Microbial risk assessment of drinking water filtration dispenser toll machines (DFTMs) in Mahasarakham province of Thailand

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ABSTRACT

This investigation aimed to assess the microbial quality of drinking water produced by drinking water filtration dispenser toll machines (DFTMs) to assess the health risk of DFTM water. A total of 210 samples were randomly collected of 70 DFTMs within a radial distance of 500 metres around Mahasarakham University. The prevalence of *Escherichia coli* and *Staphylococcus aureus* in the DFTM water was 54.17% and 16.67% with an average concentration of 1.04 log cfu/ml and 0.26 log cfu/ml, respectively. The risk calculation (using the @Risk program based on drinking water consumption of 2 L/day) indicated that the probability of exposure was 1.67 E-01, while the probability of illness due to *E. coli* and *S. aureus* was 2.08 E-03 and 1.58 E-05 and the risk of illness was 1.13 E-03 and 2.64 E-06, respectively. The sanitation status and drinking water quality related to health would be influenced by other pathogens due to the poor hygienic conditions of DFTMs and improper environments.

Key words | drinking water filtration dispenser toll machine, microbial risk assessment, water quality

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INTRODUCTION

Water is an essential natural resource for sustaining all living things (Cidu *et al.* 2011) and is important for physical and biochemical metabolisms. For health and safety, drinking water must contain very few pathogens and contaminants (Saati & Faidah 2013). Currently, water produced by DFTMs (i.e., DFTM water) is an alternative way to provide clean and safe drinking water, and DFTM water is widely popular in global urban communities and rural areas (Moniruzzaman *et al.* 2011; Tan *et al.* 2016), also in Thailand. The Department of Health revealed the quality of drinking

water from 40% of DFTMs does not meet the standard requirement (Thai Health Promotion Foundation 2017).

DFTM water is treated by reverse osmosis (RO), carbon filtration and UV radiation. RO can remove organic and inorganic contaminants and 99% of bacteria present in tap water (Park & Hu 2010; WHO 2011). However, DFTM water might be rapidly re-contaminated by other conditions, especially poor hygiene of DFTMs due to irregular maintenance, causing the drinking water to be contaminated with pathogenic microorganisms and toxic chemicals (Prasai *et al.* 2007; Ali *et al.* 2012). Several previous studies have reported microorganism contamination in DFTM water, including coliform bacteria, *Pseudomonas aeruginosa*, and heterotrophic bacteria (Chaidez *et al.* 1999; Ligouri *et al.* 2010; Moosa *et al.* 2015). In one study, there were 76.6% of samples where DFTMs were contaminated by at least one

of the following: total coliform such as faecal coliform, *Escherichia coli*, faecal streptococci, *P. aeruginosa* or *Staphylococcus* spp. (da Silva *et al.* 2008). These microorganisms are potentially associated with waterborne diseases, such as diarrhoea, typhoid fever, cholera, giardiasis, and hepatitis. The *E. coli* and *S. aureus* levels are used as microbial parameter indicators for the cleanliness and hygienic status of drinking water products because they might indicate the potential of diarrhoeal diseases (Mihdhdir 2009; Jessen *et al.* 2013). Hence, a microbial risk assessment of *E. coli* and *S. aureus* contamination in DFTM water is needed to assessment the adverse impact on public health.

A risk assessment technique for quantitative microbial risk assessment (QMRA) has been recommended by the World Health Organization. This technique is a four-step process, including hazard identification, hazard characterization, exposure assessment, and risk characterization, to quantify human health risk (USDA/FSIS 2012) associated with exposure to pathogenic microorganisms and the probability of infection and illness (Machdar *et al.* 2013; Haas *et al.* 2014). Therefore, an investigation of DFTM water quality and microbial risk assessment using QMRA coupled with Monte Carlo simulations was performed to provide a range of uncertainty for infectious illness risks to human health. In addition, a survey of the sanitation status of DFTMs was undertaken to estimate risk and consider appropriate measures for controlling DFTM water quality.

