SAFE HOUSEHOLD DRINKING WATER VIA BIOSAND FILTRATION PILOT PROJECT EVALUATION AND FEASIBILITY STUDY OF A BIOSAND PITCHER FILTER

by

Melanie I. Pincus

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Abstract

The author traveled to the Lumbini district of Nepal's southern Terai region to assess the performance of 10 recently installed concrete BioSand filters. Filter performance and source water quality were evaluated using membrane filtration for enumeration of *Escherichia coli* (*E. coli*) and total coliform bacteria, presence/absence tests for hydrogen sulfide (H₂S) producing bacteria, turbidity and flow rate measurements. Filter flow rates varied from 1.0 to 37.5 L/hr; improper sand preparation and filter commissioning for some units may be responsible for the variation observed. Turbidity removal was high for all systems; filters treating highly turbid source water (176.0 – 360.0 NTU) were observed to remove between 98.7 and 99.8% of turbidity. Results from microbial analyses were mixed. Whereas two BioSand filters were removing 99% of *E. coli* from highly contaminated influent water, three were found to be sources of *E. coli* contamination for relatively clean source water. Very poor correlation (38% false negative rate) was observed between H₂S and membrane filtration test results, even in samples with >1000 cfu *E. coli*/100 mL. Winter temperatures of approximately 10°C (50°F) were thought to significantly decrease the accuracy of H₂S tests as detectors of fecal contamination in drinking water.

The BioSand pitcher filter was conceptualized during field investigations as a smaller, cheaper alternative to the concrete BioSand filters. Pitcher filters might also potentially serve as bench-scale models of the larger BioSand filters. Field and laboratory experiments were performed to conduct a preliminary evaluation of pitcher filter viability by cross-checking their performance with the concurrent performance of concrete and plastic Davnor BioSand filters. Microbial removal performance of experimental pitcher filters was comparable to the existing BioSand filters. *E. coli* removal efficiencies of two field pitcher filters. Laboratory pitcher filters ripened with *E. coli* spiked Charles River water for 28 days, then challenged with dilute wastewater, showed removal efficiencies of 97%, as compared to 95% for a plastic BioSand filter. A strong correlation was observed between biofilm maturation periods and source water quality; lower quality influent water facilitated biofilm ripening.

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1 Introduction

1.1 Access to Safe Water

Access to safe water is a basic human right that has been denied to a large proportion of the world's population. Only 0.7% of the world's water supply is available for consumption and, unfortunately, it is disproportionately distributed. Over one half of the people living in developing countries suffer from diseases related to unsafe water supply and sanitation (WHO, 1996a). At the beginning of 2000 one-sixth (1.1 billion people) of the world's population was without access to improved water supply (UNICEF, 2002). The majority of these people live in Asia and Africa, where fewer than one-half of all Asians have access to improved sanitation and two out of five Africans lack improved water supply. These figures are all the more shocking because they reflect the results of at least twenty years of concerted effort and publicity to improve coverage (WHO, 2000).

The use of polluted waters for drinking and bathing is one of the principal pathways for infection by diseases that kill millions and sicken more than a billion people each year (World Bank, 1992). Unsafe water is implicated in many cases of diarrheal diseases. Approximately 4 billion cases of diarrhea each year cause 2.2 million deaths, mostly among children under the age of five. This is equivalent to one child dying every 15 seconds, or 20 jumbo jets crashing every day. These deaths represent approximately 15% of all child deaths under the age of five in developing countries (WHO, 2000).

The most widespread contamination of water is from disease-bearing human wastes, usually detected by measuring fecal coliform levels. Human wastes pose great health risks for the many people who are compelled to drink and wash in untreated water from rivers and ponds (World Bank, 1992). Fecal contamination of source and treated water is further exacerbated by increasing populations, urban growth and expansion, peri-urban settlement and continued and increasing pollutant transport into ground and surface water due to deforestation, global climate change, recurrent disastrous weather events (hurricanes, cyclones, floods, tsunamis, etc.) and increasing coverage of the earth's surface with impervious materials (Sobsey, 2002).

1.2 Improving Water Supply

Current estimates of the number of people using microbiologically unsafe water are probably low. This is because the assumptions about the safety or quality of water based on its source, extent of treatment or consumer handling do not take into consideration several well-documented problems. One problem is that so-called protected or improved sources, such as boreholes and treated urban supplies, can still be fecally contaminated and deliver microbially unsafe water. In some cities the water systems abstract unsafe water from unprotected or contaminated sources and deliver it to consumers with no or inadequate treatment, yet these water systems are classified or categorized as improved and safe.

Another problem contributing to the underestimation of the population served by unsafe water is contamination of water during distribution whether water is piped or carried into the home. Many communities have protected or improved water supplies and treated water that is microbiologically safe when collected or when it leaves a treatment plant. However, substandard water distribution systems, intermittent water pressure due to power outages and other disruptions, and illegal connections to the distribution system often lead to the introduction of fecal contamination and therefore, microbiologically contaminated water at the consumer's tap or collection point (Sobsey, 2002).

There is now conclusive evidence that simple, acceptable, low-cost interventions at the household and community level are capable of dramatically improving the microbial quality of household water and reducing the risks of diarrheal disease and death in populations of all ages in the developed and developing world (Sobsey, 2002). Simply, point-of-use technologies are in demand; attendees of the Second International Women and Water Conference in Nepal in 1998 (peasant women who had traveled, in some instances, 2-3 days on foot, to attend) asked presenters for simple, inexpensive household-scale water filters (Murcott, 2003). These women couldn't trust their government to provide them with a stable, long-term source of safe drinking water, nor

could they trust their neighbors to properly maintain distribution systems (i.e., keep local pumps and spigots clean). They realized that their best option was a technology to treat water as close to the point of consumption as possible.

1.3 Safe Household Drinking Water via BioSand Filtration

In response to these requests, beginning in 1999 Master of Engineering students from the Massachusetts Institute of Technology (MIT) Nepal Water Project have conducted ongoing investigations in Nepal and in Cambridge, Massachusetts, USA to determine appropriate point-of-use water treatment technologies. Building on previous MIT Nepal Water Project studies, ceramic water filters, solar disinfection and arsenic treatment technologies were studied in the 2002-2003 academic year (see Dies (2003); Flores (2003) and Tabbal (2003), respectively). One system in particular, the BioSand filter, seemed promising and was recommended as meriting further investigation. Ensuing field and laboratory investigations showed that the BioSand filter may be an appropriate technology for providing households in remote villages with safe drinking water. System strengths include simplicity, effectiveness, economic sustainability, social acceptability, and reliance on local resources. In addition, the higher flow rates of BioSand filters (20-40 L/hr¹; as compared to ceramic filters [1-4 L/hr, see Dies (2003)], for example), are better suited to meeting basic water quantity requirements for health.²

In January 2002, a BioSand pilot project was initiated by Lee Hersh and Susan Murcott in the Lumbini district of southern Nepal. Ten concrete BioSand filters were constructed and distributed to households in the region, and brief tutorials on filter operation and maintenance provided.

¹ The design flow rate for a concrete BioSand filter with an area of about 0.3 m by 0.3 m is 1 L/min (60 L/hr) when the top reservoir is full of water. As the water level in the reservoir drops, the flow rate will also drop because there is less hydrostatic pressure through the filter. Average flow rates of filters in the field are generally closer to 30 L/hr (Ron Lentz, 2003).

² Based on estimates of requirements of lactating women who engage in moderate physical activity in above-average temperatures, a minimum of 7.5 litres per capita per day will meet the requirements of most people under most conditions. This water does not account for health and well-being-related demands outside normal domestic use such as water in health care facilities, food production, economic activity or amenity use (Howard and Bartram, 2003).

The author traveled to Nepal in January 2003 to evaluate the performance of the newly installed BioSand filters. At the request of the town doctor, a brief survey of 21 Lumbini district hand pumps was also performed. Field experiments on two concrete BioSand filters were conducted to elucidate biofilm maturation rates and bacterial removal efficiencies. The results of these experiments were compared to subsequent laboratory experiments at MIT, using a plastic BioSand filter (Davnor filter).

The BioSand pitcher filter (pitcher filter) was conceptualized during field investigations in response to observed drawbacks of concrete BioSand filters. Two prototypes were constructed using locally obtained materials and tested over a 4-day period. Laboratory experiments at MIT were also conducted to supplement field data and evaluate pitcher filter viability as (a) bench-scale models of the full-size filters and (b) a new household drinking water technology.

1.4 Research Objectives

- To evaluate the technical performance of the concrete BioSand filters installed in Nepal's Lumbini district.
- To elucidate biofilm maturation rates and bacterial removal efficiencies of BioSand filters.
- To develop an improved BioSand filtration system.
 - To investigate the potential of a smaller filter unit as a laboratory model of the full-size filter.
 - To assess the feasibility of the new filter unit as an alternative household water filter.
- To assess the validity of the H₂S method for detecting fecal contamination in drinking water in colder climates.

2 **BioSand Filtration Overview**

2.1 BioSand Filter Overview

In 1987, when Dr. David Manz of the University of Calgary, Alberta, flew to the Zulu homeland in South Africa as part of an international development project, he found himself in a world of perpetual illness, high infant mortality and rampant fatalism (Pearce-McLeay, 1996). International aid organizations had come and gone, leaving in their wake scattered springs, bored holes and instructions to boil or chlorinate the community water supply (University Technologies International, 1998). Driven by the desire to help the developing world find a better way to purify drinking water, Manz spent the next few years developing a simple, cheap and effective filtration system (Legge, 1996). The result of these investigations was the BioSand Filter, an intermittently operated slow sand filter specifically designed for use by poor people in developing countries.

The BioSand filter is an example of a granular bed filter, typified by a substantial depth of sand as the primary filter media (see Figures 2.1 and 2.3). The filter may also be described by the hydraulic arrangement employed to pass water through the medium, as well as the rate of filtration, i.e., the flow rate per unit area. Specifically, the BioSand filter is a gravity filter – open to the atmosphere with gravity facilitating flow through the medium (Water Quality & Treatment, 1999). Particle removal occurs both at depth (i.e., within the granular material) and at the surface of the filter media (see section 2.2).

