swab samples were taken and analyzed for total coliform and *E. coli* using the spread plate method. Handwashing facility conditions and the handwashing practices of customers were assessed using a checklist and questionnaire. The data were analyzed by SPSS version 23 software. The median handwashing water total coliform count was 390.0CFU/ml. The median hand swab samples' total coliform and *E. coli* load were 21.6 and 4.8CFU/cm<sup>2</sup>, respectively after handwashing practice. There was an uphill correlation between the handwashing water and hand swab sample total coliform load ( $r = .34$ ,  $P = .032$ ). The mean handwashing efficacy for the removal of total coliform was 25.8%. The microbial load was varied with the type of water container used, method of taking water from the container, handwashing practice, and hand drying. The microbial quality of handwashing water and hand swab samples didn't comply with the acceptable limit. Thus, governmental bodies should work to improve the handwashing practice of communities. Food establishment owners should treat water for handwashing, clean the water containers, handle the water properly, and avail soap and poster demonstrators to the handwashing facilities. The customers should also comply with proper hand washing practices and reduce hand contamination.

**KEYWORDS:** Customer, food establishment, hand hygiene, handwashing facility, handwashing water, microbial quality

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## Introduction

Food and drink establishments are places where an individual gets food for breakfast, lunch, dinner, or snacks. Many factors including population growth, living standard, change, and temporary settlement for training or other purpose-led a person to eat from food catering establishments.<sup>1</sup> This provokes the opening of many establishments in the urban area, specifically around higher institutions like universities. However, the food from these establishments is prone to contamination due to poor personal hygiene,<sup>2</sup> improper food, and drink handling, and contaminated food surfaces and equipment.<sup>3</sup> Poor personal hygiene plays a major role in this regard and is responsible for many foodborne diseases. In some cases, food establishments missed any means of handwashing services like water and soap.<sup>4</sup>

Globally, foodborne illness is responsible for morbidity and mortality and remains a major public health concern.<sup>5–7</sup> Based on the World Health Organization (WHO) 2020 report, annually, nearly 420 000 deaths occur due to foodborne illnesses.<sup>8</sup> In developing countries, 80% of the diseases are associated with poor home and personal hygiene and about 2.2 million people die; most children die annually due

to diarrhea; the same number again die from acute respiratory infections.<sup>9</sup> Poor personal hygiene causes diarrheal<sup>10</sup> and respiratory diseases and is responsible for half of all child deaths per year globally.<sup>11,12</sup> Over 70% of diarrhea cases in developing countries are foodborne illnesses.<sup>5,6</sup> According to the 2021 report, around 670 million people had no access to handwashing facilities at the home level regardless of the utilization of the available facilities.<sup>13</sup> Only a 30 million access coverage increment was shown in the first 5-year period of Sustainable Development Goals (SDGs) achievement. The issue is more concerning in low- and middle-income countries including East African nations.<sup>13,14</sup> The problem is huge in Ethiopia where 38% of the population lack access to handwashing facilities at the household level<sup>13</sup> and have improper and insufficient handwashing practices.<sup>9</sup>

Hands are the highways for the transmission and spread of pathogens that cause diseases and foodborne illnesses. Numerous studies support the finding that handwashing reduces the carriage of pathogens on the hands.<sup>15,16</sup> In this regard, to ensure the safety of the food service and protect the service users (customers) of the establishments, it has been emphasized the handling and microbial quality of