MATERIALS AND METHODS

Data collection

Samples were collected from tap water before (inlet) and after (outlet) filtration by 210 randomized DFTMs located within a radial distance of 500 metres around Mahasarakham University in Mahasarakham Province, Thailand, during the period from May to October 2017.

In addition, other data related to the sanitation status of DFTMs were collected by using survey forms and questionnaires about hygiene conditions. Scoring criteria for the hygiene of water vending machines were adapted from the scoring criteria of a factory producing sealed drinking water bottles of Pimmason (2009).

Water quality analysis

All water samples were analysed as follows.

Physical component:

- pH (electrometric measurement method using a pH meter)
- Turbidity (clarity and transparency, which were quantified by a nephelometric method) (APHA 2012).

Chemical component:

- Total solids (by a gravimetric method)
- Total hardness (quantified as $CaCO_3$ by EDTA titration method).

Biological component:-

- Coliform bacteria (by the most probable number)
- *E. coli* and *S. aureus* (by the spread-plate technique in selective media (AOAC 2006)).

The quality of tap water before entering DFTMs was compared with the standard criteria of the Provincial Waterworks Authority (2011) for tap water quality. The quality of drinking water produced by DFTMs was compared with the standard criteria of the Ministry of Public Health (2013) for water consumed from automatic drinking water dispensers.

Quantitative microbial risk assessment

QMRA was employed to determine the health risks due to exposure to microorganisms in contaminated drinking water produced by DFTMs. QMRA consists of four categories, as follows:

(1) Hazard identification involves defining and describing pathogens associated with health effects in drinking water, and the data include the characteristics of pathogens and outbreaks (Medema 2013).
(2) Hazard characterization involves a qualitative or quantitative description of the adverse health effects due to pathogen exposure and the estimation of the risk of infection is explained by dose–response relationships. Information on disease burden, including symptoms, severity, duration of illness, and death rates from identified pathogens, has been reviewed in the literature to ensure that all people who are likely to be at risk are identified (Codex Alimentarius Commission 2014).

(3) Exposure assessment involves establishing the size and characteristics of the exposed population and the pathway, dose and exposure duration. These data are used for a quantitative assessment of the probability of pathogenic microorganisms being present in drinking water that consumers ingest.

(4) Risk characterization explains the probability of illness occurrence and the severity of health effects in a given population. The risk characterization was carried out to integrate steps (1)–(3) and determine the public health risk of infection.

Exposure assessment and risk characterization were calculated by Monte Carlo simulations with the @Risk version 4.5.1 Professional Edition program. The model calculates a number of iterations to simulate the outcome distribution. In this study, 10,000 iterations of Latin Hypercube sampling (Yunita et al. 2016) were used to determine the average value of pathogen contamination and the amount of drinking water consumed per day based on 10,000 scenarios that might occur.

RESULTS AND CONCLUSIONS

Drinking water quality

The cause or source of contamination in DFTM water is dependent on the tap water quality and DFTM maintenance (Barrell et al. 2000; Traistaru 2011). Based on the water quality study (Table 1), the physical and chemical characteristics of tap water and DFTM water were generally within the standard levels, except for one sample of tap water (1.43%) with a turbidity exceeding the standard level and one sample of DFTM water (1.43%) with a hardness exceeding the standard level. However, in terms of biological quality, both tap water and DFTM water contained total coliform bacteria, E. coli and S. aureus levels exceeding the standard levels. Fewer DFTM water samples were contaminated with microorganisms than tap water samples, indicating that DFTMs can remove some microorganisms.