2.1.1 Critical Design Parameters of the BioSand Filter

Dr. Manz's BioSand filter is a scaled-down version of an industrial slow sand filter, and optimized for intermittent, household use. Filter design incorporates two key modifications to traditional slow sand filtration technology (see section 2.1.1). The first is a faster loading rate of 0.6 m/h (flow rate of 20-40 L/hr for a 0.3 m x 0.3 m concrete unit) as compared to traditional slow sand filtration rates of 0.1 to 0.2 m/hr. The second

key design parameter is a 5 cm layer of standing water, sufficient to allow adequate oxygen diffusion to the biological layer during pause periods.³

The depth of the supernatant (standing water reservoir) is based on research conducted by Buzunis (1995) to determine the head height at which the aqueous microbial community receives the maximum oxygen while still being protected from incoming water. Because the filter is aerobic, oxygen is required to break down and destroy organic contaminants and pathogens. Bacterivores in the supernatant also require oxygen for metabolism (see below). Thus the limiting factor in a filter which is operating intermittently is the amount of oxygen available for metabolism during the paused or stopped period. For the system to remain aerobic, the rate of oxygen use must not exceed the rate of diffusion into the supernatant. A more shallow water depth is preferred since the two prototypes studied having water depths 2.5 cm and 5 cm and filters installed with a 5 cm or smaller standing water depth in Nicaragua appeared to result in higher average removal rates than the 12.5 cm intensely studied filter, which was the subject of Buzunis' Master's Thesis (Buzunis, 1995).

2.1.2 Slow Sand Filters

The fundamental operating principles of the BioSand filter originate in slow sand filtration technology. Figure 2.1 shows a simple slow sand filter (Skinner and Shaw, 1990). In slow sand filtration, the water passes slowly downwards through a bed of fine sand. Pathogenic organisms are mainly filtered out in the very top layer of the filter bed where a biological film accumulates; larger particles are removed via physical and chemical processes within the sand bed (see section 2.2).

³ Dr. Manz's patent is for a "Slow Sand Filter for use with Intermittently Flowing Water Supply and Method of use thereof." Supernatant depth is specified at 1 - 8 cm above the top of the surface sand layer (Manz, 1993).



Figure 2.1 A simple slow sand filter (Skinner and Shaw, 1990).

The reader is referred to Lee (2001) and Lukacs (2002) for an overview of slow sand filtration technology.

2.1.3 Davnor Plastic BioSand Filter

A plastic version of the BioSand filter (Davnor filter; marketed as a 6 L capacity, 20 L/hr water filter, see Figure 2.2) is produced by Davnor Water Treatment Technologies, Ltd. (Davnor, 2003), the Alberta-based company started by Dr. Manz to market his technology. Davnor produces several types of plastic BioSand filters – manual and automated – of varying sizes, flow rates, holding capacity, etc.



Figure 2.2 Davnor plastic BioSand filter (right) with filter media (left; Davnor, 2003).

2.1.4 CAWST Concrete BioSand Filter

The concrete BioSand filter (concrete shell) is a modification of the 20 L/hr Davnor plastic unit and distributed by CAWST (Centre for Affordable Water and Sanitation Technology), the humanitarian arm of Davnor. Figure 2.3 shows a cross-sectional view of a typical square-based concrete BioSand filter (0.3 x 0.3 x 0.9 m). A 2 inch (5 cm) layer of gravel covers the intake pipe and supports a 2 inch (5 cm) layer of coarse sand. Primary filter media is an 18 inch (46 cm) layer of medium-fine sand, which is covered by a 2 inch (5 cm) layer of standing water when the filter is at rest. Filter design includes a diffuser plate to block input water from disturbing the top layer of sand.



Figure 2.3 Cross-section of a concrete BioSand filter (Ritenour, 1998).

2.1.5 BioSand Pitcher Filter

The BioSand pitcher filter was conceptualized during field investigations as a smaller, cheaper alternative to the concrete Biosand filters. Figure 2.4 shows a picture and cross-sectional view of BioSand pitcher filters. Pitcher filter design includes the established 5 cm (2 inch) layer of standing water at rest; filter capacity is approximately 0.5 L.



Figure 2.4 BioSand pitcher filters.

2.2 BioSand Filter Particle Removal Mechanisms

The BioSand filter relies on natural biological, chemical and physical processes to purify raw water. The 5 cm layer of standing water supports a microbial community at the surface of the sand layer; this diverse ecosystem consists of algae, bacteria, protozoa, and small invertebrates, which are both free and attached to biofilm communities that form on the surface sand layer and sand grains (Huisman and Wood, 1974). The biofilm is derived initially from the biology in the raw water and is subsequently sustained by the organic matter in the raw water (Ritenour, 1998). Biologically mediated mechanisms, together with physical-chemical mechanisms, account for removal of particles smaller than about 2 μ m in diameter (Weber-Shirk and Dick, 1997). As influent water penetrates the standing water reservoir, motile predators either living in the supernatant or in the sand surface travel upward due to the new more abundant food source. Many fecal indicator organisms and pathogens will be consumed here (Buzunis, 1995). Predation by protozoa has been identified as the principle biological removal mechanism of harmful bacteria in source water.⁴ Physical-chemical removal processes include straining (of particles greater than about 2 μ m in diameter) and attachment via intermolecular forces between the sand grain surfaces and dissolved and/or suspended particles.

2.3 **BioFilm Maturation**

Newly installed or recently cleaned BioSand filters do not effectively remove bacteria. Bacterial removal efficiency depends on biolayer "ripeness." Ripening refers to the time necessary for the biological community or biofilm to mature such that optimal bacterial and particle removal is attained. Initially, filter performance is based solely on the physical-chemical removal mechanisms of the sand media and flowing water. Over time, particulate and organic matter settles on the solid surface resulting in system head loss and increased removal of turbidity and microorganisms. Dissolved organic carbon, dissolved oxygen, and nutrients present in the influent water support elevated biological populations within the biofilm which further enhance microbial removal efficiency (Collins et al., 1992).

Initial ripening time of a new slow sand filter is approximately 1-3 weeks (Huisman and Wood, 1974). Ripening times of intermittently operated slow sand filters (BioSand filters) are of similar duration: "The filter may require up to two or three weeks to reach

⁴ Weber-Shirk and Dick (1997) studied particle and E. coli removal mechanisms in slow sand filters. Introduction of sodium azide (an inhibitor of oxidative phosphorylation) was found to cause appreciable reduction in particle and E. coli removal, indicating biological removal mechanisms to be significant. Bacterivory was identified as the biological mechanism principally responsible for bacteria removal in a later study (Weber-Shirk and Dick, 1998).

optimum removal of bacteria, viruses and protozoa; complete removal of parasites (parasitic worms) can be expected immediately" (Ritenour, 1998). The target bacterial removal rate upon installation (before the biofilm has ripened) is 70% (Ritenour, 1998).⁵ Removal rates between 93 to 99% of fecal coliform bacteria in matured BioSand filters have been demonstrated (see section 2.4).

Filters with very high-quality source water may not achieve efficient particle removal because of lack of physical-chemical or biological ripening. Physical-chemical ripening is related to previously removed particles and is less effective when the concentration of particles in the raw water is very low (Weber-Shirk and Dick, 1997). That is, turbidity removal increases as particulate and organic matter settle and accumulate on the sand surface. Biological ripening caused by bacterivores is dependent on the concentrations of bacteria in the raw water. Raw water with low concentrations of bacteria may not achieve significant biological ripening (Weber-Shirk and Dick, 1997).

In summary, ripening periods can be greatly reduced (or biofilm growth accelerated) by the presence of organic substances in the raw water. Surface water generally contains higher levels of organic matter than groundwater, and will likely shorten ripening periods where it is used as raw water (see section 6.4.1). Conversely, surface water is likely to have elevated levels of coliform bacteria as compared to ground water; this increased contamination may decrease removal efficiency.

2.4 BioSand Filter Effectiveness

According to Davnor (2003), the BioSand filter effectively removes giardia cysts, cryptosporidia oocysts, water-borne parasites, bacteria, viruses, iron (and iron bacteria), manganese, sulphur smell and other obnoxious odors, color, poor taste, and small particles (silt, clay and organic materials) from source waters. Previous studies demonstrate the effectiveness of BioSand filters in purifying source waters. In initial tests on concrete BioSand systems run by the Public Health Laboratories at Calgary's

⁵ Ritenour (1998) used fecal coliform bacteria as indicator organisms for microbial removal evaluations.

Foothill Hospital in the fall of 1988, the filter eliminated 99 percent of influent fecal coliform (Buzunis, 1993).⁶ Buzunis (1995) found more than 96% reduction of fecal coliform indicators and turbidity reductions to less than 1 NTU.⁷ It is expected that this process behaves in a similar way to continuously operated slow sand filtration and will remove very high proportions of pathogens, including parasites, cysts, viruses, bacteria and cerarcaie (Buzunis, 1995). Laboratory investigations at MIT have found even higher microbial removal rates. Lee (2001) found that the plastic Davnor filter removes 99.5% of total coliform bacteria in raw water.

In a study of 56 concrete BioSand filters operating in Valle Menier, Nicaragua, average fecal coliform removal rates of 97%, ranging from a low of 86% to a high of 99%, were observed. Some of the water sources within this community contained contamination in the range of 10,000 fecal CFU/100 mL (Manz and Buzunis, 1995). Samaritan's Purse Canada (2002) conducted a comprehensive evaluation of 100 BioSand filters in Kenya, Mozambique, Cambodia, Vietnam, Honduras and Nicaragua. Average fecal coliform removal rate for filters in the field was 93%.

Not all studies have found high microbial removal performance, however. One study by the Environmental and Public Health Organization (ENPHO) of Katmandu, Nepal, found microbial removal efficiencies as low as 67%.⁸

⁶ Buzunis (1993) studied fecal coliform removals in concrete BioSand filters with 5 cm supernatant and 20 cm sand bed depth.

⁷ Buzunis (1995) studied fecal coliform removal in concrete BioSand filters with 12.5 cm supernatant and 35 cm sand bed depth.

⁸ ENPHO conducted a 5-month performance trial of two concrete BioSand filters and one TERAFIL filter.

3 Technical Evaluation Criteria for BioSand Filter Microbial Removal Efficiency

3.1 Bacterial Indicators of Fecal Contamination

The ability to test drinking water for fecal contamination is a powerful tool and can encourage local participation in the provision of safe drinking water and in the oversight or monitoring of its provision by other responsible parties such as governments, privatized water companies, water supply contractors, water vendors, etc. (Sobsey and Pfaender, 2002). Testing directly for bacterial pathogens is impractical for many reasons, not the least of which is the need for lengthy and involved test procedures, expensive laboratory equipment, and highly trained technical operators. It has become customary to use *indicator organisms* instead. These are bacteria, usually not pathogenic, that are present when the pathogens are present and absent when the pathogens are absent. Indicator organisms should be of fecal origin as well.

