

## Reservoirs of faecal indicator bacteria in well-head hand pumps in Bangladesh

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### ABSTRACT

The majority of the population of Bangladesh (90%) rely on untreated groundwater for drinking and domestic use. At the point of collection, 40% of these supplies are contaminated with faecal indicator bacteria (FIB). Recent studies have disproved the theory that latrines discharging to shallow aquifers are the major contributor to this contamination. In this study, we tested the hypothesis that hand pumps are a reservoir of FIB. We sampled the handle, spout, piston and seal from 19 wells in Arai-hazar Upazila, Bangladesh and identified that the spout and seal were reservoirs of FIB. These findings led to our recommendation that well spouts be regularly cleaned, including the removal of precipitated deposits, and that the seals be regularly changed. It is envisaged that one or both of these interventions will reduce the numbers of FIB in drinking water, thereby reducing the burden of diarrhoeal disease in Bangladesh.

**Key words** | Bangladesh, faecal contamination, groundwater, handpump

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### INTRODUCTION

The human right to water entitles everyone to clean, safe drinking water (United Nations 2010). However, it is estimated that 38% of protected water sources in low- and middle-income countries are contaminated with faecal indicator bacteria (FIB; organisms that indicate that water has been contaminated with human or animal faeces, e.g., *Escherichia*, *Salmonella*, etc.) (Bain *et al.* 2014), while between 35 and 60 million cases of acute gastrointestinal illness result from the consumption of untreated groundwater each year globally (Murphy *et al.* 2017).

Bangladesh is a low- to middle-income country with a population of >160 million and a GDP per capita of \$1,359 (World Bank 2016). Over 90% of the population consumes untreated groundwater and the prevalence of diarrhoea in children under five is 3.9% (BBS/UNICEF

2015). At the point of collection, 40% of water supplies are contaminated with FIB; at the point of use this increases to 60% (van Geen *et al.* 2011; BBS/UNICEF 2015; Ercumen *et al.* 2017). The origin of the contamination has been widely discussed, with most attention being focused on pit latrines that leak into the shallow aquifer (the main source of drinking water); the problem may be exacerbated by Bangladesh's frequent flooding (Graham & Polizzotto 2013). It has recently been shown that leakage from latrines into the shallow aquifer cannot account for the frequency of FIB at the point of collection, and it has been concluded that previous studies have incorrectly equated the quality of collected water with that of groundwater (Ravenscroft *et al.* 2017). Those conclusions are consistent with the results of a recent water quality survey, which showed that FIB counts were positively correlated with poor condition of well bases (Ercumen *et al.* 2017). That study also proposed that 'short-circuiting' occurs at the wellhead, i.e., contaminated water flowing down through a poorly sealed annulus

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(the space between the water pipe and the well wall), priming with contaminated water, or the addition of cow dung during well construction. A positive correlation was also identified between increased FIB numbers and a broken or absent well base (Knappett *et al.* 2012). Short-circuiting may well occur in some instances, but it is also possible that the hand pumps themselves are reservoirs for FIB. Hand pumps from FIB-contaminated wells continue to discharge FIB when removed and transferred to a sterile water source; and sterile hand pumps seeded with *Escherichia coli* continue to discharge bacteria for over three months post-contamination (Ferguson *et al.* 2011). In those seeded pumps, it was the 'elastomeric' components (the seal and piston, which are normally made of plastic and rubber, respectively), in particular, that were reservoirs of the *E. coli* (Ferguson *et al.* 2011).

In this study, we have built on the previous work by examining, *in situ*, the contamination of hand pumps, with the specific aim of identifying whether any specific part acts as a reservoir for FIB.

## METHODS

### Field location

Wells were sampled in January 2018 at two locations in Arahazar Upazila, Dhaka division, Bangladesh, coordinates: 23°47'24.78" N, 90°39'25.96" E and 23°46'.76" N, 90°38'24.70" E. Arahazar is 30 km east of Dhaka where the majority of wells (typically <30 m deep) contain >50 µg/L arsenic (van Geen *et al.* 2003, 2014).

