Nitrate and bacterial contamination in well waters in Vinh Phuc province, Vietnam

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ABSTRACT

Measurements were made of the nitrate concentration and bacterial contamination of groundwater samples taken from both dug wells and bores in the commune of Hop Thinh', in the administrative district of Vinh Phuc, Vietnam. A significant number (18%) of samples had nitrate concentrations in excess of the WHO Guideline value for drinking water of 50 mg L^{-1}, with a higher proportion in the dug wells (29%) than the bores (3.8%). High concentrations of thermotolerant coliforms were found in many of the dug wells and even in the deeper drilled bores. At the time of the study no *Shigella* or other infectious organisms were found. There was no correlation between nitrate concentration and bacterial content and it is concluded that nitrate concentration is not a good indicator for bacterial contamination.

Key words | bacterial contamination, groundwater, nitrate, Vietnam, well water

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INTRODUCTION

Vietnam has a high incidence of bacterial pathogens causing diarrhoea, children being particularly susceptible. A prospective study of 1655 children under five years of age showed Campylobacter, Shigella and enterotoxic *Escherichia coli* to be the most common isolates (Isenbarger *et al.* 2001). The incidence of diarrhoea was shown to be seasonal, increasing in the rainy season from April to September and declining in the cooler, drier months. This had been presumed to be due to surface flooding, which might be expected to spread faecal waste into drinking water supplies. It was shown in that study, however, that this is not necessarily the explanation of seasonality since, in the commune (Phu Chao) most at risk of flooding, there was consistently the lowest incidence of disease. An alternative explanation is required.

In rural regions of Vietnam most people depend for their water supply on wells or, as is increasingly the case in some areas, bores drilled deeper into the aquifer. Extensive work in Indonesia has shown that bacterial counts are often high in village wells, particularly in crowded villages with primitive sanitation (Smith *et al.* 2000). The present work was initiated with the objective of determining whether a possible alternative explanation for the incidence of some diarrhoeal disease in Vietnam is that it is transmitted through the soil into the groundwater, and thereby contaminates water used for drinking, cooking and washing.

There are several studies of the stability of bacteria in soil and water and the topic has been reviewed by Santamaria & Toranzos (2003). It is well known that soil is a reservoir of certain bacterial pathogens and it is known that Shigella, Campylobacter and *Escherichia coli*, in particular, are stable for significant periods in certain situations in both soil and water. Our plan was to investigate nitrate measurements of well water as a possible indicator of contamination with sewage and animal waste, and then to attempt to determine if there was a correlation with bacterial counts. A detailed study in Indonesia showed that, although there was no direct correlation between nitrate concentrations and bacterial counts, in all cases where nitrate levels were high, bacterial numbers were also high (Smith *et al.* 1999). Furthermore, it was shown in that

doi: 10.2166/wh.2008.027

study that nitrate levels varied as the level of the groundwater changed between the dry and rainy seasons. Although a primary objective of the present work was to investigate nitrate concentration as an indicator of bacterial contamination, nitrate itself is a health concern. The World Health Organization (WHO) has a guideline value of 50 mg L^{-1} as the maximum for drinking water (WHO 1993). This is primarily based on its ability to cause methaemoglobinaemia after conversion to nitrite in the gastrointestinal tract (Windle Taylor 1974; Fan & Steinberg 1996), particularly in young children, but it has also been suggested that nitrate is associated with other diseases (Dijk-Looyaard & Montizaan 1990; Fan & Steinberg 1996), including oesophageal (Siddiqi *et al*. 1992), stomach (Forman 1989) and urological (Volkmer *et al*. 2005) cancers.

Location and time of the work

The work was done in the commune of Hop Thinh', divided into the six administrative structures (villages) of Tan Thinh, Le Loi, Quang Trung, Tho Khanh, Hung Thinh and Lac Thinh. This is in the administrative district of Vinh Phuc. The study was done under the auspices of the Centre for Preventative Medicine in Vinh Yen. There is an increasing trend in this region to use water from pipes drilled to greater depths than the dug wells which they are replacing. The study was conducted in June 2005. The region of Vinh Phuc is flat, suggesting slow-moving aquifers. The distribution of wells, boreholes and tap water is shown in Table 1.