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drinking water<sup>17</sup> and the hygienic condition and practice of food handlers.<sup>5,18,19</sup> Poor hygienic status of the hands has been linked to the individual's handwashing practice and availability of safe water.<sup>11,18</sup> Handwashing practice at critical times is important to reduce diarrheal diseases and respiratory infections.<sup>11,20</sup> The critical times for food contamination problems are during food harvesting, preparation, handling, and processing.<sup>21</sup> Food contamination happens at any point from production to consumption or farm to fork.<sup>22,23</sup> The disregard for hygienic measures enables pathogens to come into contact with food and, in some cases, to survive and multiply in sufficient numbers to cause illness in consumers. The hands are particularly important since they are the last line of defense in the chain of transmission of gastrointestinal pathogens, directly from hand to mouth. However, hands washed with contaminated water pose a risk for higher levels of recontamination.<sup>24,25</sup> Improper or inadequate handwashing practices of food handlers have been identified as the cause of foodborne illnesses.<sup>9,19</sup> The quality of water for handwashing<sup>18</sup> and handwashing practice and hygiene of customers matter in preventing illnesses<sup>25</sup> but these are overlooked issues. Whatever the food is prepared and served safely, unless the hand of customers that take the food-to-mouth is hygienically safe, the problem will continue to occur. Thus, this study is aimed to assess (1) the hand hygiene status of customers of the food service establishments and (2) its associated factors based on the handwashing facility sanitary conditions, handwashing water quality, and customer's handwashing practice.

## Materials and Methods

### *Study setting, Study design, and period*

This study was conducted in Ginjo Kebele, Jimma town, which is located 355 km Southwest of Addis Ababa, Ethiopia.<sup>19</sup> The area lies within a latitude range from 7°13' to 8°56' N and a longitude range of 35°52' to 37°37' E and between an elevation of 1740 and 1760 m above sea level. The average maximum and minimum temperatures of the area are 25°C to 30°C and 7°C to 20°C respectively. The area receives annual precipitation ranging from 1200 to 2000 mm<sup>26,18</sup> and is known for its coffee plantations.<sup>27</sup> According to the Kebele Health post office, there are a total of 50 food service establishments in Kebele. Which include 4 hotels, 10 bars and restaurants, 27 Migib bets (small establishments that serve only food), and 5 groceries. A cross-sectional study design was employed in July 2022.

### *Sample size and sampling technique*

A total of 40 food service establishments were considered. Only those establishments that are licensed by the local authorities and actively functioning during the study period were included. Based on this at the time of data collection, the

remaining 10 were not included. One customer from each establishment was selected randomly for a hand hygiene examination. The sample was taken before and after a free-will customer's handwashing practices, making the total hand rinse sample to be 80. In addition, 40 handwashing water samples were collected for microbial quality investigation.

### *Inclusion and exclusion criteria*

A customer who came to the food service establishment for service was considered, while those customers who had skin irritation, eczema, inflammation, and wounds at the time of sampling were excluded.

### *Data collection*

After verbal consent was obtained from food service establishment management and the customers, data related to the socio-demographic status of the customers, source of water, handwashing facility conditions, and handwashing practices of the customers were collected through a face-to-face interview (with customers and establishment head) and direct observation methods using a structured questionnaire and an observational checklist. These data were collected side-by-side together with sample specimen collection (handwashing water and hand swab samples).

### *Sample collection*

From each of the 40 food service establishments, a 100 ml water sample was collected using a sterile glass bottle directly from tap water and water storage tanks (containers) where the customers used to wash their hands. The participants (customers) were asked to give a hand rinse sample at the beginning of data collection. Forty hand rinse samples were collected by swab method using sterile cotton-headed swabs from the dominant (eating) hands of the customers of the establishment using a sterile template measuring 25 cm<sup>2</sup>.<sup>28</sup> The hand was rolled between the thumb and index finger as it has high contact with food at the time of eating. The hand swab samples were collected before the customers washed their hands and after washed and get dried them. The participants were kept unaware of the sampling after the handwashing practice. Thereafter, the swab stick was placed into the tube containing 0.1% of 5 ml sterile peptone-buffered water.<sup>5,28-30</sup> Finally, the tubes and water samples were labeled and put into a cold box (<4°C) and transported to the Laboratory of Environmental Health Sciences and Technology Department at Jimma University for analysis within 4 hours of collection.<sup>18,28</sup>

