Epidemiol. Infect. (1990), **105**, 363–375
Printed in Great Britain

Faecal contamination of water and fingertip-rinses as a method for evaluating the effect of low-cost water supply and sanitation activities on faeco-oral disease transmission. I. A case study in rural north-east Thailand

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(*Accepted 25 May 1990*)

SUMMARY

Most villagers in north-east Thailand carry water to their homes and store it in separate containers depending on its subsequent use. In one village, information on water use was collated with the bacteriological quality of stored water, water sources and fingertip-rinses. Stored water quality was a function of water-related activities rather than quality at source ($P < 0{\cdot}0001$). Specifically water used for toilet, washing dishes and cooking-related activities was much more contaminated with faecal bacteria than that used for drinking and cooking. *Salmonella* spp. was significantly more common in water used for washing dishes than drinking ($P < 0{\cdot}05$). *Escherichia coli* contamination of fingertip-rinses was strongly associated with the individual's activity prior to testing ($P < 0{\cdot}0001$); child care, food and water-related activities produced much higher levels of fingertip contamination than others. Dirty utensils used for cooking and eating were usually left to soak and faecal bacterial growth occurred in this grossly contaminated soak-water. Cross-contamination via water handling was the main mechanism of stored water pollution. These results were used to develop a hygiene intervention study presented in a companion paper.

INTRODUCTION

Although the potential transmission of water-borne diseases by a water supply is generally accepted, many of these diseases have multiple means of transmission and are not necessarily prevented by improvements to water quality alone. Most water-borne diseases may also be water-washed and these have been reclassified as faeco-oral diseases [1]. This group is made up of mainly the diarrhoeal diseases which are a major cause of morbidity and infant mortality in developing countries [2]. A review of studies evaluating the health impact of water supply and sanitation found that although methodological problems are inherent in such studies [3], water quality improvements have less impact on diarrhoeal disease than improvements to water availability or excreta disposal [4]. Moreover it was suggested that the combined effects of improvements to all water supply and sanitation activities are greater than the sum of their separate parts.

In developing countries, most rural and poor urban areas do not have household water connections [5]. Generally this means that water must be carried to the home, although some domestic activities may be conducted at source. Cairncross [6] emphasizes the importance of more water to maximize health benefits but demonstrates that the relationship between distance to the source and the amount used is not a simple one. Various studies in Africa show that as journey time decreases, water consumption tends to increase, however a plateau is reached when the return journey takes less than 30 min (equivalent to about 1 km in the absence of queuing). Only when the water is supplied in the house or the yard does consumption increase further, but then it rises by a factor of three or more. Most of this increase is used up in personal and domestic activities as there is an obvious limit to the amount of water used for drinking and cooking.

Where water has to be carried, or intermittent piped supplies prevail, it is common practice to store the water in a container(s) for its subsequent use. Feachem and colleagues [7] found in rural Lesotho that contamination of stored water greatly exceeded that of source, and levels of contamination were positively associated with the time since water collection. However, conflicting evidence has been found elsewhere. In rural Egypt, water samples taken from public taps and stored water had similar levels of protozoan contamination, however helminth ova were found repeatedly in samples of stored water but never in tap water [8, 9]. Most tap water samples contained faecal streptococci but *E. coli* were rarely present; the stored water contained consistently higher levels of both bacteria. They suggest that the expected reduction of bacterial counts over time due to the activities of other organisms, natural wastage and nutrient deficiency, is compensated by subsequent contamination during the filling and taking of water. However, this proposed effect did not lead to a significant increase in protozoa. Conversely, the bacteriological quality of stored water in Malawi was found to be primarily a function of quality at source [10]. However, water was usually taken from storage containers by a cup with a handle which should reduce water contamination. Another study in Zambia also found little difference between contamination of water sources and stored water used for drinking and cooking [11].

