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

Thomas H. Osborne, Seamus A. Ward, Kazi M. Ahmed and Joanne M. Santini

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 Araihazar 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

Thomas H. Osborne Seamus A. Ward Joanne M. Santini (corresponding author) Institute of Structural and Molecular Biology, Division of Biosciences, University College London, Gower Street, London WC1E 6BT, UK E-mail: j.santini@ucl.ac.uk

Kazi M. Ahmed Geology Department, University of Dhaka, Dhaka 1000, Bangladesh

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

doi: 10.2166/wh.2018.042

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 aguifer (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

This is an Open Access article distributed under the terms of the Creative Commons Attribution Licence (CC BY 4.0), which permits copying, adaptation and redistribution, provided the original work is properly cited (http://creativecommons.org/licenses/by/4.0/).

(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 Araihazar Upazila, Dhaka division, Bangladesh, coordinates: $23^{\circ}47'24.78''$ N, $90^{\circ}39'25.96''$ E and $23^{\circ}46'.76''$ N, $90^{\circ}38'24.70''$ E. Araihazar 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 μ m 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 μ L of each dilution plated onto EMB agar and incubated overnight at 37 °C. EMB inhibits the growth of Grampositive 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





	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	

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

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	268.5*	298.5**
Handle		-	262.5	285.5*
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.

ACKNOWLEDGEMENTS

This work was funded by a BBSRC GCRF Impact Acceleration Award (BB/GCRF-IAA/17/21) to Celia Caulcott (UCL). We thank Celia Caulcott and Mariana Trejo for the funding opportunity. We would like to acknowledge the laboratory of Anowara Begum for technical assistance, and Al Amin Mishu and Riaz Uddin for field assistance.

REFERENCES

- Bain, R., Cronk, R., Wright, J., Yang, H., Slaymaker, T. & Bartram, J. 2014 Faecal contamination of drinking-water in low- and middle-income countries: a systematic review and metaanalysis. *PLoS Medicine* 11 (5), e1001644.
- BBS/UNICEF 2015 Bangladesh Multiple Indicator Cluster Survey 2012-2013, Final Report. Bangladesh Bureau of Statistics and UNICEF Bangladesh.
- Ercumen, A., Naser, A. M., Arnold, B. F., Unicomb, L., Colford, J. M. & Luby, S. P. 2017 Can sanitary inspection surveys predict risk of microbiological contamination of groundwater sources? Evidence from shallow tubewells in rural

Bangladesh. *The American Journal of Tropical Medicine and Hygiene* **96** (3), 561–568.

- Ferguson, A. S., Mailloux, B. J., Ahmed, K. M., van Geen, A., McKay, L. D. & Culligan, P. J. 2011 Hand-pumps as reservoirs for microbial contamination of well water. *Journal of Water and Health* **9** (4), 708.
- Graham, J. P. & Polizzotto, M. L. 2013 Pit latrines and their impacts on groundwater quality: a systematic review. *Environmental Health Perspectives* 121 (5), 521–530.
- Knappett, P. S. K., McKay, L. D., Layton, A., Williams, D. E., Alam, M. J., Mailloux, B. J., Ferguson, A. S., Culligan, P. J., Serre, M. L., Emch, M., Ahmed, K. M., Sayler, G. S. & van Geen, A. 2012 Unsealed tubewells lead to increased faecal contamination of drinking water. *Journal of Water and Health* 10 (4), 565.
- Levine, M. 1918 Differentiation of *B. Coli* and *B. Aerogens* on a simplified eosin-methylene blue agar. *Journal of Infectious Diseases* **23** (1), 43–47.
- Murphy, H. M., Prioleau, M. D., Borchardt, M. A. & Hynds, P. D.
 2017 Review: epidemiological evidence of groundwater contribution to global enteric disease, 1948–2015. *Hydrogeology Journal* 25 (4), 981–1001.
- Ravenscroft, P., Mahmud, Z. H., Islam, M. S., Hossain, A. K. M. Z., Zahid, A., Saha, G. C., Zulfiquar Ali, A. H. M., Islam, K., Cairncross, S., Clemens, J. D. & Islam, M. S. 2077 The public health significance of latrines discharging

to groundwater used for drinking. Water Research 124, 192-201.

- Sokal, R. R. & Rohlf, F. J. 1995 *Biometry*. W.H. Freeman & Co., New York, USA.
- United Nations 2010 *Resolution on Human Right to Water and Sanitation*. United Nations General Assembly. A/64/292. United Nations, New York.
- van Geen, A., Zheng, Y., Versteeg, R., Stute, M., Horneman, A., Dhar, R., Steckler, M., Gelman, A., Small, C., Ahsan, H., Graziano, J. H., Hussain, I. & Ahmed, K. M. 2003 Spatial variability of arsenic in 6000 tube wells in a 25 km² area of Bangladesh. *Water Resources Research* **39** (5), 1140.
- van Geen, A., Ahmed, K. M., Akita, Y., Alam, M. J., Culligan, P. J., Emch, M., Escamilla, V., Feighery, J., Ferguson, A. S., Knappett, P., Layton, A. C., Mailloux, B. J., McKay, L. D., Mey, J. L., Serre, M. L., Streatfield, P. K., Wu, J. & Yunus, M. 2011 Faecal contamination of shallow tubewells in Bangladesh inversely related to arsenic. *Environmental Science and Technology* **45** (4), 1199–1205.
- van Geen, A., Ahmed, E. B., Pitcher, L., Mey, J. L., Ahsan, H., Graziano, J. H. & Ahmed, K. M. 2014 Comparison of two blanket surveys of arsenic in tubewells conducted 12 years apart in a 25 km² area of Bangladesh. Science of the Total Environment 488-489, 484-492.
- World Bank 2018 Data, Bangladesh. https://data.worldbank.org/ country/bangladesh (accessed 16 February 2018).

First received 20 February 2018; accepted in revised form 6 April 2018. Available online 20 April 2018