### *Sample analysis*

In the laboratory, both water and hand swab samples were analyzed for total coliform and *E. coli*. Total coliform was

determined to assess the quality of water for handwashing and the effectiveness of hygienic practices (handwashing)<sup>28,31,32</sup> of the customers. However, *E. coli* was used to see the health risk of handwashing water and the hands of the customers.<sup>18,28</sup> Hence, only the swab sample is taken after handwashing was analyzed for *E. coli*. The spread plate technique was applied to culture both total coliform and *E. coli* on nutrient agar and MacConkey agar (Oxoid, UK), respectively. The media were prepared and sterilized following the manufacturer's instructions. Serial dilutions of  $10^{-1}$  and  $10^{-2}$  were made based on the pretest made before the actual sample analysis and plates with a countable number of colonies (30–300) were considered to avoid the chance of error due to overcrowded growth of microbes and a small number of samples. After the sample specimen was spread over the agar media aseptically using a spreader, it was incubated at 32°C and 37°C for about 24 to 48 hours for total coliform and *E. coli*,<sup>21,33</sup> respectively. This was done in duplicate together with 1 negative field control and 1 negative laboratory control for both water and hand swab samples to check for the procedures (sampling, handling, and analysis) followed. Thereafter, the growth of the culture was enumerated for the individual colony forming units manually with the help of a magnifying glass. Only colonies appearing with non-mucoid pink color on MacConkey agar (Oxoid, UK) were considered as *E. coli*. The average value of the duplicate analysis was taken and finally, the result was expressed in the form of colony-forming units per milliliter (CFU/ml) and per square centimeter (CFU/cm<sup>2</sup>) for water and hand swab samples respectively.

### Statistical analysis

Data were entered into Microsoft Excel 2016 and exported to SPSS version 23 for further analysis. Descriptive analyses like; frequency, percent, mean, median, min, max, etc. were computed. As the bacterial count data were not normally distributed, Spearman's correlation was used to show the relationship between the handwashing quality and the hand hygiene status of the customers. The microbial quality of handwashing water and customer's hand hygiene status association with other predictor variables were checked. The Chi-square test was used to test the association among predictor variables, whereas the Mann-Whitney test and Kruskal Wallis test were used to compare the statistical mean difference in microbial load of handwashing water and hand hygiene status of customers between and/or among predictor variables groups. For the statistically significant Kruskal Wallis test, a multiple comparison test was done using the Mann-Whitney test to see the specific mean difference between variable groups. On the other hand, the Wilcoxon Signed Rank test was used to see the mean difference in the hand hygiene status of customers before and after the handwashing practice. A *P*-value of less than or equal to .05 was considered a significant statistical result.

### Data quality assurance

To assure the quality of this study, all necessary field and laboratory standard procedures were strictly followed. To know the completeness of the assessment tools and the appropriate dilution requirement of the specimen, samples were taken from the neighbor Kebele and pretested 1 week prior to the actual data collection. In addition, field and laboratory negative controls were used by taking sterilized water and peptone water for both water and swab samples. In the laboratory, duplicate analysis was carried out. Up-to-date media and reagents were used and prepared following the manufacturer's instructions.

### Operational definitions

**Proper handwashing:** Wetting the hands with clean running water, followed by the use of soap and rub of the lathered hands together for at least 20 seconds, rinsing them under clean running water.<sup>34</sup> Otherwise, if 1 or more of the procedures is/are missed, it is considered as an improper handwashing practice.

**Clean water container:** If there is no filth material or defect on the coverage, at the outlet or opening, or on the external part of the water container.

**Stored protected water:** Water stored in a temporary container (not directly from the tap) with coverage and a sink used exclusively for handwashing and separated clearly from the potential sources of contaminants such as near the latrines.

**Formal training:** If the customers get training in a scheduled and organized way on hand hygiene, its public health significance, and how effective handwashing should be for half a day or more duration with practical demonstrations. Otherwise, informal if it is in the form of short health information dissemination and public awareness creation campaign programs.

**Repairment requirement:** When the hand washing facility's storage container and sink have a cleanness problem, they are visually old and leaking, it was considered that the facility requires repairment. A facility without all of these problems does not require repairmen.