Total coliform bacteria and E. coli are used as faecal contamination indicators in drinking water (Gruber et al. 2014). Skin or wound S. aureus (Schillinger & Du Vall Knorr 2004) levels can indicate clean and hygienic drinking water production (Gwimbi 2011). The findings showed that DFTM water contamination is due to poor sanitation and maintenance and improper hygiene operations. Similar studies have shown coliform bacteria (Chaidez et al. 1999) and Staphylococcus spp. contamination in DFTM water due to improper maintenance (Schillinger & Du Vall Knorr 2004; da Silva et al. 2008). Hence, coliform bacteria in drinking water might be potentially unsafe enteric pathogens that affect consumer health and increase gastrointestinal disease risk (da Silva et al. 2008; Khatoon & Pirzada 2010).

QMRA

Hazard identification

Pathogen hazards of DFTM water showed E. coli and S. aureus contamination, which might be associated with poor

Table 1 | Tap water and DFTM water quality (n = 70)

Parameters	Tap water before DFTMs			Drinking water from DFTMs		
	Min–max	Std[a]	Number (%) of samples exceeding standard levels	Min–max	Std[b]	Number (%) of samples exceeding standard levels
pH	6.5–7.62	6.5–8.5	0	6.66–7.86	6.5–8.5	0
Turbidity (NTU)	0–12.14	<4	1(1.43)	0–0.21	<5	0
TS (mg/l)	21–596	600	0	1–373	500	0
Hardness (mg/l as CaCO₃)	0–240	300	0	0–140.67	100	1 (1.43)
Total coliform (MPN/100 ml)	<1.8–910	ND	21 (30.0)	<1.8->1,600	<2.2	16 (22.86)
E. coli (cfu/ml)	0–1,316.7	ND	27 (38.57)	0–1,050	ND	38 (54.29)
S. aureus (cfu/ml)	0–176.7	ND	11 (15.71)	0–1,376.7	ND	11 (15.71)

[a]The Provincial Waterworks Authority standard criteria for tap water quality (Provincial Waterworks Authority 2011).
[b]The standard for water consumed from automatic DFTMs following from the Ministry of Public Health (2013).

sanitation and improper maintenance (Schillinger & Du Vall Knorr 2004; Moosa *et al.* 2015; Tan *et al.* 2016). *E. coli* and *S. aureus* bacteria are considered sources of waterborne diseases, principally diarrhoeal diseases (WHO 2011). Outbreaks of diarrhoea due to the consumption of *E. coli*-contaminated drinking water have been reported (Jessen *et al.* 2013).

Hazard characterization

This section is mostly drawn from a literature review and is subsequently explained.

E. coli bacteria cause diarrhoeal diseases, and the virulence depends on the strain. The most virulent strain is *E. coli* O157:H7 of the EHEC group (Park *et al.* 2018; Public Health Agency of Canada 2018). EHEC stimulates the release of Vero-cytoxin-producing *E. coli* (VTEC), also known as Shiga-toxin-producing *E. coli* (STEC) (Nguyen & Sperandio 2012). The syndromes of EHEC range from mild and non-bloody to highly bloody (e.g., haemorrhagic colitis), and disease duration ranges from a few days to a few weeks. Between 2% and 7% of cases can develop potentially fatal haemolytic urinemic syndrome (HUS), which is characterized by acute renal failure and haemolytic anaemia (Obrig 2010; WHO 2011). The number of organisms required to infect 50% of exposed people (ID 50) for *E. coli* O157:H7 ranges between 10^2 and 10^6 (Hunter 2003). It has also been reported that between 10 and 10^2 *E. coli* O157:H7 can possibly cause illness (Jeena *et al.* 2006; Schelin *et al.* 2011; Wu *et al.* 2016). ETEC produces a cholera-like labile toxin or heat-stable peptide toxin. Symptoms caused by ETEC are mild watery diarrhoea, abdominal cramps, nausea and headache. Infection with EPEC causes persistent diarrhoea and symptoms of non-bloody diarrhoea, vomiting and fever in infants for more than 10 to 12 days. EIEC causes watery and occasionally bloody diarrhoea, and the symptoms are similar to those of shigellosis or bacillary dysentery. EAEC infection may cause persistent diarrhoea. Elderly adults, young children and immunocompromised persons have the highest risks of infection (Pavankumar & Sankaran 2008; WHO 2011; Jessen *et al.* 2013).