Table 1 | The nature of the water supplies in Hop Thinh'

	Water source			
Village	**Dug wells**	**Boreholes**	**Tap water**	**Total**
Tan Thinh	10	170	0	180
Le Loi	8	188	2	198
Quang Trung	6	99	0	105
Tho Khanh	6	168	4	178
Hung Thinh	29	196	2	227
Lac Thinh	10	98	0	108
Total	69	919	8	996

METHODS

General approach

Over a three day period 117 nitrate measurements were made on water that was freshly drawn in buckets or by pumps. Of these, 63 were dug wells and 54 were drilled bores. At the same time, a questionnaire was filled out. It was intended to subsequently collect one litre water samples from all of the dug wells and from 37 of the bores, for bacterial analysis. In practice, 61 of the dug wells and 33 of the bores were sampled. The attrition was due mainly to some house owners not being home at the time that the samples were being collected. Appropriate sample handling and processing procedures were in place to avoid any possible cross contamination between samples.

Nitrate measurement

Nitrate was measured using nitrate test strips with a Nitracheck reflectometer as described previously (Wetselaar *et al*. 1998; Smith *et al*. 1999). Values were calibrated against standard nitrate solutions measured at the same temperature, later in the day (Smith *et al*. 1999).

Questionnaires

The questionnaires were completed at each household and included personal details of all members of the household, the nature and protection of the water source, its distance from latrines and animals and incidences of diarrhoeal sickness in the previous year.

Bacterial analysis

Samples (1 L) of water were taken back to the laboratory in Hanoi and immediately the process of filtration, enrichment and growth on selective media for various bacterial species was begun. Together with the enrichment growth, bio-chemical tests were done to identify specific organisms. These included total and thermotolerant coliforms and, particularly, Shigella.

RESULTS

Incidence of nitrate in wells and bores

In Figures 1(a) and (b) the nitrate concentrations in the water drawn from the dug wells and drilled bores are shown, respectively. Clearly, the concentrations were significantly higher in the former than in the latter, although even in some drilled bores significant concentrations of nitrate were observed, when considered in relation to the WHO Guideline value for drinking water of 50 mg L^{-1}. Of all the water samples, 18% had nitrate concentrations above this concentration. The percentage was 29% for the dug wells and 3.8% for the bores. The depths of wells and bores varied. The depth to the water table of dug wells varied between 0.5 and 7 m with the depth to the bottom of the wells being 9–10 m. The depth of the drilled bores usually varied from 12–18 m, with a few significantly deeper than this.