## Results

### Sociodemographic characteristics of the customers

Out of the 40 respondents from the food service establishments included in the study, 29 (72.5%) of them were male and the majority of them, 37 (92.5%) aged between 18 and 35 years. Regarding educational status, 26 (65.0%) of the study participants had College and above level. Thirty (75%) of the study participants had trained in handwashing practice, and only 15 (50.0%) customers had taken formal training (Table 1).

### Handwashing facility sanitary conditions of the establishment

In this study, of the 40 food service establishments included in the study, 22 (55.0%) of them were Migib bets and 5 (12.5%)



**Table 1.** Socio-demographic characteristics of the customers of food service establishments in Ginjo Kebele, Jimma town, Southwest Ethiopia, 2022 (N=40).

VARIABLE	CATEGORY	FREQUENCY	PERCENT
Sex	Female	11	27.5
	Male	29	72.5
Age (year)	18-35	37	92.5
	36-50	3	7.5
Educational status	Primary school	4	10
	Secondary school	10	25.0
	College and above	26	65.0
Training status on handwashing	Trained	30	75.0
	Not trained	10	25.0
Training type	Formal	15	50.0
	Informal	15	50.0
Fingernail status	Trimmed	29	72.5
	Untrimmed	11	27.5

were Groceries. In 11 (27.5%) establishments, the handwashing facilities had water containers without coverage. A poster demonstrating the proper handwashing practice was not available in all the studied food service establishments attached to the handwashing facility. Fourteen (35%) of the handwashing facilities used pouring methods of taking handwashing water (Table 2).

#### *Handwashing practice of customers*

The findings of this study revealed that more than half of the customers practice improper handwashing. Twenty-one (52.5%) of them washed their hands within less than 20 seconds and 16 (40%) of them rubbed their hands on their clothes to dry their hands (Table 3).

#### *Microbial quality of water and hand hygiene status of customers, and association factors*

The median handwashing water total coliform and *E. coli* records were 390 and 70 CFU/ml, respectively. The median total coliform loads of hand rinse samples before handwashing hand samples were 28.4 CFU/cm<sup>2</sup>, while the median total coliform and *E. coli* counts after handwashing were 21.6 and 4.8 CFU/cm<sup>2</sup>, respectively (Table 4).

#### *Association between the training history of the customers and their handwashing practices*

The chi-square test statistics revealed that customer training on handwashing practice had a significant association with the

**Table 2.** Conditions of handwashing facilities of food service establishments in Ginjo Kebele, Jimma town, Southwest Ethiopia, 2022 (N=40).

VARIABLE	CATEGORY	FREQUENCY	PERCENT
Type of establishment	Hotel	3	7.5
	Bar and restaurant	10	25.0
	Migib bet	22	55.0
	Grocery	5	12.5
Source of water used for handwashing	Stored protected water	13	32.5
	Stored unprotected water	15	37.5
	Tap water (direct)	12	30.0
Type of water container	Bucket	9	22.5
	Jarkan	10	25.0
	Tanker	9	22.5
	Tap	12	30.0
Presence of container coverage	Without coverage	10	35.7
	With coverage	18	64.3
Method of taking the water from the container	By pouring	14	35.0
	By dipping other material	9	22.5
	Use tap	17	42.5
Cleanness of the facility	Clean	28	70.0
	Not clean	12	30.0
Frequency of cleaning the container for holding water	Daily	10	25.0
	3x a week	10	25.0
	2x a week	9	22.5
	Weekly	11	27.5
Repairment requirement of a handwashing facility	Not required	14	35.0
	Required	26	65.0
Availability of soap	Yes	25	62.5
	No	15	37.5

time taken to wash hands, the method of drying the hand, and proper handwashing practice (Table 5).

The Kruskal Wallis test showed a statistically significant mean difference in water total coliform load among the sources of handwashing water ( $X^2 = 9.506$ ,  $df = 2$ ,  $P = .009$ ), the type of temporary water container ( $X^2 = 12.724$ ,  $df = 3$ ,  $P = .005$ ), the method of taking water from the temporary water container

**Table 3.** Customer's handwashing practice in food service establishments in Ginjo Kebele, Jimma town, Southwest Ethiopia, 2022 (N=40).