The current criteria of an ideal or preferred indicator of fecal contamination have been defined and stated by the World Health Organization (WHO) and other authorities. According to these authorities, the essential criteria of a fecal indicator are the following (WHO, 2002 in Sobsey and Pfaender, 2002):

- The indicator should be absent in unpolluted water and present when the source of pathogenic organisms of concern (fecal contamination) is present.
- The indicator should be present in greater numbers than the pathogenic microorganisms.
- The indicator should respond to natural environmental conditions and water treatment processes in a manner similar to the pathogens of concern.
- The indicator should be easy to isolate, identify and enumerate.

- The test should be inexpensive, thereby permitting numerous analyses to be taken.
- The indicator should not be a pathogenic microorganism (to minimize the health risk to analysts).
- The indicator should not multiply in the environment.

The rationale for this last criterion is that the presence and concentration of fecal indicators should be in proportion to the level of fecal contamination. Hence, microbial proliferation in the environment could result in the microbe being present at high concentrations when no fecal contamination (and its associated pathogens) or very low levels of fecal contamination are actually present (Sobsey and Pfaender, 2002).

The use of bacterial indicators to detect fecal contamination has its limitations. No one organism or group of organisms satisfies all of the criteria for an ideal indicator. For example, in temperate climates, total coliform bacteria are commonly used as indicator organisms in potable water supplies. In many tropical climates, however, indigenous *E. coli* are present in pristine water sources where no fecal contamination exists; yet they will produce positive results in total coliform tests (Lisle, 1993). Conversely, it has been well documented that waters considered bacteriologically safe (less than 1 bacterial fecal indicator per 100 mL) can contain sufficient pathogenic enteric viruses and protozoans to cause disease outbreaks (Berry and Noton, 1976; Craun and Gunn, 1979; and MacKenzie et al., 1994).

Other fecal indicator microbes, such as enterocooci, spores of *Clostridium perfringens* and coliphages, can be detected in drinking water when the usual coliform bacteria (total or thermotolerant) or *E. coli* are not detectable. Furthermore, there is some evidence that coliforms possibly including *E. coli* can proliferate in tropical and subtropical waters. Warmer water temperatures may contribute to the growth of coliforms, thermotolerant coliforms and *E. coli* and the greater survival of some enteric bacteria, notably

Salmonella (Hazen, 1988; Iverson and Fleay, 1991; Jimenez et al., 1989; Townsend, 1992 all in Sobsey and Pfaender, 2002). The latter constraint, however, was not likely to have confounded test results obtained during an unusually cold Nepali winter in the Terai, where the temperature ranged from approximately 5-10°C.

3.2 Detecting and Quantifying Fecal Contamination

Several types of methods may be used to detect and quantify fecal contamination. The H_2S test for drinking water contamination is based on the presence (P) or absence (A) of the microbial indicator in a specified volume of water, a so-called P/A test.⁹ According to some standards and guidelines, the H_2S indicator is expected or required to be absent in all of (zero tolerance) or most of (e.g., 95%) the sample volumes successively tested over time (see, for example, drinking water guidelines for bacteriological quality in WHO, 1996). Alternatively, the water is analyzed for the fecal indicator microbe or microbe group by an enumerative method (membrane filtration) in which the concentration of bacteria per unit volume can be expressed as colony forming units (cfu) per unit volume (Sobsey and Pfaender, 2002).

3.3 H₂S Test Overview

 H_2S tests were originally developed by Manja et al. (1982) as a simple field alternative to enumerative methods of detecting pollution in drinking water. The tests detect the presence or absence of hydrogen sulfide produced by so-called H_2S producing bacteria. These bacteria are generally found in high concentrations in human and animal feces and often associated with coliform bacteria in fecal contaminated drinking water.¹⁰ The tests do not measure the presence of either total coliform bacteria, specific groups of fecal

 $^{^{9}}$ The H₂S test may also be conducted as a Most Probable Number (MPN) test. See Hwang (2002) for field studies in Nicaragua using H₂S MPN assays.

¹⁰ Many bacteria are capable of producing hydrogen sulfide from organic materials. Some of these are unique to or strongly associated with fecal contamination and many others are not. A major group of environmental bacteria producing H_2S is the sulfate reducing bacteria group. These bacteria are ubiquitous and occur in a variety of habitats, including marine and freshwaters and their sediments, soils, biofilms, microbial mats, intestinal contents, termite guts, walls of "black smokers" and in association with marine worms (Sobsey and Pfaender, 2002). Human feces contain high concentrations of sulfate reducing bacteria, which can be as high as up to $10^{10}/g$ (Levett, 1993).

bacteria (e.g., fecal coliforms) or a specific fecal bacterium (*E. coli*); rather, the test is based on measuring bacteria that produce hydrogen sulfide under the test conditions employed (Sobsey and Pfaender, 2002). The presence of H_2S producing coliform bacteria (e.g., *Citrobacter* spp.) and some enteric bacteria (e.g., *Clostridium perfringens*) associated with fecal contamination may be detected by the H_2S method.



Figure 3.1 H₂S test vials and powdered media packets. Vials with black precipitate indicate presence of H₂S producing bacteria (HACH, 2003).

Test results are based on the observable formation (or lack thereof) of a black iron sulfide precipitate, which forms when H_2S gas produced by the microorganisms reacts with iron in the test media in powdered or paper strip form (Figure 3.1).

3.4 H₂S Test Viability

Sobsey and Pfaender (2002) investigated the validity and reliability of H_2S tests to detect and quantify fecal contamination in drinking water. They reported no expert judgement or analysis to have contributed to the development of H_2S tests as a P/A test:

Instead, P/A H_2S test results were compared to those of other fecal indicator tests to determine the extent of correlation. The use of this comparative approach has never been subject to review of its scientific merit and validity. Considering the differences in the target bacteria being detected, absent any consideration of pathogen presence in water, and without formal efforts to determine how well they fulfill the essential criteria of an ideal or acceptable indicator of fecal contamination (see section 3.1), the validity of H_2S tests, the meaning and reliability of interpretation of their results, and their ability to predict microbial health risks is a matter of concern (Sobsey and Pfaender, 2002).

Nonetheless, Sobsey and Pfaender did cite numerous studies of the H₂S method which reported good correlation between H₂S test results and those from other bacterial indicator tests for fecal contamination (Ratto et al., 1989; Kromoredjo and Fujioka, 1991; Kaspar et al., 1992; Castillo et al., 1994; Venkobachar et al., 1994; Martins et al., 1997; Rijal and Fujioka, 1998; and Genthe and Franck, 1999). See also Grant and Ziel, 1996; Hewison et al., 1988; Sivaborvorn and Dutka, 1989 all in Pillai et al., 1999. They recommend the H₂S method as a reasonable approach for determining the suitability of drinking water with respect to fecal contamination, but caution the following:

- In general, the use of bacterial indicators has its limitations; water systems should be evaluated on a site-specific basis in order to best gauge which microorganisms are appropriate for detecting the presence of fecal contamination.
- Because no systematic efforts have been made to determine if H_2S tests fulfill the essential criteria for an indicator of fecal contamination in drinking water, it is not possible to unequivocally recommend the method for said purpose.
- The H₂S method has yet to be adequately tested in temperate and cold climates (see sections 5.5 and 6.3.2).
- False positive results may be obtained when water samples contain hydrogen sulfide from sources other than fecal contamination (e.g., abiotic chemical reactions, reduction reactions by some enteric bacteria, etc.).
- The limitations of P/A testing should be considered. P/A testing was developed for and is applicable where most tests provide a negative result (Sobsey and Pfaender, 2002). Where a significant number of microbial tests indicate contamination to be present, quantitative testing is preferable in order to more precisely quantify health risks. The reader is referred to Hwang (2003) for a study of the H₂S Most Probable Number test.

3.5 Temperature Dependence of H₂S Tests

Pillai et al. (1999) investigated temperature effects on the reliability of H_2S tests to accurately detect fecal contamination in drinking water. Specifically, they studied the influence of temperature on the incubation period, and whether the contamination level has any influence on the incubation period. The incubation period for H_2S test bottles was found to be highly dependent on incubation temperatures. An 18-hour incubation period was required for samples stored at 37°C. In contrast, no iron sulfide precipitate formation was observed for samples incubated at 0, 8, 14 and 47°C (5 day testing period, with fecal coliform concentrations varying from 1 to >1000 cfu/100 mL). Samples stored at 28°C and 22°C required 36 and 90 hour incubation periods, respectively. In general, H_2S tests did not require constant temperature incubation if the room temperature was between 20 and 44 °C. These results suggest the applicability of H_2S tests in detecting the presence of fecal contamination in tropical climates.

An increase in the incubation period was necessary with the lowering of fecal coliform concentrations at all temperatures. This showed that the growth of H_2S producers was slowed down at those temperatures and the H_2S production was delayed (Pillai et al., 1999).

3.6 Using H₂S Tests to Detect Fecal Pollution in Drinking Water

The requirements for laboratory resources or field analysis kits for standard bacteriological tests for fecal contamination of drinking water are major barriers to their accessibility in many parts of the world. The need for sterilized bacteriological materials (media, sample bottles, sterile diluent, culture tubes, bottle or plates, membrane filters, pipettes or other volumetric dispensing devices, etc), controlled temperature incubators, the required use of aseptic technique by trained individuals, and relatively high costs make it difficult, impractical or impossible to perform these tests in many places. Need for a rapid, simple, inexpensive test for the microbial quality of drinking water is especially great for small community and household water supplies that lack access to and cannot afford conventional bacteriological testing of drinking water. On-site testing

using portable equipment and use of simplified tests, such as the H_2S tests, may both contribute to overcoming these constraints (Sobsey and Pfaender, 2002).

Because of their ease of application and interpretation, the author performed presence/absence microbial testing during field investigations of source waters and BioSand filters in Nepal. Specifically, presence/absence tests for H₂S producing bacteria were used to evaluate source water quality and treatment system efficacy, as positive H₂S test results indicate potential contamination from human and animal feces, and imply presence of pathogenic organisms in local drinking water supplies.