S. aureus can cause disease by two established mechanisms. The first mechanism is based on the ability of organisms to increase their number and spread in tissues, and the second mechanism is based on the ability of organisms to produce extracellular enzymes and toxins. Diseases caused by multiplying organisms in tissues include boils, skin sepsis, post-operative wound infections, enteric infections, septicaemias, endocarditis, osteomyelitis and pneumonia. The duration from the beginning of symptoms for these infections is relatively long, usually several days (WHO 2011). *S. aureus* can produce staphylococcal enterotoxin (SE), which can cause gastrointestinal disease. Infection can be caused by as few as $20-10^2$ ng of enterotoxin (Asao *et al.* 2003; Schelin *et al.* 2011), especially in children and highly sensitive, vulnerable populations. Acute symptoms of ingesting staphylococcal enterotoxin are nausea, vomiting, abdominal cramps, and diarrhoea; during disease incubation, these symptoms are short in duration and range between 1 and 8 hours. However, no dose–response relationship data were found in the literature (Asao *et al.* 2003; Wu *et al.* 2016).

Exposure assessment

The *E. coli* and *S. aureus* levels in water from DFTMs ($n =$ 210) located within a radial distance of 500 metres around Mahasarakham University in Mahasarakham Province, Thailand were determined. The results showed that 54.17% and 16.67% of samples were contaminated with *E. coli* and *S. aureus*, respectively. The concentrations of the pathogens in drinking water were in the range of 0.52–3.02 log cfu/ml (average = 1.04 log cfu/ml) and 0.52–3.14 log cfu/ml (average = 0.26 log cfu/m), respectively (Table 2).

Exposure assessment is a determination of the probability of exposure to *E. coli* and *S. aureus* from consuming drinking water. The probability of exposure to microorganisms was calculated from the prevalence value and average concentration of pathogens in drinking water. The calculation determined the risk consumption in 1 day, and the amount of drinking water consumption for individuals was 2 L/day. The probability of exposure to microorganisms was assessed by exponential analysis using the @Risk program. The probability of exposure to *E. coli* and *S. aureus* from the consumption of drinking water was calculated to be 1.67 E-01 (Table 3). This result indicates the number of people exposed to *E. coli* and *S. aureus* for every 100 people that consume drinking water.

Table 2 | Survey of the prevalence and levels of *E. coli* and *S. aureus* in drinking water from DFTMs ($n = 70$)

E. coli		S. aureus	
Level (log cfu/ml)	No. of samples	Level (log cfu/ml)	No. of samples
Not detected	32	Not detected	59
0.52–1.00	5	0.52	1
1.21–1.40	4	0.83	1
1.41–1.60	3	1.22	1
1.61–1.80	4	1.3	2
1.81–2.00	3	1.37	1
2.01–2.20	5	2	1
2.21–2.40	4	2.04	1
2.41–2.60	3	2.2	1
2.61–2.80	4	2.29	1
2.81–3.02	3	3.14	1
Average = 1.04 log cfu/ml		**Average** = 0.26 log cfu/ml	
Prevalence = 54.17%		**Prevalence** = 16.67%	

Table 3 | Exposure assessment and risk characterization of *E. coli* and *S. aureus* in drinking water from DFTMs

Factors	E. coli	S. aureus
Probability of exposure to microorganisms	1.67 E-01	1.67 E-01
Probability of illness from exposure to microorganisms	2.08 E-03	1.58 E-05
Risks of illness from exposure to microorganisms	1.13 E-03	2.64 E-06

Risk characterization

This step estimates the probability and risk of illness from exposure to *E. coli* and *S. aureus* in drinking water, and the probability is explained by beta-Poisson. The values of the input variables, including prevalence, amount of water consumed and intake frequency, were randomly taken from the appropriate distributions obtained after 10,000 iterations by Latin Hypercube sampling.