VARIABLE	CATEGORY	FREQUENCY	PERCENT
Utilization of soap	Yes	18	45.0
	No	22	55.0
Handwashing duration (s)	Less than 20	21	52.5
	20 and more	19	47.5
Handwashing practice condition	Proper	16	40.0
	Improper	24	60.0
Method of hand drying	Air dried	24	60.0
	Rubbed on their clothes	16	40.0

**Table 4.** Microbial quality of handwashing water and customer's hand hygiene status in food service establishments in Ginjo Kebele, Jimma town, Southwest Ethiopia, 2022 (N=40).

MICROBIAL LOAD	MIN.	MAX.	SUM	MEAN	PERCENTILES		
					25	50	75
Water total coliform (CFU/ml)	2.0	968.0	17502.0	437.6	223.5	390.0	641.5
Water <i>E. coli</i> (CFU/ml)	2.0	512.0	4280.0	107.0	38.5	70.0	134.5
Hands total coliform (CFU/cm <sup>2</sup> ) before handwashing	9.6	176.0	1650.4	41.3	20.2	28.4	50.9
Hands total coliform (CFU/cm <sup>2</sup> ) after handwashing	4.0	120.8	1292.8	32.3	15.7	21.6	36.1
Hands <i>E. coli</i> (CFU/cm <sup>2</sup> ) after handwashing	0.0	56.0	358.0	9.0	1.6	4.8	10.7

Abbreviations: Max. = maximum; Min. = minimum.

**Table 5.** Chi-square test of association between training status of customers and their handwashing practice in food service establishments, Ginjo Kebele, Jimma town, Southwest Ethiopia, 2022 (N=40).

VARIABLES WITH CATEGORY			TRAIN BEFORE		$\chi^2$	DF	P-VALUE
			TRAINED	NOT TRAINED			
Handwashing time	<20s	Observed count	13	8	4.043	1	.044
		Expected count	15.8	5.3			
	≥20s	Observed count	17	2			
		Expected count	14.3	4.8			
Method of drying the hand	Air dried	Observed count	20	3	4.126	1	.042
		Expected count	17.3	5.8			
	Rubbing on their cloth	Observed count	10	7			
		Expected count	12.8	4.3			
Handwashing practice	Proper	Observed count	15	1	5.00	1	.025
		Expected count	12.0	4.0			
	Improper	Observed count	15	9			
		Expected count	18.0	6.0			

( $X^2=7.133$ ,  $df=2$ ,  $P=.028$ ), and frequency of cleaning the handwashing facility ( $X^2=9.273$ ,  $df=3$ ,  $P=0.026$ ). The

multiple comparison tests for the Kruskal Wallis tests are shown in Table 6.

**Table 6.** Mann-Whitney *U* test shows the mean total coliform bacteria load (CFU/ml) difference among variable groups in food service establishments, Ginjo Kebele, Jimma town, Southwest Ethiopia, 2022 (N=40).

MANN-WHITNEY <i>U</i> TEST AS A MULTIPLE COMPARISON TEST					
BACTERIA (CFU/ML)	VARIABLE GROUPS		MEAN RANK	MANN-WHITNEY <i>U</i> TEST	P-VALUE
WTC	Source of handwashing water	SuP*SP	14.9	92	.800
			14.1		
		SuP*Tap	17.9	31	.004
			9.1		
		SP*Tap	16.5	33	.014
			9.3		
WTC	Type of water container	Bucket*Jarkan	10.1	44	.967
			10.1		
		Bucket*Tanker	10.7	29.5	.331
			8.3		
		Bucket*Tap	15.1	17	.009
			7.9		
		Jarkan*Tanker	11.7	28	.165
			8.1		
		Jarkan*Tap	16.2	13	.002
			7.6		
		Tanker*Tap	14.3	24.5	.036
			8.5		
WTC	Method of taking water from the container	Pouring*Dipping	10.3	39	.131
			14.7		
		Pouring*Use tap	18.6	82	.142
			13.8		
		Use tap*Dipping	18.7	30	.012
			10.8		
WTC	Frequency of water container cleaning	Daily*3x a week	9.9	44	.650
			11.1		
		Daily*2x a week	7.9	23.5	.079
			12.4		
		Daily*Weekly	8.0	25	.035
			13.7		
		3x*2x a week	7.2	17	.022
			13.1		
		3x*Weekly	7.7	22	.020
			14.0		
		2x*Weekly	10.4	48.5	.939
			10.6		