It was deemed appropriate to use H_2S tests in conjunction with a quantitative analytical technique to more precisely measure levels of fecal contamination. Correlation between H_2S tests and enumerative bacterial analyses were then compared to results obtained by Lukacs (2002), who performed similar assays in Nepal at approximately 21°C or 70°F. In contrast, field operating temperatures for the present work (at the same site as Lukacs' study) were generally at or below 10°C or 50°F.

3.7 Using Membrane Filtration Assays to Detect Fecal Pollution in Drinking Water

Concentrations of *Escherichia coli* (*E. coli*) and total fecal coliform bacteria in source waters and filtrate were enumerated via membrane filtration (see section 4.4.3). Results are presented as colony forming units per 100 mL sample volume (cfu/100 mL).

This work does not focus on arsenic contamination, i.e., the threat posed by naturally occurring arsenic to drinking water supplies. For more on this subject, see Halsey (2000), Hurd (2001), Poole (2002), Hwang (2002), Ngai (2002), Ngai and Walewijk (2003), and Tabbal (2003).

3.8 Evidence-Based Evaluation Criteria

While this work focused on microbiological data to assess BioSand filter performance, it is important to note that microbial data alone are insufficient to indicate intervention success. Simply showing that microbiological quality of filtrate is improved is insufficient evidence of system efficacy in reducing waterborne disease. Specifically, health data from intervention studies (e.g., on disease reduction) are critical to performance evaluations (see section 7.1.4).

4 Methodology

4.1 Field Site Description

4.1.1 Lumbini BioSand Filter Survey

The author visited 10 1-year old concrete BioSand filters (BSFs) in Nepal's southern Terai region during the month of January, 2003. Specifically, 6 villages in the Lumbini district – Sekhuwadand, Khambe, Sonbarshi, Ramawa-pur, Mujhana and BuddhaNagar – were visited. Figure 4.1 shows a typical example of a concrete BioSand filter installed in the village of Sekhuwadand. All concrete casings were constructed locally using a steel mold, the design drawings for which were provided by CAWST. Concrete molding and sand preparation (sifting and washing) was performed by Durga Ale (see Figure 4.6), a local technician trained in BioSand filter construction and operation, without any MIT Nepal Water Project supervision (see sections 4.2.1.1 and 6.3.4). Filter commissioning was observed by Lukacs (2002) and/or International Buddhist Society (IBS) staff.



Figure 4.1 Concrete BioSand filter (right) installed in village (Sekhuwadand).

IBS Dr. Narendra Mallik and several women motivators (Manorama Tripathi, Pobetra Panday and Susma Aryal) generally accompanied the author during village visits, acting as translators and communicating to the author health information and general observations.



Figure 4.2 International Buddhist Society (IBS) women motivators Pobetra Panday & Susma Aryal.

The women motivators (Figure 4.2) work with IBS to facilitate a health and wellness outreach program. Each woman motivator is assigned and is responsible for several villages that she visits; her responsibilities include checking in on villagers to discover who is sick and in need of care, encouraging individuals to take advantage of IBS health clinic services, and giving informal lessons on personal hygiene and community health and well-being.

The locations of the concrete BioSand filters are shown on village maps in Appendix A. All villages were accessed by jeep. Global Positioning System (GPS) coordinates of
sampling locations were obtained (see Appendices B and C) using a Garmin GPS III. Appendix C includes data on hand pump types (local, private or IBS), well depths and ages, and remarks on well locations.

4.1.1.1 Analytical Techniques

BioSand filter performance was evaluated using membrane filtration for enumeration of *E. coli* and total coliform bacteria, presence/absence tests for H_2S producing bacteria, turbidity and flow rate measurements. Source water quality was evaluated using enumeration of *E. coli* and total coliform bacteria, presence/absence tests for H_2S producing bacteria, and turbidity measurements. Microbial testing was performed in an empty room provided by the International Buddhist Society (IBS), a local village development program and Buddhist center (Figure 4.3). All laboratory equipment and supplies were brought from Cambridge, Massachusetts to Lumbini. Fortunately, there were no problems with airport security or customs agents; all supplies and equipment arrived safely in Nepal.



Figure 4.3 Author in IBS laboratory space.

4.1.2 Lumbini Well Survey

As per requests from IBS, Dr. Malik and several villagers, the author tested water from 21 wells (hand pumps) in 6 Lumbini district villages: Dhodahawa, Bhagawanpur, Lamtihawa, Mujahana, BuddhaNagar and Muhuwari (Figure 4.4). Two of these wells were private hand pumps (BUD_SK and BUD_CK), seven were installed by the government, 11 were financed and constructed by IBS, and one tapped an artesian aquifer at a depth of 350 feet. Wells were purged of approximately 20 L prior to sampling, however, spouts were not sterilized (e.g., flamed) prior to obtaining samples (see section 7.1.2).



Figure 4.4 Surveying a public well (hand pump). Author, center, with IBS women motivators Manorama Tripathi, left, and Pobetra Panday, right.

4.1.2.1 Analytical Techniques

Well water quality was evaluated using enumeration of *E. coli* and total coliform bacteria, presence/absence tests for H_2S producing bacteria, and turbidity measurements. Microbial testing was performed in a room at IBS.

4.2 Lumbini BioSand Filter Experiments

4.2.1 Concrete BioSand Filter Experiments

4.2.1.1 Experimental Set-Up

Experiments at IBS to elucidate biofilm maturation rates and bacterial removal efficiency were performed over an 8-day period. Filter ripening was defined in this context as an improvement in the ability of a filter to remove *E. coli*. *E. coli* were chosen as test particles because they were not expected to multiply in the filter columns (due to low temperatures and insufficient oxygen levels) and thus could be used as tracer particles (Weber-Shirk and Dick, 1997). Two concrete BioSand filters (Concrete Filter 1 [CF1] and Concrete Filter 2 [CF2], see Figure 4.5) were set up with sand obtained from a local (Butwal) rock-crushing operation.



Figure 4.5 Concrete BioSand filters used in field experiments. IBS students, foreground.

Set up was performed according to the recommendations of CAWST (2003) and Ritenour (1998). Filter commissioning was conducted with the help of Durga Ale (see Figure 4.6), a local technician trained in BioSand filter construction and operation by Samaritan's Purse. The two concrete filters were initially set up by Ale without the author's or other MIT Nepal Water Project supervision (the author was in the field surveying household filters at the time). Subsequent flow rate measurements (less than 10 L/hr) indicated improper construction techniques.¹¹ An inspection of the surface sand layer revealed non-uniform grain sizes (i.e., gravel mixed in with the fine sand), further verifying that improper sand preparation had taken place. At the author's insistence, the two filters were decommissioned and set up again by Durga Ale and the author, according to the CAWST methods outlined in section 4.2.1.3.

¹¹ The grain size of its sand is one critical variable (surface area is another) that determines the flow rate of a sand filter. If the flow rate is too low, then the sand contains too many fine particles and requires more washing, or sifting. If the flow rate is too high, you have either washed or sifted too many of the fine grains from the sand or the initial effective diameter of the sand was too large. If the initial flow rate of a 0.3 x 0.3 x 0.9 m concrete BioSand filter is in the range from 38 - 70 L/hr, then the preparations were sufficient (Ritenour, 1998). Flow rates of ripened filters are generally somewhat less than initial flows; flow rates of Lumbini concrete BioSand filters should ideally be between 20 and 40 L/hr.

4.2.1.2 Filter Sand Contamination

Sand used for filter set-up was obtained from a local rock-crushing operation (originally presumed to be rock quarry sand). Sand produced at a crushing operation is considered to be pure, clean, and relatively uniform in size and shape. It requires the least preparation and is the best possible sand source for BioSand filtration purposes (Ritenour, 1998). Filter media for the BioSand filter should be free of organic contamination, such as is often found in riverbank sand. Riverbed sand often contains the very contaminants filtration is attempting to remove (Ritenour, 1998). The organic material present in river sand provides food for microorganisms at depth within the filter. This may encourage microbial growth at depth, whereas in a properly functioning filter, the activity of the microorganisms is limited to the surface of the sand. Eventually, the organic material/food will be consumed. However, this process could take months and could result in more contaminated water leaving the filter than going in (Baker, 2002). In addition, riverbed sand grains are more rounded and smooth in their shape, which decreases their effectiveness in trapping contaminants (Ritenour, 1998).

Filter sand for IBS experiments was subsequently discovered to be a source of *E. coli* contamination (anecdotal evidence suggests that the crushing operators obtained their materials from river banks), as filtrate from both filters tested positive for *E. coli* bacteria when source water was free of contamination (see Appendices D and E, and Figure 5.1). Section 5.3.1.1 summarizes the results of these self-cleansing trends.



Figure 4.6 Durga Ale, Lumbini BioSand filter technician.

4.2.1.3 Filter Commissioning

In brief, sands and gravels were separated (sifted) into appropriately sized gravel, coarse and medium-fine sands (see Figure 4.7) and washed to remove finer silts and clays. IBS water that tested free of *E. coli* bacteria (obtained from well BUD2, see Appendix F) was used for sand preparation and filter washing. Sand and gravel layers were deposited according to instruction, being careful to always add sand to water.¹² Prior to commencing experiments, filters were washed (using approximately 30 L of water in all) until filtrate came out clear. Throughout experimentation the existence of a 5 cm (2 in) layer of standing water was verified (see section 2.1). For a more detailed description of filter casing construction, sand preparation, set-up, operation and maintenance, the reader is referred to Ritenour (1998) and CAWST training materials (CAWST, 2003).

¹²Adding sand to water allows air in the sand to escape. Water only flows where water is, and where there is air there cannot be water. The surface of any sand grain surrounded by air is not available for adsorption; it will never meet with water or anything in the water (Ritenour, 1998).



Figure 4.7 Filter media: clockwise from left, gravel, coarse sand and fine sand.

4.2.1.4 Experimental Procedures

Filters were challenged with an average of 21 L of water per day (see Table 5.7). Raw water was temporarily stored in plastic buckets prior to use, and time lag between water collection and filtering never exceeded 20 minutes. Raw water samples were obtained by sampling directly from collection buckets. Filtrate samples were obtained after approximately 10 L of water drained through the filter column.