Based on the analysis, the probability of illness from *E. coli* exposure was 2.08 E-03. The risk of illness from exposure to microorganisms was 1.13 E-03. The risk assessment of the exposure probability to *E. coli* in drinking water from DFTMs was low (Table 3).

The probability of illness caused by exposure to *S. aureus* was 1.58 E-05. The risk of illness caused by exposure to this microorganism was 2.64 E-06. Therefore, the risk assessment of the exposure probability to *S. aureus* in drinking water from DFTMs was very low.

Even though *E. coli* and *S. aureus* are associated with waterborne diseases, the health risk to people is different. The susceptible population, such as elderly adults, young children and immunocompromised individuals, has the highest risk of infection and severe illness. Although the risk assessment of the exposure probability to microorganisms indicated low and very low levels, *E. coli* and *S. aureus* and other pathogen contamination in drinking water is still a health concern due to poor DFTM sanitation.

DFTM sanitation survey

The relation between the sanitation or hygiene and the quality of drinking water produced by DFTMs was examined by using a biological quality assessment. The results were divided into two groups: samples that did not pass and those that passed the standard criteria. No relation was found between the hygiene of DFTMs and the quality of drinking water (data not shown). Additionally, a relation between cleaning DFTMs and the quality of drinking water produced by the DFTMs was not observed (Table 4).

The results found no relation between the sanitation of DFTM water and the biological quality of the drinking water. However, previous studies have reported that poor sanitation leads to microorganism contamination of drinking water from DFTMs. Irregular maintenance and cleaning lead to higher levels of bacteria, especially coliform bacteria, on faucets (Jeena *et al.* 2006). Three factors directly affecting the quality of drinking water produced by DFTMs were reported in previous studies, and these factors are the location of the DFTMs, the drip tray water drainage system, and the pathogen source around the DFTMs. In addition, a pathogen source near DFTM water might cause cross-contamination. The quality of drinking water also depends on the tap water used in the production process (Pratum & Khananthai 2017). According to the notification of the Ministry of Public Health, hazardous health activities by the local government shall be subject to the issuance of local regulations for the control of such

Table 4 | Relation between cleaning DFTMs and drinking water quality

Factors	Quality of drinking water		x^2	P-value (P < 0.05)	OR	95% CI
	Not passing the standard criteria Number (%)	Passing the standard criteria Number (%)				
The filtering system is incorrect	5 (10.9)	1 (4.2)	0.34	0.66	2.81	0.31–25.49
The water storage tank is incorrect	37 (80.4)	21 (87.5)	0.45	0.53	0.59	0.14–2.41
The nozzle is incorrect	15 (32.6)	7 (29.2)	0.76	0.98	1.18	0.4–3.44
The water stand is incorrect	9 (19.6)	6 (25)	0.59	0.83	0.73	0.23–2.37
The water receiving door is incorrect	9 (19.6)	6 (25)	0.59	0.83	0.73	0.23–2.37

business in the area of responsibility. Control and supervision of the owners or service providers of drinking water from DFTMs should ensure quality standards for the safety of consumers.

CONCLUSIONS

Although DFTMs are well designed and provide effective treatment processes, including RO, RO and UV and RO and ultrafiltration treatments, this study showed that some water samples from DFTMs had total hardness, total coliform bacteria, *E. coli* and *S. aureus* levels that exceeded the standard levels. The QMRA results indicated that the *E. coli* and *S. aureus* levels cause illness and are linked to diarrhoeal diseases. In the risk assessment, the values were low and very low. However, *E. coli* and *S. aureus* contamination in drinking water produced by DFTMs is a health concern caused by other pathogens due to poor DFTM sanitation conditions. Regular maintenance and cleaning of DFTMs are important to control bacterial growth and the presence of waterborne pathogens. Reducing the health risk for people included using agencies for security surveillance of the drinking water from DFTMs.

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