\*Comparison between; WTC = water total coliform; SP = stored protected source; SuP = stored unprotected source.

**Table 7.** Shows Mann-Whitney *U* mean total coliform bacteria count (CFU/cm<sup>2</sup>) comparison test between different variables groups in food service establishments, Ginjo Kebele, Jimma town, Southwest Ethiopia, 2022 (N=40).

MANN-WHITNEY <i>U</i> TEST					
BACTERIA (CFU/CM <sup>2</sup> )	VARIABLE GROUPS		MEAN RANK	MANN-WHITNEY <i>U</i> TEST	P-VALUE
HTC	Method of hand drying used	Air dried	14.9	66.0	.000
		Rubbing on cloth	28.1		
HTC	HW time	20s and more	14.8	92.0	.004
		Less than 20s	25.6		
HTC	HW Practice of customers	Proper	13.8	84.5	.003
		Improper	25.0		

Abbreviations: HTC = hand swab total coliform; HW = handwashing.

Spearman's correlation revealed that the water and hand swab sample coliform loads had an uphill weak correlation ( $r = .34$ ,  $P = .032$ ), whereas it is insignificant for *E. coli*. On the other hand, the Wilcoxon Signed Rank test revealed a significant reduction in microbial load after handwashing practice for total coliforms ( $P = .012$ ). The mean handwashing efficacy for the removal of total coliforms was 25.8% which ranged between -9.8% and 85.2%. This coliform removal efficiency had a weak downhill correlation with water quality ( $r = -.364$ ,  $P = .021$ ). Mann-Whitney test results indicated a significant difference in the mean swab sample total coliform load (after handwashing) for different variable groups (Table 7).

## Discussion

The findings of this study revealed that the minimum and maximum handwashing water total coliform counts recorded were 2 and 968 CFU/ml, respectively with a mean value of 437.6 CFU/ml. This finding is below the work of Berhanu et al<sup>18</sup> which was conducted in Jimma town, Southwest Ethiopia, where the mean value was  $5.4 \pm 0.6 \log$  CFU/ml. On the other hand, a study conducted in Southern Nations, Nationalities, and People's Region (SNNPR), Ethiopia, found 27.5% of handwashing facility water had zero total coliforms.<sup>25</sup> This is by far lower than our finding in which 100% of the handwashing water was positive for total coliform. It might be due to the seasonal variation of the studies. The former study was conducted in the dry season where the effect of rain and runoff on the water source pollution is minimal. Regarding *E. coli*, similarly, our findings (mean 107 CFU/ml) revealed a lower load relative to the previous study (mean  $5.0 \pm 0.6 \log$  CFU/ml).<sup>18</sup> These differences might be explained by the fact that the establishments had improved in the handling of handwashing water due to the attention gained during the era of the Coronavirus Disease 2019 (COVID-19) pandemic. Besides, the concentration of *E. coli* in the present study of handwashing water is still higher than the limit concentration (<1000 CFU/100 ml) which allow at least a reduction of the load from soiled hand as modeled by the previous scholars.<sup>35</sup>

The effect at this level is reducing risks if the rate of pathogen removal exceeds the rate of addition. Pathogens can be transferred to hands from non-potable water and pose a risk of infection from subsequent hand-to-mouth contact even in lower concentrations than the limit.