In rural Bangladesh, *E. coli* was found in 50% of drinking water samples, and levels of contamination increased with environmental temperature [12]. It was reported that water sources had similar levels of contamination, but water samples were taken from children's cups at meal times and there is no mention of sampling the sources. In hot climates, regrowth of *E. coli* in water has previously been reported when associated with rotting vegetation [13]. Enterotoxigenic *E. coli* (ETEC) has also been found to grow in stored water [14]. However, most of the literature would imply that regrowth of *E. coli* is a rare event, in fact this particular bacterium is chosen as a faecal indicator because of its *inability* to grow in waters [15].

In rural Bangladesh [12], a clearer relationship was established between increased environmental temperatures and *E. coli* contamination of weaning foods, and between food contamination and storage time. The proportion of food containing *E. coli* contamination was positively correlated with children's annual incidence of ETEC diarrhoea ($r = 0{\cdot}35$; $P < 0{\cdot}05$) but not all diarrhoea; no such associations resulted from water contamination. Studies in the Gambia also suggest that poor bacteriological quality of infant foods is a significant causal actor in the aetiology of weaning diarrhoea [16]. Explanations for the contamination of freshly cooked food are given as inadequate cooking, poor hygiene or a combination of both. However it is unlikely that *E. coli* could survive the cooking process particularly as the types of foods considered (e.g. rice, maize) should not have temperature penetration problems [17]. It is contended here, therefore, that contamination occurred after cooking by mechanical processes such as contaminated utensils, containers, fingers, or the addition of other foods (e.g. milk, groundnut paste). The positive relationship found between *E. coli* in water and environmental temperature may then be explained by cross-contamination from the immediate environment (e.g. food).

It is well documented that in warm climates many non-faecal coliforms can mimic *E. coli* using the standard methods of detection [18]. Mara and Oragui [19] found in Nigeria that only 28–35% of presumptive *E. coli* from water samples produced indole from tryptone water. Another point of confusion stems from a difference in definition; the recommended procedure used in the UK accepts the need for confirmation tests to show that faecal coliforms are in fact *E. coli*. However, the American definition of faecal coliforms is equivalent to thermotolerant coliforms and thus does not require confirmation tests. This implies that presumptive faecal coliforms in the British procedure are the same as confirmed faecal coliforms in the American standards. It would appear that the studies in Egypt and rural Bangladesh did not use rigorous tests to confirm the presence of *E. coli*.

In the Philippines, Moe and colleagues [20] examined the relationship between bacterial indicators of drinking water quality and childhood diarrhoea. *E. coli* and enterococci were found to be better predictors of risk of disease than presumptive faecal coliforms. However only children drinking grossly contaminated water (> 1000 *E. coli* per 100 ml) had significantly higher rates of diarrhoeal illness than children drinking less contaminated water ($P < 0{\cdot}01$). Although this study used an improved method for the enumeration of *E. coli* [21], it would appear that samples from the water sources were analysed as opposed to the child's drinking cup.

This study compares the bacteriological quality of water stored in rural households in north-east Thailand with the water sources. Fortunately water was stored in separate containers depending on its subsequent use and consequently analysis was able to compare water activities with water quality. At the same time fingertip-rinses were also tested for faecal bacteria to help substantiate where cross-contamination was important. These results are used to develop a hygiene intervention study which is described in a companion paper [22].

MATERIALS AND METHODS

Study outline

Ten households, with children of any age, were arbitrarily selected from a village (Ban Sahart) in north-east Thailand. Each house was visited on five separate occasions during the period October 1987 to May 1988. Information on socio-economic status, water users' practices, water source, knowledge and attitude of diarrhoea transmission was obtained by questionnaire and observation. All water containers were coded on a plan of the household, and all persons present were coded accordingly. On each visit, stored water samples and fingertip-rinses of individuals present were examined for certain faecal bacteria. Research assistants responsible for administering the questionnaire could all speak Lao (dialect of north-east Thailand). Wherever possible responses were checked by direct observation and assistants were trained to reword and repeat questions, particularly if the informant appeared uncertain or the answer seemed unlikely.