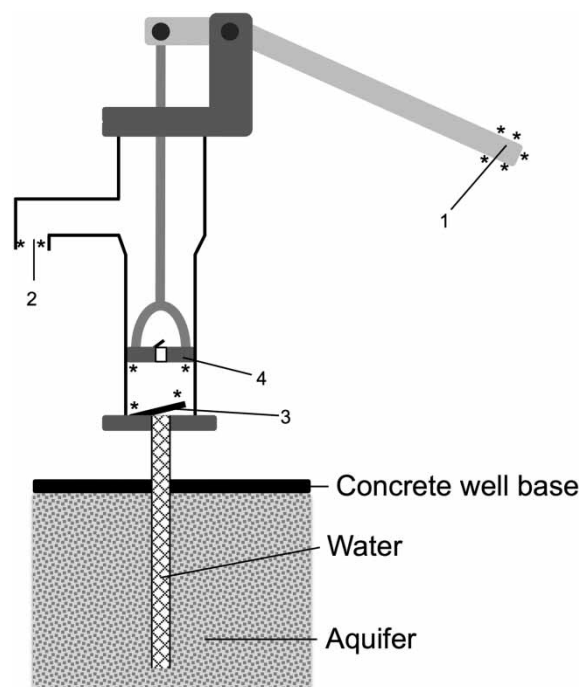
### Sampling

All wells sampled contained Bangladesh No. 6 hand pumps. For pre-screening of wells, 100 mL of water was collected into sterile tubes, filtered through a sterile 0.45 µm filter (Millipore), which was placed onto an eosin-methylene blue (EMB) agar plate (Levine 1918) and incubated overnight at 37 °C. In 19 wells that were positive ( $\geq 1$  FIB per 100 mL sampled) for FIB, samples were taken using sterile swabs from four positions: the handle (upper and lower sides of the handle grip), the inner surface of the spout (not

pre-cleaned), the top edge of the seal at the base of the well, and the edge of the underside of the piston (Figure 1). Each position was swabbed three times. (It was not possible to swab certain parts of the pump, including the rubber part of the piston, without cross contamination.) The swabs were transported to the laboratory at the University of Dhaka and the material suspended in 1 mL sterile phosphate buffered saline (PBS; Sigma). The suspensions were serially diluted and 100 µL of each dilution plated onto EMB agar and incubated overnight at 37 °C. EMB inhibits the growth of Gram-positive bacteria but allows growth of both lactose- and non-lactose-fermenting bacteria. Counts are presented per swab (see Table S1, available with the online version of this paper).

## RESULTS

Table 1 shows the means, standard deviations and variances of the bacterial counts from the four positions on the 19 pumps; and Table 2 shows their medians, first and third



**Figure 1** | Schematic of typical suction mode hand pump. Sampling positions of well-heads. Handle: 1; spout: 2; seal: 3; piston: 4. The handle, spout and bottom of the piston are all made from metal while the seal is made from plastic. The piston assembly comprises a rubber plug, which seals the pump barrel, and a metal base. \*Indicates positions that were swabbed.

**Table 1** | Means, standard deviations and variances of FIB counts from the four positions

	Piston	Handle	Spout	Seal
Mean	161.57	207.89	1,988.94	2,044.73
S.D.	502.76	516.00	4,808.06	7,028.67
VAR	252,768	266,256	23,117,441	49,402,202

Positions are arranged in increasing order of mean count.

**Table 2** | Medians, first and third quartiles, and ranges of FIB counts from the four positions

	Piston	Handle	Spout	Seal
Maximum	2,200	1,720	20,560	31,000
3rd Quartile	70	75	1,345	635
Median	0	0	230	360
1st Quartile	0	0	35	85
Minimum	0	0	0	0

quartiles, and ranges. Because of the large differences in variance and the strongly skewed distributions of the counts, analysis of variance was inappropriate. Instead, data were analysed using a non-parametric method: the Friedman test for randomized blocks (the blocks are the 19 pumps) (Sokal & Rohlf 1995).

The FIB counts differed significantly among positions:  $\chi^2 = 15.93$ , 3 d.f.,  $P < 0.005$ . Table 3 gives the U values for the post-hoc multiple comparisons (Sokal & Rohlf 1995).

The samples from the seal contained significantly more FIB than those from the piston and the handle; those from the spout had significantly higher counts than those from the piston, though not those from the handle ( $U = 262.5$ ,  $0.05 < P < 0.1$ ). Handle and piston samples did not differ from each other, and there was no significant difference between the counts for spout and seal.

**Table 3** | U values for multiple comparisons among the FIB counts from the four positions

	Piston	Handle	Spout	Seal
Piston	–	196.5	<b>268.5*</b>	<b>298.5**</b>
Handle		–	262.5	<b>285.5*</b>
Spout			–	186.5
Seal				–

Values in bold are significant at  $P < 0.05$  (\*) or  $P < 0.01$  (\*\*).

## DISCUSSION AND CONCLUSIONS

The high bacterial counts from seals and spouts, relative to handles and pistons, have implications for possible ways of limiting contamination of drinking water. Since the spout is downstream from both the seal and the piston, it cannot be the original source of bacteria; but they can clearly colonise it from the water passing through the pump, so regular cleaning of the spout may help limit contamination. The seal, however, has at least as high a bacterial load as the spout, and it is upstream so it may be the more important source of FIB. If so, periodic replacement may result in a cleaner water supply.

Further work is needed to test whether replacing the seal, alone or in combination with cleaning of the spout, does indeed reduce bacterial contamination of drinking water, as our results imply. Further sampling of the plug and valve should be conducted to determine other reservoirs of FIB if seal changing and spout cleaning does not reduce bacterial contamination significantly.

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