

## Analysis of microbial communities in groundwater of the Kathmandu Valley, Nepal

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In the Kathmandu Valley of Nepal, groundwater is an important water resource for drinking and other domestic uses. Approximately 50% of water supply is derived from the groundwater. However, microbial contamination exceeding the standard for drinking water set by the WHO has been reported. Conventional microbial surveys have mainly focused on *Escherichia coli* and coliform bacteria. There is concern about possibility of underestimating the contamination of disease-causing bacteria except *E. coli* and coliform bacteria. Therefore, we analyzed the microbial communities in the groundwater to detect the other pathogens.

Six water samples were collected from shallow wells and river (five samples for shallow wells and one sample for river). The samples were filtrated with Durapore membranes and DNA extraction was performed from the filters. PCR amplification of the 16S rRNA genes from each DNA sample was carried out with the primer set, EUB8F and EUB907R. The 16S rRNA gene clone library was established and the clonal DNA was sequenced by the dideoxy chain-termination method. The sequence data were compared to those in the GenBank database by using BLAST search program. Moreover, the presence of pathogenic bacteria was detected by nested PCR assay.

Results of phylogenetic analysis of 16S rRNA gene sequences showed that some clones were similar to *Lactobacillus* and *Staphylococcus* that associated with human life. In addition, a total of 57 clones were closely related to nine kinds of pathogenic bacteria such as *Acinetobacter* and *Legionella*. Furthermore, results of nested PCR assay revealed that multidrug-resistant *Acinetobacter* were presented in the shallow well groundwater.

Keywords: Kathmandu Valley, groundwater, microbial analysis, pathogenic microbes, multidrug-resistant *Acinetobacter*