Information on the household routine was obtained by general observations and this was compared to the bacteriological results in order to generate hypotheses regarding the causes of faecal contamination. The third and fourth visits to each household took place on consecutive days and an observation chart was used to record the activities of all individuals present. Particular attention was paid to water use, water handling and objects touched by each individual. In addition, a separate chart recorded the use of each water container which included water collection, water-related activities, objects and fingers coming into contact with the water. During this period extensive sampling of stored water and fingertips was conducted, the timing of which was recorded on observation charts and samples coded accordingly.

Bacteriology

Membrane filtration equipment was used on site rather than transporting the water and fingertip-rinse samples to the laboratory; this greatly increased the number of samples taken in one day. Water samples were measured into a sampling jar (100 ml) and media used for fingertip-rinses contained 1/4 strength Ringer's solution supplemented with 0·1% (v/v) Tween 20. Sterile containers contained enough solution to cover the fingertips when a hand was inserted and the subject was told to rub the thumb back and forth across the fingertips for approximately 5 seconds. All samples were divided equally between two membrane filters (0·45 μm pore size), one was placed on pads saturated with Membrane Lauryl Sulphate Broth (SLS; Oxoid MM615) for the enumeration of faecal coliforms, and the other placed on KF streptococcus agar (Oxoid CM701) for the enumeration of faecal streptococci.

All non-sterile equipment was sterilized with acetone before each use, and forceps sterilized over a flame. Periodic checks were made to test the thoroughness of this procedure by sampling the sterile fingertip media. All plates were placed in plastic bags to avoid drying and stored in a dark box. At the end of the day samples were incubated at 44·5 °C: faecal coliforms for 24 h and faecal streptococci for 48 h. The numbers of bacteria were determined following the membrane filtration technique [23]. Periodically, samples from the soak-water of dishes and utensils were taken to the laboratory and serial dilutions prepared before following the procedure outlined above. In addition, some stored water samples (1 litre) were tested for the presence of *Salmonella* spp. using the enrichment-selective method [24]. Isolates were confirmed by slide agglutination tests using polyvalent antisera.

Particular attention was paid to the appearance of the faecal coliform colonies on each plate and those of similar shape, colour and sheen were counted into groups. A selection of colonies from each group were inoculated directly into test tubes containing tryptone water and lactose peptone water. After incubation (24 h) isolates were identified as *E. coli* by their ability to produce indole from tryptophan and gas from lactose at 44 °C [23]. Thermotolerant coliforms were identified by their ability to produce gas at 44 °C. These results were periodically