Initially, both filters were challenged with water that had previously tested positive for *E. coli* contamination (water from holding tanks on IBS roof, >100 cfu/100 mL). Concrete filter 2 (CF2) was fed a 1:1 mixture of CF1 raw water and *E. coli* free IBS water (i.e., CF1's *E. coli* concentration diluted by half). Subsequent testing proved the contamination in the holding tank water to be transient (possibly from bird feces) – no *E. coli* were discovered in this source water after the first day of experimentation. However, filter sand was discovered to be a source of fecal contamination, as filtrate tested positive for *E. coli* bacteria (see Appendices D and E, and section 5.3.1.1). For consistency, the two concrete filters were challenged with this same source water make-up (i.e., pure

holding tank water for CF1 and 1:1 dilution for CF2) until 0 cfu/100 mL *E. coli* bacteria were detected in filtered water.

On the fifth day of experimentation the concrete filters were challenged with *E. coli* rich source water from a stagnant pond on IBS property. Source water concentrations for CF1 varied between 75 cfu/100 mL and at least 500 cfu/100 mL. CF2 was fed a 1:1 mixture of CF1 raw water and *E. coli* free IBS water (i.e., CF1's *E. coli* concentration diluted by half, see Appendices D and E). However, *E. coli* concentrations in raw water for both filters were variable, and influent *E. coli* concentrations for CF2 varied from at least 100 cfu/100 mL to 500 cfu/100 mL. The author was never able to verify that actual concentrations of *E. coli* in CF2 raw water were half those of CF1. Nonetheless, source water used during the latter four days of testing did have elevated levels of *E. coli* bacteria, in contrast to the first four.

4.2.2 BioSand Pitcher Filter Experiments

4.2.2.1 Experimental Set-Up

The author conceptualized the BioSand pitcher filter (see Figure 2.4) as an alternative household water purification technology. Pitcher filters could also serve as bench-scale models of larger BioSand filters, and would be far easier to use for experimental purposes than the full-size units. Two prototypes (Green Pitcher Filter [GF] and Blue Pitcher Filter [BF]) were constructed using materials bargained for at the local market. Field experiments at IBS were performed over a 4-day period. The purpose of these investigations was to conduct a preliminary evaluation of pitcher filter viability by cross-checking their performance with the concurrent performance of the concrete BioSand filters. The dependence of microbial removal efficiency (if any) on type of filter media was also explored.

4.2.2.2 Dependence of Microbial Removal Efficiency on Filter Media

Pitcher filters were used to perform preliminary bench-scale studies of microbial removal dependence on sand type. While Ritenour (1998) and Baker (2002) recommend using the cleanest available crushed rock for filter media, studies by University of Berkelely, California students (Coan and Stoller, 2002) suggest that bacterial removal rates are not significantly affected by sand type or method of sand preparation.

To this end, two identical pitcher filters were constructed and studied, differing only by fine sand type (see section 4.2.2.5). A green pitcher filter was constructed using sand obtained from a local riverbank. A blue pitcher filter was constructed with riverbank sand which had been dried in the sun for 2 days. Sand collection was performed with the help of Durga Ale (see Figure 4.6). Specifically, sand was collected from low on the riverbank, directly adjacent to the water's edge.

4.2.2.3 Pitcher Filter Viability

Secondly, pitcher filters were studied to evaluate their viability as a drinking water purification technology. Each filter was challenged with 2 L of *E. coli* rich source water per day, and subsequent performance evaluated. Laboratory experiments conducted at MIT in March and April of 2003 (see section 4.3) explored pitcher filter capabilities for treating greater volumes of water.

4.2.2.4 Analytical Techniques

Pitcher filter performance was evaluated using enumeration of *E. coli* bacteria removal, presence/absence tests for H₂S producing bacteria, turbidity and flow rate measurements.

4.2.2.5 Pitcher Filter Commissioning

The main principles of BioSand filter set-up and operation were adhered to in pitcher filter commissioning. In brief, sands were separated into appropriately sized coarse and fine sands. A gravel layer was omitted due to spatial considerations. IBS water that tested free of *E. coli* bacteria (obtained from well BUD2, see Appendix F) was used for sand preparation and filter washing. The coarse sand (1 inch layer) used for both pitcher filters was sterilized by immersion in boiling water for 10 minutes. Fine sand (~ 4 inch layer) was deposited next, taking care to always add sand to water. The green filter (GF) fine sand was obtained from a local riverbank. Fine sand for the blue filter (BF) was riverbank sand which had been sun-baked for 2 days on the IBS roof. Appendix G gives local pyronometer readings of UV-A radiation for both days (January 14 and January 15). Levels of radiation necessary to destroy aqueous pathogenic microorganisms were reached and exceeded on both days (SANDEC, 2002; Sobsey, 2002 and Wegeling et al., 1994).¹³ Levels needed to destroy microorganisms in sand were not known at the time these experiments were conducted. Subsequent research in this area is needed to determine levels of solar radiation necessary to destroy pathogenic microorbes in sand.

Prior to commencing experiments, filters were washed (using approximately 1 L of water in all) until filtrate came out clear. Instructions for pitcher filter construction may be found in Appendix H.

¹³At a water temperature of about 30°C, a threshold solar radiation intensity of at least 500 W/m² (all spectral light) is required for about 5 hours for SODIS [solar disinfection] to be efficient. This dose contains energy of 555 Wh/m² in the range of UV-A and violet light, 350nm-450nm, corresponding to about 5 hours of midlatitude (European) midday summer sunshine. At a water temperature of 30°C, a fluence of 555 W*h/m² (350-450 nm, dose of solar radiation corresponding to approximately 6 hours of mid-latitude midday summer sunshine) is required to achieve a 3-log reduction of fecal coliforms. Under these conditions, only the effect of UV-A radiation is present. However, the die off rate of fecal coliforms exposed to sunlight increases significantly, when two stress factors, UV-A radiation and increased water temperature are present. At a water temperature of 50°C, a synergetic effect of UV-A radiation and temperature occurs: a 3-log reduction of fecal coliforms only requires a fluence of 140 W*h/m². This is equivalent to an exposure time of only one hour (SANDEC, 2002).

4.2.2.6 Experimental Procedures

Filters were challenged with an average of 2 L of water per day (see Table 5.7). Raw water was temporarily stored in plastic buckets prior to use, and time lag between water collection and filtering never exceeded 20 minutes.

Source water for pitcher filters was obtained from a stagnant, highly turbid pond on IBS property. Raw water *E. coli* concentrations varied from 400 cfu/100 mL to at least 1000 cfu/100 mL.

4.3 MIT BioSand Filter Experiments

Laboratory experiments were performed to compare the performance of two BioSand pitcher filters, a green pitcher filter (MIT-GF) and a white pitcher filter (MIT-WF), with a Davnor plastic BioSand filter (MIT-DF, Davnor, 2002, see Figure 2.2).

Experiments were performed to determine break-through curves of start-up times, i.e., biofilm maturation times. Overall filter performance was evaluated to determine feasibility of pitcher filters as a new water filtration technology, and assess pitcher filter viability as a bench-scale testing platform for the full-size units.

4.3.1 E. coli Culturing Methodology

Filter performance was evaluated by measuring *E. coli* and turbidity removal, and flow rates. K12 *E. coli* from a pure culture were added to all influents to serve as test particles for evaluating filter performance (see section 4.2.1.1).

E. coli were grown at 37°C overnight on agarose gel. Cells were resuspended in dilute nutrient broth¹⁴ (25 g nutrient broth/L) and cultured overnight on a BIGBill Thermolyne shaker. Liquid cultures were transferred to 2 mL plastic cuvettes and enumerated at 600

¹⁴ Luria-Bertani broth

nm using a Beckman DU640 spectrophotometer. Cultures were diluted to obtain a concentration of approximately 100 *E. coli*/100 mL (corresponding to an absorbance of ~1.1 diluted 100 fold), however, actual influent concentrations measured were variable (see section 5.4.1 and Appendices L - N).

4.3.2 Experimental Set-Up and Filter Commissioning

The author attempted to set up both pitcher filters so that they differed only by type of filter casing (i.e., identical holding capacity, identical supernatant depth, etc.). The green pitcher filter casing was obtained in Lumbini (see above); the white pitcher casing was obtained at an Economy Hardware store in Cambridge, Massachusetts.

Filter media for both the Davnor and pitcher filters was sand obtained from a Home Depot store in Somerville, Massachusetts.¹⁵ Gravels, course and fine sands were separated, washed and installed according to Ritenour (1998). Pitcher filter sand layers included 1 inch of gravel, 1 inch of coarse sand, and 4 inches of fine sand (approximate values, see Figure 2.4). Prior to commencing experiments, filters were washed (using approximately 30 L of water for the Davnor filter and approximately 10 L of water for each pitcher filter) until filtrate came out clear. Distilled water was used for sand preparation and filter washing. Throughout experimentation the existence of a 5 cm (2 inch) layer of standing water was verified. Holding capacity for experimental filters is as follows: 6 L of water for Davnor filter, and 0.5 L and 0.3 L for green and white pitcher filters, respectively.

4.3.3 Analytical Techniques

Filter performance was evaluated using enumeration of *E. coli* bacteria removal, turbidity and flow rate measurements.

¹⁵ Home Depot personnel verified that their sand source was a rock-crushing operation.

4.3.4 Experimental Procedures

Twenty-nine days of filter experiments were conducted (March 7th – April 4th). Source water was obtained daily from the Charles River, in Cambridge, Massachusetts. River water was spiked with fresh *E. coli* cultures grown during the previous night. River water temperatures were around 6°C (approximately 43°F) and may have contributed to *E. coli* die-off during the first 12 days of experimentation (see below). Experiments were paused on Saturday, March 29th and resumed on Monday, March 31st. Flow rate and turbidity data were obtained every day except March 29th and 30th. *E. coli* removal was measured March 7th – 18th (days 1 – 12), March 20th – 26th (days 14 – 20), April 1st (day 26) and April 3rd – 4th (day 28 – 29).

The Davnor filter was challenged with an average of 8.6 L of water per day; the green pitcher filter averaged 4.6 L/day and the white pitcher filter averaged 4.5 L/day (see Table 5.8). For the first 12 days of experimentation (March $7^{th} - 18^{th}$), filters were fed *E. coli* spiked river water that had been collected no less than 1 hour before (i.e., time lag between water collection and filtering never exceeded 1 hour). Difficulties ensued maintaining a constant level of live *E. coli* in influent water, likely due to temperature shock upon contacting freshly thawed river water. Room temperature river water (22°C or approximately 72°F), allowed to stand in covered buckets, overnight) was used for the remainder of the testing period.