In the present work, the total coliform count from the hands of the customers before practicing handwashing ranged between 9.6 and 176 CFU/cm<sup>2</sup> with a median value of 28.4 CFU/cm<sup>2</sup>. However, for hand hygiene after handwashing, the minimum and the maximum total coliform load were 4 and 120 CFU/cm<sup>2</sup>, respectively, with a median value of 21.6 CFU/cm<sup>2</sup>. This revealed the mean handwashing efficacy for the removal of total coliforms after handwashing was 25.8% and ranged between 9.8% and 85.2%. This lower coliform removal efficiency might be attributed to the high initial load of coliforms on soiled hands and poor quality of handwashing water due to potential microbial transfer from handwashing water.<sup>35</sup> However, the removal efficiency had a weak downhill correlation with water quality ( $r = -.364$ ,  $P = .021$ ). This indicates the effect of handwashing water on the hand hygiene of customers in which hands could not be free from coliforms. Besides, after handwashing, the median *E. coli* load was 4.8 CFU/cm<sup>2</sup> and ranged from 0 to 56 CFU/cm<sup>2</sup>. This result is alarming, as almost all (95%) of the hand swab samples (n=38) were not free of *E. coli*.

The comparison was made with the food handler's hand hygiene status as there is no study conducted on food service establishment's customers so far. When the microbial load after handwashing practice is compared with Berhanu et al<sup>18</sup> found for total coliform and *E. coli* (mean  $4.9 \pm 0.6$  and  $4.2 \pm 0.6 \log$  CFU/ml respectively), ours is lower. The difference might be explained by the fact that the establishments had improved the handling of handwashing water due to the attention gained from the COVID-19 pandemic.<sup>36</sup> This in turn affected the microbial removal from hand as indicated by their correlation ( $r = .34$ ,  $P = .032$ ) in the present study. The lower hand swab microbial load in the present study can also be attributed to the contaminant exposure level of the hands of food handlers and



customers might have differences due to the involvement of handlers in different activities. Similarly, our findings revealed a lower *E. coli* load relative to the previous study.<sup>18</sup> This might be again due to differences in the study participants and the activities they are involved in. The rise in awareness and behavioral change toward the proper handwashing and hygienic practice of the participants after the era of COVID-19<sup>36</sup> and the emergence of other outbreaks like monkeypox might also be a reason as different hygienic practices or interventions including hand hygiene campaign were recommended and applied through international and national institutions to the communities.<sup>37-39</sup>

The positive correlation between water microbial quality and hand hygiene status after handwashing in terms of total coliform ( $r = .34$ ,  $P = .032$ ) in the present study, is in line with the finding of those of Berhanu et al<sup>18</sup> regardless of the strength of correlation. The correlation was strong ( $r = .71$ ;  $P \leq .001$ ) in the former study. However, unlike Berhanu et al<sup>18</sup> there was no significant correlation between handwashing water and hand swab (after handwashing) samples of *E. coli* count in our findings. This variation might be due to the difference in the initial load of hands of customers and food handlers for this microorganism before handwashing. The contact and activity of the food handler and customers involved also vary and might affect the initial load of microorganisms of fecal origin relative to that of total coliforms.

The quality of handwashing water is considered the same quality as potable water<sup>18,25</sup> which does not allow any detection of total coliforms and *E. coli* in 100 ml of water.<sup>17</sup> However, the microbial count recorded for handwashing water was not in line with the WHO standard limit (when a 100 ml sample is free of *E. coli* or thermotolerant coliforms). This implies the hand hygiene status of customers after their handwashing practice has to be *E. coli-free*. However, in this study, it was not at the level that safeguards them in which the median *E. coli* concentration was found to be 4.8 CFU/cm<sup>2</sup> and ranged from 0 to 56 CFU/cm<sup>2</sup>. The presence and high load of *E. coli* indicates fecal contamination and the presence of other pathogens<sup>17</sup> which might have led to the contamination of ready-to-eat foods.<sup>12,40,41</sup> Thus, the probability of acquiring disease-causing microbes and the risk of developing food-borne infections among customers is high.<sup>18</sup>