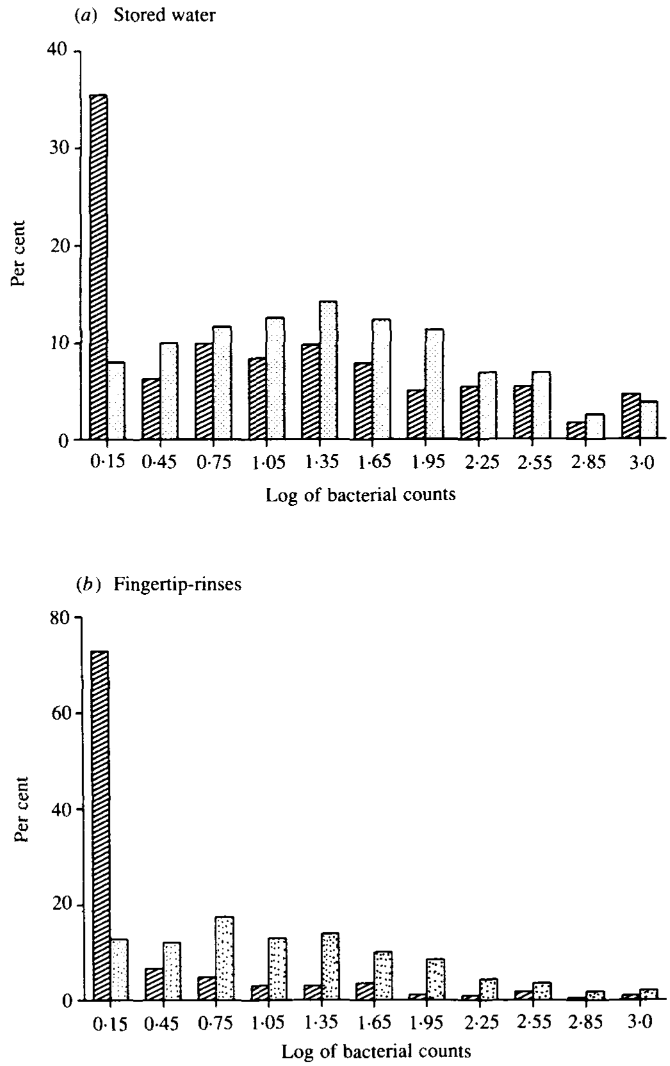

Fig. 1. The numbers ($\log_{10}$) of faecal bacteria in samples of (*a*) stored water (/50 ml) and (*b*) fingertip-rinses. ▨, *E. coli*; ▩, faecal streptococci.

confirmed by the Microbiology Section at Sri Nakorin Hospital, Khon Kaen. *E coli* represented only 48% of the total thermotolerant coliforms; a random selection of colonies similar in appearance to *E. coli* were identified as *Klebsiella* spp. (75%) and *Enterobacter* spp. (25%).

It was previously observed that *Staphylococcus epidermidis* from fingertip-rinses would grow under the same conditions as faecal coliforms [25]. The media (SLS) was modified with Penicillin 'B' (50 mg/l) in order to stop the growth of this organism, without suppressing the growth of faecal coliforms. Experiments designed to test the survival of *E. coli* and faecal streptococci on fingertips were conducted in the laboratory and a measured loop was used to inoculate a known number (range 10^3–10^6) of bacteria. The fingertips were rinsed at different time intervals and bacteria enumerated following the procedure outlined above.

https://doi.org/10.1017/S0950268800047956 Published online by Cambridge University Press

Analysis of data

All data were coded and recorded on a micro computer. Figure 1 presents the frequency distribution of bacterial counts. Although data transformations are often employed in order to normalize irregular distributions, these data sets ($\log_{10}$) still tend to have *non*-normal distributions due to the high frequency of zero counts, especially those of *E. coli*. For this reason non-parametric statistics were employed when analysing bacterial contamination, and it should be noted that these statistics are generally more conservative than their parametric equivalents. The Kruskal–Wallis One-Way Analysis of Variance was used to test whether all the samples are the same population; the Mann–Whitney *U* test was employed when comparing two samples. All statistical comparisons of categorical variables were made using the chi-squared test.

RESULTS

North-east Thailand

The north-east is the poorest region of Thailand and, for the most part, relies on subsistence agricultural production of glutinous rice. Most of the rural population lives in compact settlements of 40 to several hundred households. These villages, shaded by their trees, are usually located on the uplands and form a sharp contrast with the surrounding paddy fields. There are three typical seasons during the year including cool/dry (November to January), hot/dry (February to April) and rains (May to October). The study period began at the end of the rainy season and finished at the start of the next rains.

Baseline data

Socio-economic information for the 10 households from the village Ban Sahart is summarized in Table 1. There was an average of 6·6 persons per household and the median period of adult education was 4 years. Answers to questions regarding the knowledge and attitude to the prevention and cause of diarrhoeal disease revealed that the maternal heads appeared to be fairly well informed on the subject (Table 2). The major deviation from this related to questions regarding child care such as breast-feeding, bottle feeding and contact with children's faeces. Moreover, when asked to prioritize handwashing for five different activities, this revealed following priority; before going out > after toilet > before cooking > before eating > after cleaning a baby's bottom.