On April 3rd and 4th (the 28th and final day of experimentation, respectively), filters were challenged with a 1:1 mixture of room temperature Charles River water and wastewater from the Deer Island Wastewater Treatment Plant in Boston, Massachusetts.

4.4 Analytical Techniques

4.4.1 Laboratory Conditions in Nepal

Field experiments conducted in Nepal in January, 2003, were based out of a room provided by IBS, equipped with supplies brought mostly from the United States (see Figure 4.3). IBS kindly provided a propane stove and pressure cooker for boiling water and sterilizing glassware. The laboratory was generally fairly congested as children and adults often stayed to observe experimental procedures. Overcrowding made it challenging to maintain sterile surfaces and microbial testing equipment. Even so, *E. coli* free quality assurance blanks were consistently obtained (see Appendix O).

Visibility in the laboratory was poor, so head lamps were often worn to augment the light provided by two light bulbs and several small windows. Head lamps were always worn in the evenings, as evening brown-outs were common. Dilution and rinse water were obtained from a tap outside (approximately 20 feet or 6 meters distant) and sterilized by boiling for 10 minutes on a portable propane stove. All glassware was sterilized by immersion in boiling water for 10 minutes.

4.4.2 Laboratory Conditions at MIT

Laboratory experiments were conducted at MIT in March and April, 2003 in a Civil and Environmental Engineering Department laboratory space. Though the author was frequently the only person working at the back bench of the laboratory and hence could maintain some degree of control over the sterility of the workplace, the laboratory was utilized for department classes at other times. Though this may have compromised the sterility of surfaces and testing equipment, *E. coli* free quality assurance blanks were consistently obtained (see Appendix P).

Laboratory lighting was excellent. As electricity was continuous – 24 hours a day, 7 days a week – grid electricity was used to power the Millipore incubator (see section 4.4.3). Dilution and rinse water were obtained by boiling distilled water for 10 minutes on a portable electric stove. All glassware was sterilized by immersion in boiling water for 10 minutes.

4.4.3 Quantification of total coliform and E. coli bacteria

Both *E. coli* and total coliform bacteria were enumerated following Standard Methods membrane filtration procedures. In Nepal, samples were collected in 100-mL presterilized whirl-pack bags and transported in an insulated cooler for same-day analysis upon return to IBS. Samples were filtered through 0.45 um pore size paper (see Figure 4.8) and placed in a petri dish containing m-ColiBlue24TM nutrient broth, then incubated for 24 hours at 35 ± 0.5 °C in a Millipore portable field incubator.



Figure 4.8 Millipore portable membrane filtration assembly (left) with vacuum hand pump, lighter, pliers, forceps, petri dish, 0.45 um filter paper, m-Coliblue24TM nutrient broth ampules and sterilized water.

Samples (diluted or undiluted) less than 10 mL were added to approximately 50 mL of sterile dilution water prior to filtering through the membrane apparatus. This increase in

water volume aids in uniform dispersion of the bacterial suspension over the entire filtering surface (Standard Methods, 1998).

To ensure continuous electricity supply in Lumbini, incubator power was supplied by Millipore Ni-Cd batteries, as evening brown-outs (during periods of peak demand) were common. Following incubation, petri dishes were removed from the incubator and colony counts recorded. Red and blue colonies indicated total coliforms and blue colonies specified *E. coli*. Sampling results are presented as colony forming units per 100 milliliters of water (cfu/100 mL).

All laboratory surfaces were sterilized by wiping down with ethanol. Sterile rinse water was obtained by bringing IBS water to a rapid boil and continuing to heat for 10 minutes. To ensure accuracy of results, quality assurance blanks were obtained during each day of experimentation (see Appendix O). Duplicate sampling was also performed (see Table 5.6). Where duplicate samples were obtained, tabulated results are presented as mean values (see Appendices D and E, and J and K). Where microbial measurement results are presented as bounded values, the lower limits were used to calculate removal efficiencies. For a more detailed outline of experimental procedures, see Lukacs (2002).

4.4.4 Presence/absence tests for H₂S producing bacteria

Presence/absence tests for H₂S producing bacteria were conducted using HACH PathoScreenTM Medium MPN Pillows for 20 mL sample bottles. These tests are suitable for detecting *Salmonella*, *Klebsiella*, *Proteus*, *Citrobacter*, *Clostridium*, *Edwardsiella* and other H₂S producing organisms proven to be associated with fecal contamination and the presence of coliforms (see section 3.3). Samples were collected in 20-mL sample bottles which had been sterilized by immersion in boiling water for 10 minutes. Following sample collection, the contents of one PathoScreen MediumTM MPN Pillow was added to each bottle (see Figure 4.9). Each bottle was capped and shaken, then incubated at 35 ± 0.5 °C for 24 hours. A color change from yellow to black or the formation of a black precipitate was considered a positive for H₂S producing bacteria.



Figure 4.9 H₂S test field equipment: hand sanitizer (left) with 20 mL sterilized glass vials, nail clippers, lighter, marking tape and powdered media capsules.

4.4.5 Turbidity measurements

Turbidity measurements were conducted using a HACH Pocket TurbidimeterTM Analysis System (see Figure 4.10).



Figure 4.10 Turbidimeter field kit. Counter-clockwise from left: calibration standards, pocket turbidimeter, instruction manual and sample vials (HACH, 2003).

The HACH turbidimeter measures turbidity in the range of 0.1 to 400 NTU. Sampling containers were rinsed with sample water three times prior to sample collection. Following sampling, the outside surfaces of the collection vessel were wiped free of debris, moisture, etc. Recalibration of the turbidimeter was performed weekly.

4.4.6 Flow rate measurements

Flow rate measurements were conducted according to the recommendations of Ritenour (1998). Flow rate measurements for both full-size and pitcher filters began when the water level was midway between the diffuser plate and the top of the filter. Rates were obtained by using a stopwatch to measure the time it took to fill a 100 mL polypropylene graduated cylinder. Where flow rates were low, a 25 mL or 50 mL polypropylene graduated cylinder was used instead.

5 **Results and Project Implementation**

5.1 Lumbini BioSand Filter Survey

5.1.1 Microbial Removal

5.1.1.1 <u>E. coli Bacteria</u>

Microbial measurements of source water and filtrate were obtained from 9 out of 10 BioSand filters visited. Appendix B summarizes field observations of filters during village visits. All but one filter (BSF 5 – Khambe) were in good condition, i.e., the concrete casing and diffuser plate were intact, and the cover and diffuser plate were in place. The diffuser plate of BSF 5 was chipped. Two of the filters (BSF 8 – Ramawapur and BSF 9 – Mujhana) had spouts stopped with straw and thus a standing water layer greater than 5 cm (as per design).¹⁶ However, filtered water from these BioSand filters was *E. coli* free (i.e., 0 cfu/100 mL *E. coli* bacteria), as source water was of high quality to begin with (*E. coli* concentrations of 0 and 1 cfu/100 mL, respectively, see Table 5.1 and Appendix I). BSF 1 and BSF 4 (Sekhuwadand) were also treating high quality source water (raw water *E. coli* concentrations of 2.5 and 0 cfu/100 mL) with high removal efficiencies.

Filter	Rav	v Water	Water Filtered Water		Filtered Water		%	Log
	E. Coli	Total Coliform	E. Coli	Total Coliform	Removal	Reduction		
	(cfu/100 mL)	(cfu/100 mL)	(cfu/100 mL)	(cfu/100 mL)	E. Coli	Value		
BSF1	2.5	10	0.5	1.5	80	0.7		
BSF2	0	110	>10	>90	(-1000)	-1		
BSF3	0	1.5	>400	>800	(-40000)	-2.6		
BSF4	0	20	0	15	N/A	N/A		
BSF5	>1000	>2000	10	>1010	99	2		
BSF6	>110	>110	0	>120	99	2		
BSF7	0.5	29	>1000	>1033	(-199900)	-3.3		
BSF8	0	5.3	0	>1000	N/A	N/A		
BSF9	1	101	0	1	(100)	N/A		
BSF10	N/A	N/A	N/A	N/A	N/A	N/A		

 Table 5.1 Microbial data from Lumbini district BioSand filters

¹⁶ Filter owners indicated that they plugged spouts to control flows.

While 2 filters, BSF 5 and BSF 6 (Khambe), were successfully removing 99% (Log Reduction Value [LRV] of 2)¹⁷ of *E. coli* from raw water with elevated bacterial concentrations, 3 filters (BSF 2 and BSF 3 – Sekhuwadand and BSF 7 – Sonbarshi) were contaminating clean source water. For example, BSF 3 was adding more than 400 cfu *E. coli*/100 mL to source water with 0 cfu/100 mL. BSF 7 is also a likely health threat, adding more than 1000 cfu/100 mL to relatively clean raw water (0.5 cfu/100 mL) of low turbidity (2.8 NTU, see Table 5.2 and Appendix I).¹⁸

Filter	Flow Rate (L/hr)	Turbidity (± 0.1 NTU)				
		Raw Water	Filtered Water	% Removal		
BSF1	5.6	3.9	1.7	57.7		
BSF2	1.0	360.0	0.8	99.8		
BSF3	14.5	15.0	1.7	88.7		
BSF4	24.1	176.0	2.3	98.7		
BSF5	2.8	179.0	2.3	98.7		
BSF6	2.4	1.2	1.8	(-50.0)		
BSF7	2.4	2.8	1.3	53.6		
BSF8	37.5	3.4	2.1	38.2		
BSF9	34.9	5.0	2.0	60.0		
BSF10	0.0	N/A	N/A	N/A		

Table 5.2 Turbidity and flow rate data from Lumbini district BioSand filters.

Even so, one or more of these filters may still be helping to improve health and reduce sickness overall as measured via turbidity removal. For example, BSF2 was removing 99.8% of turbidity from highly turbid raw water (360.0 NTU). The owner of this filter specifically commented on less sickness and improved health following filter introduction. Though introducing *E. coli* into drinking water, BSF 2 is significantly reducing suspended particulate concentrations and may be filtering out disease-causing organisms.

¹⁷ Log Reduction Value (LRV) = $\log_{10}(\text{raw water } E. \ coli \ \text{concentration/filtered water } E. \ coli \ \text{concentration})$. 1 LRV = 90% reduction, 2 LRV = 99% reduction, 3 LRV = 99.9% reduction, etc.