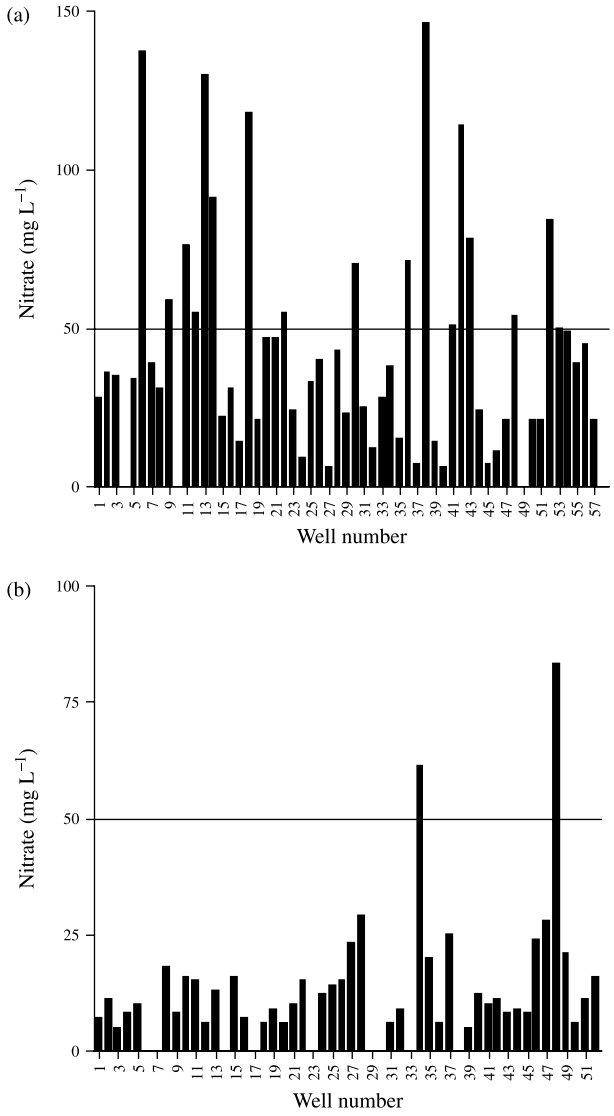

Figure 1 | The nitrate concentrations in the water drawn from the dug wells (a) and drilled bores (b).

Bacterial analysis of water samples

High concentrations of thermotolerant coliforms were found in many of the dug wells and even in the drilled bores. Plots were made of bacterial counts *versus* nitrate concentrations for both dug wells and drilled bores and are shown in Figures 2(a) and (b).

Clearly, there was no overall correlation between nitrate concentrations and bacterial numbers. As with nitrate, bacterial numbers were generally much higher in the dug wells than in the bores, although even in the latter there were two cases of very high coliform numbers; these had depths of 15 and 18 metres. As far as specific bacterial types are concerned, there was no evidence of Shigella or other pathogens in any of the samples.

Questionnaires

The questionnaires revealed that there had been outbreaks of diarrhoeal disease in some households but the cases did not correlate with high coliform numbers at the time they were measured in this study. The questionnaires also revealed the proximity of individual wells and bores to latrines and animals. These are not presented as there were no clear correlations observed and it was not possible to tell whether a well or bore was upstream or downstream of a latrine in the flow of the groundwater. It could be noted that in many cases latrines, and animals, were located very close to wells or bores.

DISCUSSION

The present study has shown that many village wells in the commune which was studied in Vinh Phuc province contained high concentrations of nitrate, often well above

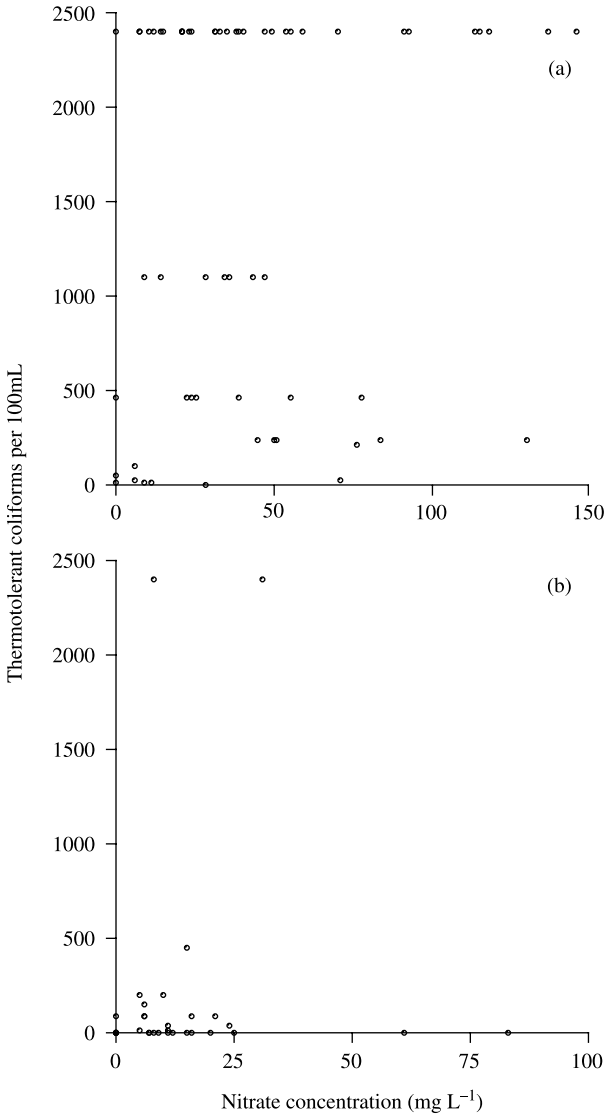

Figure 2 | Numbers of thermotolerant coliforms in the dug wells (a) and in the drilled bores (b). The plots show bacterial counts *versus* nitrate concentration.