Water sources

All the households collected rainwater, and its subsequent availability depended on the storage capacity of jars and the amount of water used. A shallow well (1·5 km outside the village) was the preferred drinking water source when rain was not available. Tubewells were the most reliable water source and many were conveniently located within the village. However, the water from these tubewells, although clear in appearance, often contained dissolved salts which rendered it unacceptable for drinking, thus limiting its use to domestic purposes. Pond water competed with tubewells for the houses nearby and in many cases clothes were washed at the pond. For drinking purposes then, only rain or shallow well water were used due to the 'taste' criterion. Tubewell and pond water were primarily

https://doi.org/10.1017/S0950268800047956 Published online by Cambridge University Press

Table 1. *Socio-economic information for 10 households (HH) studied*

Household ownership of the following	No. of HH	Household sources of income	No. of HH
Electricity	10	Farming	8
Toilet	9	Trader	3
T.V.	7	Labourer	2
Motor bike	6	Salaried job	2
Rotavator	2	Home crafts	6
Pumped water	3	Shop/restaurant	1
Glass windows	2		
Refrigerator	1		

Table 2. *The maternal head's knowledge and attitude to the spread of diarrhoea disease in 10 households*

Cause diarrhoea? Questions	Response			Prevent diarrhoea? Questions	Response		
	Yes	Unsure	No		Yes	Unsure	No
Dirty water	9	0	1	Clean water	9	0	1
Stored cooked food	8	1	1	Washing hands	10	0	0
Flies in food	10	0	0	Clean piped water	10	0	0
Babies faeces	3	5	2	Clean house	9	1	0
Animal faeces	3	6	1	Clean environment	9	1	0
Bottled baby food	0	8	2	Eating well	10	0	0
Clean water	0	0	10	More water	10	0	0
Breastfeeding	8	1	1	Breastfeeding	1	2	7
Boiled water	2	1	7	Dirty water	0	0	10
Exercise	1	3	6	Stored cooked food	1	3	6
Spirits	3	6	1	Bottled baby food	1	8	1
Spicy food	8	0	2	ORS	7	3	0

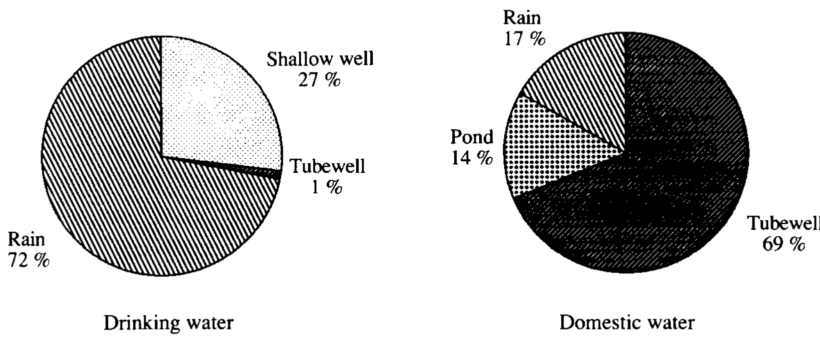

Fig. 2. Water sources by water use of households studied in Ban Sahart.

used for domestic purposes, as was rainwater when available (Fig. 2). Apart from the criteria of water preference, selection of a water source followed a pattern of convenience and availability. Water collection, therefore, required greater time and effort as rainwater became scarce.

Water quality

During the study, it soon became apparent that stored water was often much more contaminated than its water source. In order to quantify this, the bacteriological quality of water from storage containers is compared to their water

Table 3. *Geometric and arithmetic () means of faecal bacteria in samples (50 ml) of stored water and water sources*

Water source	*Escherichia coli* Stored water ($n = 305$)	*Escherichia coli* Water source ($n = 102$)	Faecal streptococci Stored water ($n = 295$)	Faecal streptococci Water source ($n = 102$)
Rain	4·5 (81)	1·3 (7)	22·3 (136)	8·3 (33)
Shallow well	1·9 (7)	8·5 (22)	10·5 (38)	17·0 (26)
Tubewell	25·5 (219)	0·5 (3)	31·2 (119)	1·8 (3)
Pond	11·1 (182)	16·3 (45)	27·7 (110)	11·8 (27)
Average	9·2 (136)	1·8 (11)	23·3 (112)	7·8 (8)
Total bacteria	41435	3300*	33105	5800*

* Estimated contribution of water source to stored water contamination.