¹⁸ The author did not have time to revisit any of the Lumbini household BioSand filters and resample raw water and filtrate. Had time permitted, the author would have attempted to sterilize (e.g., flame) the BioSand filter and tubewell spouts prior to resampling. In the case of BSF7, a negative resampling result (i.e., 0 cfu *E. coli*/100 mL) would have pointed to spout contamination (e.g., from handling) as opposed to BioSand Filter contamination (see section 7.1.2).

One filter, BSF 10 (Buddhanagar, IBS kitchen), was being used as a counter-top, as its height and flat lid made it a convenient work space. However, all IBS raw water sources tested free of microbial contamination (see Appendix F), so the filter did not appear to be needed.

5.1.1.2 Total Coliform Bacteria

Concentrations of total coliform bacteria in raw waters varied considerably from 1.5 cfu/100 mL (BSF 3 – Sekhuwadand) to more than 2,000 cfu/100 mL (BSF 5 – Khambe). In general, water sources with elevated concentrations of *E. coli* had elevated concentrations of total coliform as well. The reverse was also true, i.e., high concentrations of coliform bacteria (>100 cful/100 mL) were observed in source waters free of *E. coli* contamination (0 cfu/100 mL).

Filters which significantly reduced influent *E. coli* concentrations did not perform similarly with respect to total coliform concentrations. While BSF 6 reduced the concentration of *E. coli* in raw water from more than 110 cfu/100 mL to 0 cfu/100 mL, total coliform concentrations, excluding *E. coli*, increased from 0 cfu/100 mL to greater than 120 cfu/100 mL. BSF 5, which reduced *E. coli* concentrations from more than 1,000 cfu/100 mL to 10 cfu/100 mL, maintained total coliform concentrations (excluding *E. coli*) well above 1,000 cfu/100 mL, the influent value. These results suggest growth of total coliform bacteria within the filter or contamination of the filter spout (see section 7.1.2).

Elevated levels of total coliform bacteria do not necessarily mean that BSF 5 and 6 are failing to purify contaminated water, however. Specifically, *total* coliforms do not always represent *fecal* coliforms in water samples. Besides fecal coliforms, which are generally found in human and animal (warm-blooded) intestinal tracts, many other harmless coliforms proliferate in the environment (e.g., in soils, plants and animals). These bacteria would all be subject to detection by the microbial test method employed, but not necessarily indicate presence of pathogenic microorganisms.

5.1.2 Flow Rates

Design flow rates for CAWST concrete BioSand filters are intended to fall in the range of 20 - 40 L/hr. Lumbini filter flow rates varied from 1.0 to 37.5 L/hr (see Table 5.2). Of the 9 functioning filters, 5 (BSF 1, 2, 5, 6, and 7) had flow rates less than 6 L/hr, and 4 of those 5 had flow rates less than 3 L/hr. These flows are clearly below the design specified flow rates of 30 ± 10 L/hr. Four filters (BSF 3, 4, 8, and 9) had flow rates between 14.5 and 37.5 L/hr, well within the design range.

Low flow rates did not always correspond to high bacterial removal efficiency.¹⁹ Though BSFs 5 and 6 did have high bacterial removal efficiency with low flow rates (2.8 and 2.4 L/hr, respectively), BSF 7, which added more than 1000 cfu/100 mL to raw water with 0.5 cfu/100 mL, had a flow rate of 2.4 L/hr.

5.1.3 Water Quality Aesthetics

All functioning filters had high turbidity removal, reducing influent turbidity to less than 3 NTU (see Table 5.2). Filters also significantly cooled warmer source water. Although it was not specifically mentioned by any filter user in this survey, BioSand filters have also been historically reported to reduce bad taste and odor (Davnor, 2003) of raw water.

5.1.4 General Observations

Most filters were reported to serve between 10 and 20 people; filtered water was used either for drinking only, or drinking and cooking. No villagers reported using filtered water for bathing or washing laundry. Cleaning frequency varied from 2 to 4 times per month, with 1 person generally reported as responsible for maintenance.

¹⁹ Low flow rates may be expected to correspond to high bacterial removal efficiency because contact time between raw water and biofilm is enhanced.

In general, filters were well liked by users, and many villagers expressed an interest in aquiring one. Although no social acceptability or health-based survey was conducted (see section 7.1.4), filter owners reported improved health and reduced sickness incidence following filter introduction. A more formal assessment would be necessary to determine whether these communications were reported truthfully or to please MIT Nepal Water Project staff.

Appendix Q presents a summary of morbidity statistics (for Leucorrhea, Amoebiasis, Diarhea and Gastritis) in Lumbini district villiges obtained from a display in the IBS health clinic. A summary of illness symptoms is also presented, obtained from notes from a conversation between Susan Murcott and Rajess Yadav, the IBS Community Health Assistant. These data show that there is a health monitoring system in place at the IBS clinic, specifically, a program for monitoring incidence of the waterborne diseases, Leucorrhea, Amoebiasis and Diarhea.

5.1.5 Comparison with Previous Work

Lukacs (2002) performed preliminary fecal coliform testing of BioSand filters upon installation in villages. Raw water (private well water) was found to contain little contamination (only one well had fecal coliform counts >2 cfu/100 mL). Overall, results were inconclusive as samples were obtained prior to filter ripening (see section 2.3). Flow rate measurements of newly installed filters were also obtained. Flow rates for new filters averaged 29 L/hr, with a marked reduction in flow rate following the first 5-7 days of operation (to 18 L/hr). In general, these initial flow rates are much higher than flow rates obtained during the 2003 filter survey (see Table 5.3). However, it is to be expected that flow rates will decrease as filters ripen and particle deposition occurs. The low flow rates obtained during the 2003 survey may indicate one or both of the following: filters are functioning normally and simply need to be cleaned, or filter media was prepared incorrectly and filters should be re-commissioned entirely (see section 6.3.4).

Filter	Village	Owner	Flow Rate (L/hr)	Flow Rate (L/hr)
			Lukacs (2002)	Pincus (2003)
BSF1	Sekhuwadand		20	5.6
BSF2	Sekhuwadand		15	1.0
BSF3	Sekhuwadand	Ram Chandra		14.5
BSF4	Sekhuwadand	Kushylia		24.1
BSF5	Khambe	Pusspalata	20	2.8
BSF6	Khambe		20	2.4
BSF7	Sonbarshi	School	34	2.4
BSF8	Ramawa-pur	Keshav Pari Yadar	31	37.5
BSF9	Mujhana	School		34.9
BSF10	BuddhaNagar	IBS		0.0

Table 5.3 Comparison of flow rate data for Lumbini district BioSand filters from 2002 and 2003 surveys.

5.2 Lumbini Well Survey

5.2.1 Microbial Concentrations

5.2.1.1 E. coli Bacteria

Twenty-one wells (hand pumps) were visited over a 10-day period. Most wells tested free of *E. coli* bacteria; the rest had relatively low concentrations (see Appendix F). Of the 5 wells with non-zero *E. coli* concentrations, 4 (B15 – Bhagawanpur, L1 – Lamtihawa, BUD6 – BuddhaNagar and MUH8 – Muhuwari) had concentrations between 1 and 10 cfu/100 mL and 1 (B16 – Bhagawanpur) had a concentration of 28 cfu/100 mL. Well L2 (Lamtihawa) was inoperative.

5.2.1.2 Total Coliform Bacteria

Total coliform bacteria concentrations varied from 0 cfu/100 mL (BUD 3 and BUD_SK – BuddhaNagar) to more than 1,004 cfu/100 mL (B15 – Bhagawanpur, see Appendix F). In general, coliform concentrations were low, though samples from most wells showed some evidence of total coliform presence. Of the 21 wells visited, 2 tested free of coliform bacteria (0 cfu/100 mL), 12 had total coliform concentrations less than 100 cfu/100 mL and 5 had concentrations between 100 and 500 cfu/100 mL. Wells B15 and B16 (Bhagawanpur) had higher concentrations of >1,004 and >628 cfu/100 mL, respectively. As mentioned previously, these coliforms may be naturally occuring bacteria, and not necessarily indicative of fecal contamination.

5.2.2 Turbidity

Turbidity readings were obtained from all wells but the artesian well in Dhodahawa (DW9, see Appendix F). Well water was generally low in suspended particles – all turbidity levels were less than 3.0 NTU.

5.2.3 Comparison with Previous Work

Lukacs (2002) and Sullivan (2002) conducted a more extensive well survey in Lumbini a year prior to this work. Eighty-six tubewells were tested for H_2S bacteria, of which 1/3 tested positive. Sixty-seven wells were sampled for fecal coliform bacteria. Of the 39 deep public wells tested for fecal coliform contamination, 4 were new wells and highly contaminated. Of the remaining 35 deep wells, 7 or 20% contained relatively low fecal coliform concentrations (<15 cfu/100ml). The 18 private wells surveyed were nearly twice as likely (39%) to be contaminated, according to the fecal coliform results. In addition, new wells were found to have the largest concentrations of fecal coliform bacteria, with the highest concentrations detected exceeding 10,000 cfu/100 mL for a well sampled the day following installation. The presence of fecal coliform in new wells is not unexpected due to the standard practice of using cow dung slurry during well drilling (Lukacs, 2002).

Table 5.4 presents a portion of the author's *E. coli* sampling results alongside fecal coliform test data obtained by Lukacs (2002) for several Lumbini district public wells. Noticeable is the significant variability in contamination from year to year.

Table 5.4 Comparison of microbial test data for Lumbini district public wells from 2002 and 2003

Well	E. coli	Total Coliform	Fecal Coliform
	(cfu/100 mL)	(cfu/100 mL)	(cfu/100 mL)
	Pincus (2003)	Pincus (2003)	Lukacs (2002)
DW9	0	15	0
B15	4	>1004	0
B16	28	>628	0
Ll	5	130	0
M3	0	>20	0
MUH4	0	85	187
MUH5	0	4	>300
MUH6	0	1	0
MUH7	0	2	0
MUH8	1	33	0

well surveys.

For example, Muhuwari wells 4 and 5 (MUH4 and MUH5, respectively) tested free of contamination (0 *E. coli*/100 mL) during the 2003 well survey, as compared to the elevated levels of fecal coliform bacteria detected during the 2002 well survey (187 cfu/100 mL and >300 cfu/100 mL, respectively). However, these wells were one week old when visited by Lukacs in 2002, and may have showed initially high levels of contimation due to the common practice of using cow dung slurry during well boring (see Lukacs, 2002).