Different significantly associated factors were identified for the poor (high microbial load) quality of water for handwashing in the present study (Table 6). These factors include the source of water, the type of temporary water containers used by the establishments, the method of taking water from those containers, and the frequency of storage container cleaning. This finding is in agreement with those of Tolosa et al.<sup>27</sup> There was a statistically significant higher mean total coliform count in stored water than in tap water sources in our finding similar to the work of Berhanu et al.<sup>18</sup> This could be due to the age of the stored water and the availability of residual chlorine<sup>25</sup> in

the direct tap water. Similarly, a statistically significant higher mean total coliform count was recorded for handwashing water taken from a bucket relative to the tap water ( $P = .009$ ). In this regard, the present study finding is in line with those of Tolosa et al.<sup>27</sup> The water quality difference from these sources might be attributed to the possible exposure of bucket water at the time of removing and placing a cover to fetch and add water. Regarding the method of fetching water, in the current study, the only significant difference in the mean load of total coliform was observed between the utilization of tap and dipping methods ( $P = .012$ ) and supported by the other's work.<sup>27</sup> It was higher in the handwashing water used by the dipping method. This might be due to the entry of coliform organisms together with the dipping material as it could be contaminated from different sources including the hands of the users and contact surfaces.

Consequently, the water quality in turn affected the hand hygiene status of customers together with the other factors as described in Table 7. This finding is supported by the other's work as the source of water,<sup>18</sup> handwashing facility conditions (unavailability of soap<sup>42</sup>), and customers' improper handwashing practice,<sup>31,43</sup> inadequate handwashing time, lack of training<sup>5,24,44</sup> and unavailability of poster demonstration on good handwashing practice, inappropriate method of hand drying<sup>34,45,46</sup> and method of fetching water from the container<sup>32</sup> had an association with the effectiveness of handwashing practice in the removal of pathogens from hands thereby hand hygiene status.

The present study has several limitations. It was conducted only in the wet season and we recommend the next researcher to consider seasonal variation along with other physicochemical water parameters including the level of residual chlorine in the handwashing water. For hand hygiene status determination, only 1 customer per establishment was considered due to resource constraints. All statistical comparisons in this study are at crude associations as the low number of observations ( $n = 40$ ) might limit the choice of appropriate regression models which allow adjustment for potential confounders.

## Conclusion

The findings of this study revealed that the quality of water for handwashing and the hand hygiene status of the customers were poor and exceeded the WHO guideline. The water quality affected the hand hygiene status of the customers. The microbial load of handwashing water was significantly associated with some factors including the source of water, the type of temporary water container, and the method of taking water from the container. On the other hand, the microbial quality of hand swab samples was significantly associated with the training status of customers on handwashing practices, inadequate handwashing time, improper handwashing practice, and method of hand drying used. The median *E. coli* count in handwashing water and hand swab samples after handwashing

practice were 70CFU/ml and 4.8CFU/cm<sup>2</sup>, respectively. Furthermore, as all water samples and almost all (95%) of hand swab samples were positive for *E. coli*, the probability of acquiring disease-causing microbes and the risk of developing infections is high among customers.

Based on the results we recommend: (A) the owners of the establishments to (1) treat handwashing water by disinfecting using chlorine (bleach) and have a frequent follow-up on the quality in collaboration with the other stakeholders, (2) avail soap and posters that demonstrate proper handwashing at washing facility, (3) use handwashing water container with coverage or use direct tap water if possible, (4) use tap methods of taking water to prevent cross-contamination. (B) The customers to (1) practice proper handwashing, (2) avoid rubbing hands on the clothes they wear, use air drying in the absence of paper towels and reduce contamination of hands in their daily activities. It can be achieved by using hand disinfectants. (C) The concerned government bodies to give training to the community on proper handwashing practices.

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### Author Contributions

MA conceived and designed the study. FF and ME collected the data, all authors analyzed and interpreted the data, and MA prepared and wrote the manuscript. The final manuscript was critically reviewed and approved by all authors.

### Availability of Data and Materials

Data will be available upon request, by the corresponding author.

### Ethical Consideration

Ethical clearance was obtained from the ethical review board, the Institute of Health Science, Jimma University.

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