Table 4. *Geometric and arithmetic () means of faecal bacteria in stored water samples (/50 ml) by water use*

Water use	No. of samples	*E. coli*	Faecal streptococci
Drinking water (D/W)	72	2·4 (38)	11·5 (46)
D/W & cook	64	2·9 (31)	14·7 (60)
All domestic (W/W)	26	5·4 (101)	24·9 (121)
Wash dishes	51	40·0 (349)	41·6 (195)
Wash clothes	7	1·1 (2)	5·4 (10)
Bathing	32	21·9 (144)	44·7 (189)
Toilet	55	40·7 (228)	49·4 (142)
All	307	9·8 (136)	23·3 (112)
Kruskal–Wallis H statistic:		79·2*	47·8*

* $P < 0·0001$.

sources (weighted according to the frequency of occurrence). The quality of stored water is clearly not a function of quality at source (Table 3). Tubewells usually produced good water quality, except those located next to a toilet, but it was the most contaminated stored water. These figures are conservative as no account is taken of bacterial die-off and it is clear from the shallow well water, that this is a significant factor. Overall there was 41435 *E. coli* and 33105 faecal streptococci found in samples taken from stored water, whereas the estimated contribution from the water sources was only 3300 and 5800 respectively.

All households had a selection of water containers each used for a specific water-related activity. These activities have been categorized into the following: drinking water (D/W), drinking and cooking (D/W & Cook), combined domestic purpose excluding toilet (W/W), washing dishes, washing clothes, bathing and toilet. Table 4 reveals that the bacteriological quality of stored water is a function of water use; the statistic confirms that categories of water use are not from the same population ($P < 0·0001$). Water used for drinking and cooking has relatively little contamination compared to water used for domestic activities. Water for washing clothes was also quite clean, but the sample size is small. As may be expected, water used for anal cleansing was the most contaminated. However a very close second, in terms of *E. coli* contamination, was the water used for washing dishes and cooking-related activities. Shallow well water was used exclusively for drinking and cooking, whereas tubewell water was for domestic purposes and this helps explain the difference in their stored water quality (Table 3).

It is contended that the majority of faecal bacteria are transferred to the stored water from the environment encountered through the water-related activities, by the practice of water handling. Evidence to support this assumption is provided by detailed observation of individual households:

(i) The main method of obtaining water from a container was by use of a 'dipper' (usually a plain metal bowl). The dippers, and the water therein, often come into contact with surfaces, fingers and objects related to a particular water activity. It was observed that water used for drinking was handled in a more hygienic manner than domestic waters. Care was taken to limit the contact between fingers and water when using the dipper; in some cases mugs or wooden ladles were used. The dippers for domestic waters usually floated on the water surface and the user's fingers often came into direct contact when taking water or through activities such as hand rinsing. Any residual water remaining in the dipper after use was returned to the container, but residual water from drinking or cooking was generally thrown away.

(ii) The only containers that were exceptional to the above were rainjars and raintanks fitted with taps. Their levels of water contamination were unaffected by water activity because water was drawn through taps; the sheer size of these containers made it difficult to obtain water any other way. These have been counted as the rainwater source in Table 3 and are examined in more detail elsewhere [26].

Other potential sources of contamination are eliminated as follows. Samples from the plastic containers used for collecting water had similar levels of contamination to that found at source. Waters used for drinking were far more likely to be covered than domestic waters, so it is difficult to calculate the protection awarded by a lid when controlling for water use. However, there were similar levels of contamination for covered (52 cases) and uncovered (18 cases) drinking water with geometric means of 2·4 and 1·5 *E. coli* per 50 ml respectively. Levels of water contamination were also checked by location and type of container (rainjars excluded) but there was no appreciable difference between these factors. The presence of animals did not appear to affect water quality as some containers were located upstairs where no animal resided whereas others, located in the compound, were often in close proximity to animal faeces.