the WHO guideline value of 50 mg L^{-1}, and also very high levels of thermotolerant coliforms. As stated in Results, it was found that in many cases latrines, and animals, were located very close to wells or bores, often within a few metres of each other. Although there was no general correlation between nitrate concentrations and coliform numbers, in most cases where nitrate concentrations were at the higher end of the range, bacterial counts were also usually very high. Based on our data, however, it would appear that nitrate measurements are not a good indicator of levels of bacterial contamination. There are several factors which might contribute to this lack of correlation. For example, the study area was very flat and so it was not possible to judge the direction of flow, if any, of the groundwater aquifer. It was therefore not possible to determine the relationship of nitrogen sources to wells in terms of groundwater flow. Also, the mobility, longevity and other behavior patterns of nitrate and bacteria in soil and groundwater are very different. If clear evidence of pathogen contamination of wells were to be obtained, a detailed study of the site, of the type reported in Indonesia (Smith *et al.* 1999), would provide information which would allow assessment of the links between nitrate and bacteria. Such a study would assess the nitrogen and bacterial sources and their strengths, soil and aquifer characteristics, and other factors such as the proximity to wells of nitrogen sources, trees and other vegetation.

The study does show a significant improvement in water quality by the use of water from drilled bores, both in terms of nitrate concentrations and coliform numbers. Nevertheless, there were exceptions in which drilled bores produced water with high nitrate concentrations and high coliform numbers. It could be helpful to advise the relevant house owners of this observation. It is consistent with previous reports of coliforms having been measured at great depth (Santamaria & Toranzos 2003); these earlier reports showed that this situation coincided with the heaviest rainfalls. In this respect it would be useful to repeat the present study in Vinh Phuc again towards the end of the wet season.

Although many people in this area reported that they boil their well water before use, and thereby prevent any risk of infection from contaminated water, nevertheless this study shows that bacteria do in fact contaminate water used for drinking and therefore pose a potential health risk. It could also be mentioned that even if people boil water for drinking, they probably do not do so for washing and this could be a source of contamination. Further work should involve measurement of bacteria in wells at the times of significant outbreaks of diarrhoeal disease.

The present study was conducted in one location and at one time of the year. Based on previous work in Vietnam (Isenbarger *et al.* 2001) and Indonesia (Smith *et al.* 1999), it is likely that there is significant regional and seasonal variability, as well as the local variability also observed in the present study.

CONCLUSION

Significant concentrations of nitrate were found in groundwater from wells in villages in the study area, and in a significant number of cases these exceeded the WHO Guideline value for drinking water of 50 mg L^{-1}. Levels of contamination were significantly different between the dug wells and the deeper bores, although contamination was found in both cases. High levels of thermotolerant coliforms were found in many of the dug wells and even in the drilled bores. Again there was a significant difference between dug wells and bores. A primary aim of the work was to determine whether in a village context, measurements of nitrate concentration might be used as a measure of levels of bacterial contamination. It was shown that there was no direct correlation and hence nitrate measurement would appear to have little general use in this regard. At the time of the study no pathogenic bacteria were found in the water and hence it was not proven whether transmission through the groundwater was likely to be a significant route for infection. It is recommended that measurements be taken in this area, and also in other places where *Shigella, Campylobacter* or *ETEC* outbreaks occur, at the actual times of such outbreaks. We consider it an important public health matter to establish whether or not groundwater is a significant transmission route for such disease agents.

ACKNOWLEDGEMENTS

We are grateful Drs Ho Huu Tho, Bui Tien Sy, Nguyen thi Gam and Nguyen thi Be for collecting the water specimens and doing the bacterial analysis in the laboratory.

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First received 25 July 2006; accepted in revised form 2 April 2007. Available online January 2008