Fingertip contamination

Fingertip-rinses (362 cases) were obtained at the same time as the water samples and the respective geometric means were 0·8 and 13·2 counts for *E. coli* and faecal streptococci respectively. The relatively low numbers of *E. coli* reflect the results from laboratory experiments designed to test the survival time of these bacteria on human skin. In 16 separate tests 99% of *E. coli* died within 10 min and only twice did any organisms survive longer than 30 min; similar findings are reported elsewhere [27, 28]. In contrast, an average of 50% of faecal streptococci died off in the first 30 min while still 10–30% of these organisms remained viable after 2–4 h. These results would imply that *E. coli* is less likely to survive remote cross-contamination and therefore, it should show a closer association with activities conducted immediately before or during water use. In Table 4, there is indeed a greater variation between *E. coli* contamination of stored water by water use (chi-squared = 79·2) than for faecal streptococci (chi-squared = 47·8). Other studies have shown that *Salmonella* spp. and *Shigella* spp. survive longer on the skin than *E. coli* but not as long as faecal streptococci [27, 29]. This implies that cross-contamination via fingertip may well be selective for different pathogens.

Part of the study involved detailed observation and repeated sampling of individuals' fingertips. It soon became apparent that a relationship existed between fingertip contamination and the individual's activities immediately prior to sampling. In order to simplify analysis, these activities have been categorized into five groups:

(i) *Water related activities*: washing dishes, handwashing, water carriage, toilet use, washing clothes, bathing.

(ii) *Food-related activities*: breast-feeding, eating, preparing food, cooking, selling food.

(iii) *Care of infant*: looking after infants or babies.

(iv) *Arrive/Enter*: immediate arrival by either a resident or a visitor.

(v) *Other*: craft activities, sericulture (silkworm breeding), sitting, sleeping, playing.

The variation between these activities and fingertip contamination was highly significant for *E. coli* ($P < 0{\cdot}0001$) but not for faecal streptococci (Table 5). The most contaminated fingertip-rinses were taken from water-related activities and this may be expected as bacteria are susceptible to desiccation. On the one occasion an individual's fingertips were sampled after returning from the toilet, no *E. coli* were found, whereas the only sample taken after washing dishes produced > 200 counts. From food-related activities, cooking returned a geometric mean of 3·3 counts (10 cases), and eating 1·2 (39 cases). The lowest *E. coli* counts were for sitting 0·0 (18 cases), and sleeping 0·07 (11 cases). The Pearson correlation was used to test for association between fingertip contamination and any independent variable that contained interval or ratio measurements. The only significant correlation found was between climatic humidity and both *E. coli* ($r = 0{\cdot}17$; $P < 0{\cdot}01$) and faecal streptococci ($r = 0{\cdot}17$; $P < 0{\cdot}01$).

Soak-water

During the study, as the patterns of contamination became apparent, further attention was focused on providing information on possible sources of the contamination. Observation revealed the common practice of soaking used pots, dishes and utensils in a basin or bowl. Only a small quantity of water was used to soak these utensils and this generally became murky from food particles. This soak-water was found to be grossly contaminated particularly during the hot season. Up to 10^7 faecal indicator bacteria per 1 ml of soak-water were found; 9/10 samples contained $> 10^3$ thermotolerant coliforms, 6/10 contained $> 10^3$ *E. coli* and 8/10 contained $> 10^3$ faecal streptococci. Significant bacterial growth occurred in 3/8 samples after they had been taken to the laboratory. In view of the mechanisms of cross-contamination, described earlier, this helps explain the high levels of contamination found in stored water used for washing dishes.

Table 5. *Geometric and arithmetic () means of faecal bacteria in fingertip-rinses by preceding activity*

Activities	No. of samples	*E. coli*	Faecal streptococci
Water-related	23	2·9 (126)	19·7 (118)
Food-related	53	1·5 (32)	13·8 (85)
Care of infant	17	1·4 (27)	9·7 (47)
Arrive/enter	57	0·2 (3)	7·4 (31)
Other	87	0·2 (1)	11·9 (86)
All	237	0·6 (22)	11·3 (86)
Kruskal–Wallis H statistic:		26·34*	4·3

$P < 0.0001$.

Salmonella in stored water

A selection of water samples used for drinking and washing dishes was tested for the presence of *Salmonella* spp. and this pathogen was found in 13·3% (16/120) of these samples. It was significantly ($P < 0.05$) more common in water used for washing dishes (22%) than that used for drinking (7%). Contamination levels of faecal indicator bacteria for water samples positive to *Salmonella* spp. were significantly higher than others, with *E. coli* bearing a stronger association ($P < 0.01$) than faecal streptococci ($P < 0.05$). This would suggest that *E. coli* is a better indicator of *Salmonella* spp. than faecal streptococci.

DISCUSSION

In north-east Thailand, the quality of domestic waters (not used for drinking) does not directly constitute a danger to health as it is not ingested. In this study indicators of faecal pollution are used to evaluate risk of contamination from practices related to water use and sanitation. Particular attention has been focused on cooking/food-related activities due to the obvious risk of ingestion. Storage of prepared food was practised by all households and the high levels of contamination found in water used for washing dishes was thought particularly dangerous when considering the following:

(i) Although the observed method of washing dishes would dilute the soak-water, it is expected that the washed utensils would still be grossly contaminated; and in some cases dirt and scraps of food particles were still present after washing. The timing of this activity often meant that these utensils did not dry before they were used again.

(ii) The person responsible for washing the dishes usually prepared and served the food, thereby providing a good opportunity for cross-contamination.

(iii) Cooked food, with the exception of glutinous rice, was often stored on a recently washed dish, rather than left in the cooking pot, thus providing the necessary ingredients for possible bacterial growth.

If all water were to be stored in only one vessel, the risk of contamination to drinking water from other water-related activities would be greater and this may help explain conflicting results presented in other studies [7, 9–11]. This implies that for drinking water any improvements to quality at source should be considered in relation to proportion of contamination occurring after water collection. Improvements to water quantity would definitely assist in reducing cross-contamination through water-related activities but it is difficult to do this without introducing expensive piped supplies [6]. Moreover the provision of taps with running water would actually change the behaviour of users as water activities would usually be conducted at the tap. Waste-water would be contained to one area, and the action of running water provides a more hygienic washing action that the practices observed in this study. By way of a comparison, fingertip-rinses were taken from 46 Thai householders who lived in modern houses with in-house water connections. These samples were significantly less contaminated than those taken in Ban Sahart ($P < 0{\cdot}05$); similar findings have been reported elsewhere [25].

Change in water-related practices alone, without improving water quantity may help reduce faeco-oral disease transmission. The results presented here are used to develop a hygiene intervention study which is described in a companion paper. *E. coli* contamination of stored water and fingertip-rinses has been used as the main outcome indicator for the intervention. The advantages of using this rather than diarrhoeal disease are twofold:

(i) Health indicators are inherently difficult to measure due to problems of definition and recall. In addition, indicators such as diarrhoeal disease require a large sample size as it is a relatively rare event.

(ii) Many other activities apart from water supply and sanitation can effect disease (e.g. child rearing practices). These confounding variables are difficult to control where health indicators are used.

Water supply and sanitation initiatives directly effect the ingestion of pathogenic microorganism, so indicators of faecal contamination should be more specific to these activities.

ACKNOWLEDGEMENTS

I am grateful for technical assistance provided by Dr Wanpen Wirojanagud, Dr Chariya Chomvarin, Mr Cho Cha Na, and to Dr Sandy Cairncross and Mr Ed Stentiford for reviewing an earlier draft of this paper. This research was supported by the Science and Engineering Research Council, 85300104.

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