Conversely, wells B15 and B16 (Bhagawanpur) and L1 (Lamtihawa) tested positive for *E. coli* in 2003, where none had been detected previously. It is recommended that additional and more frequent sampling be conducted to augment these results and characterize (if any) seasonal changes in well contamination (see section 7.1.1), however, the author recognizes that this is difficult to accomplish in the context of available skills and equipment, or other more pressing needs.

5.3 Lumbini BioSand Filter Experiments

5.3.1 Concrete BioSand Filter Experiments

Eight days of IBS filter experiments were conducted. The first 4 days of experimentation (January 11th – January 14th) were devoted to an investigation of microbial self-cleansing trends, while the last 4 (January 15th – January 18th) were conducted with the intent of more precisely quantifying bacterial removal efficiency.

5.3.1.1 Filter Sand Contamination and Self-Cleansing Trends

Subsequent to setting up the filters on January 11^{th} , membrane filtration analyses for *E*. *Coli* were performed and filter sand was determined to be a source of *E*. *coli* contamination. That is, filtrate tested positive for *E*. *coli* bacteria when source water was free of contamination. As shown in Figure 5.1, both filters cleansed themselves quickly.



Figure 5.1 Self-cleansing trend of E. coli concentrations in filtered water.

CF1 outflow water decreased from at least 100 cfu/100 mL to 0 cfu/100 mL in 2 days, while CF2 outflow decreased from at least 500 cfu/100 mL to 0 cfu/100 mL within 1 day. Though sample size is small (n=2), it is reasonable to suppose that the cleansing period would vary in proportion to the extent of residual contamination of filter sand. It is possible that the sand was clean upon leaving the crushing facility, and that superficial contamination occurred during subsequent human handling.

5.3.1.2 Quantifying Bacterial Removal Efficiency of Concrete BioSand Filters

By January 15th (the fifth day of experimentation), filtrate from both concrete BioSand filters tested free of *E. coli* (0 cfu *E. coli*/100 mL). On this day and all subsequent days of experimentation, the filters were challenged with *E. coli* rich, highly turbid raw water obtained from a small, stagnant pond on IBS property. The bacterial removal efficiency of CF1 increased fairly regularly from 25% on January 15th to 95% on January 18th (Log Reduction Values [LRVs] in influent *E. coli* concentrations of 0.1 to 1.3,²⁰ respectively; see Figures 5.2 and 5.3, below, and Appendix D).



Figure 5.2 *E. coli* removal efficiency data obtained during January 2003 Lumbini BioSand filter experiments.

 $^{^{20}}$ Log₁₀ Reduction Value (LRV) = log₁₀(raw water E. coli concentration/filtered water E. coli concentration).

This increase in microbial removal may be attributed to growth of a metabolically active community within the biofilm over time (see section 2.3).

Further testing would have been necessary to verify that 95% microbial removal was not a transient response to influent *E. coli* concentrations, but truly indicative of a matured biofilm. For example, removal efficiency of CF1 dipped from 80% on January 16th to 69% on January 17th, but increased above the January 16th value to 95% on January 18th – an overall increase in 3 days. Given that CF1 was already 4 days mature when challenged with this source water, total ripening time for this filter may thus be approximated at 8 to 10 days (see Table 5.5).

These results support the use of concrete BioSand filters in colder climates, as daytime field operating temperatures for this work were generally at or below 10°C or 50°F (temperatures as low as 0°C or 32°F may have been reached some nights). Cold weather may be reasonably expected to diminish bacterial removal capacity of the supernatant by slowing bacterivore growth (see section 2.2 - 2.3), however *E. coli* removals observed are comparable to results from other studies on concrete BioSand filters (see section 2.4).

The possibility that the CF1 data points are erroneous (e.g., the result of random data variability) should not be discounted. This is unlikely, however, as the data exhibit a reasonable trend which correlates well with findings from previous studies (see University of Calgary, 1994; Buzunis, 1995 and Lee, 2001). Good correlation between duplicate samples (see section 5.3.1.3) and *E. coli* free quality assurance blanks (see Appendix O) also lend credibility to these results.





BioSand Filter	Estimated Ripening Period	Source Water Quality	
	(days)		
Lumbini Experiment	s		
Concrete Filter 1	8-10	poor/organic rich	
Concrete Filter 2	8-10 (uncertain)	poor/organic rich	
Green Pitcher Filter			
Blue Pitcher Filter			
MIT Experiments			
Davnor Filter	30-40	high/organic poor	
Green Pitcher Filter	30-40	high/organic poor	
Blue Pitcher Filter	30-40	high/organic poor	

Table 5.5 Estimates of BioSand filter ripening periods.

E. coli removal trends for CF2 are more difficult to explain. *E. coli* removal efficiency decreased from 99% on January 15th to 68% on January 18th (LRVs of 2.0 to 0.5, respectively, see Appendix E). It is possible that sampling results from January 15th and 16th (at 99% and 99.8% microbial removal, respectively) are erroneous, and removal efficiencies for those days are in reality less than 99%. Similarly, data from January 17th and 18th, which show decreasing removal efficiencies, may also misrepresent system conditions; data may simply be naturally skewed. It is likely that an increase in microbial removal efficiency should have occurred if experiments had continued January 18th, i.e., past 4 days.

Sample accuracy with respect to volume tested should also be considered. As shown in Figure 5.2, the first 2 CF2 data points, each at 100%, may not necessarily represent microbial removal efficiencies on those days. The author found that sample volumes of 100 mL (or 1 mL, for that matter) were not always representative of 20 L test volumes. For example, on January 16th (day 6 of experimentation), 1 mL influent²¹ (raw water) and effluent (filtrate) samples for CF1 tested free of *E. coli*, while 100 mL samples of raw and filtered water tested positive at >500 and >100 cfu/100 mL, respectively. In contrast, a 100 mL sample of CF1 effluent on January 15th tested positive for *E. coli* at 50 cfu/100 mL, while a 1 mL sample²¹ was found to have twice as much – 100 cfu/100 mL. If the values of the January 17th and 18th CF2 data points are averaged (i.e., to 80% microbial removal efficiency), a more generally upward trend in *E. coli* removal efficiency is obtained, where the January 18th data point at 68% may be attributed to natural skew (see Figure 5.4, below).

Given that CF2 was already 4 days mature when challenged with *E. coli* rich source water, total ripening time for this filter is similarly approximated at 8-10 days (see Table 5.5). This estimate is uncertain, however, due to difficulties encountered in generalizing a trend from this filter's microbial removal data (see above).

See sections 5.5 and 6.3.2 for a discussion of H_2S presence/absence test results.

 $^{^{21}}$ 1 mL samples were added to approximately 50 mL sterile rinse water prior to filtering through membrane apparatus (see section 4.4.3).





5.3.1.3 Duplicate Sample Correlation

Fifty-nine samples were analyzed via the membrane filtration method during concrete BioSand filter experimentation. Of these 59, 17 were duplicate samples. Table 5.6 presents correlation coefficients for *E. coli* duplicate samples obtained during both Lumbini and MIT BioSand filter experiments. Where bounded values were obtained from microbial measurements, the lower limit was used to calculate correlation coefficients.

BioSand Filter Experiment	Correlation Coefficient	Correlation Coefficient (omitting high volume/low volume pairs with poor correlation)
Lumbini Concrete BioSand Filters	0.75 (n=16)	0.77 (n=14)
Lumbini BioSand Pitcher Filters	0.25 (n=3)	
MIT BioSand Filters	0.91 (n=20)	

Table 5.6 Correlation coefficients for duplicate E. coli membra	ne filtration samples.
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Poor correlation was sometimes observed between samples differing significantly in volume (e.g., 100 mL vs 1 mL). Table 5.6 also gives correlation coefficients for a subset of data which exclude high volume/low volume (e.g., 100 mL/1 mL) duplicate pairs for which the high volume sample showed evidence of *E. coli* contamination and the low volume sample did not (e.g., >100 cfu/100 mL for the 100 mL sample and 0 cfu/100 mL for the 1 mL sample). Duplicate correlation is somewhat higher when these pairs are excluded for the analysis, presumably because smaller sample volumes contain too few particles to provide statistically significant estimates of microbial concentrations.

A strong correlation was observed between sample size and duplicate correlation; correlation coefficients increased from 0.25 (field pitcher filter experiments) to 0.91 (MIT experiments), corresponding to increasing sample sizes of 3 and 20, respectively (see sections 5.3.2.4 and 5.4.2). This may be because larger sample sizes give more statistically significant estimates of precision.

5.3.1.4 <u>Flow Rates</u>

Flow rates for CF1 averaged 25 L/hr; CF2 averaged somewhat higher, at 28 L/hr. Flow rates were initially high and significantly decreased as time passed (as was the trend reported by Lukacs; see section 5.1.5). Flow of CF1 decreased from 41 L/hr on January 11th (day 1 of experimentation) to 21 L/hr on January 18th a 50% reduction. CF2 experienced a 68% decrease in flow, from 61 L/hr on January 11th to 20 L/hr on January 18th (see Figure 5.5).



Figure 5.5 Flow rate data from January 2003 Lumbini BioSand filter experiments.

The observed decreases in flow rate may be attributed to microbial growth within the biofilm and particle deposition (i.e., clogging). As the biological layer thickens and as particles deposit in the topmost layer of sand, the flow rate of a filter will decrease. Very turbid water contains a large amount of fine particles and silts, which are trapped in the uppermost layer of sand and biology. The higher the content of fine particles and silts, the more quickly the filter will clog. Again, if water contains large populations of biology, the biological layer that feeds on this content will rapidly grow or thicken. This too results in a kind of clogging at the top of the filter (Ritenour, 1998). As source water for experiments was obtained from a small (~15 ft diameter), eutrophic pond on IBS property, it likely contained relatively high concentrations of organic matter which contributed to filter clogging. Moderately high turbidities (see Table 5.7) also suggest the presence of fine suspended grains, which would have accumulated on sand surfaces and slowed transport.

Filter	Volume of water introduced each day (L)	Flow Rate (L/hr)	Turbidity (± 0.1 NTU)		
			Raw Water	Filtered Water	% Removal
Concrete Filter 1	21	25	19.2	3.3	89.3
Concrete Filter 2	21	28	18.7	2.0	93.0
Green Pitcher Filter riverbank sand	2	6	57.1	5.2	91.0
Blue Pitcher Filter sun-dried riverbank sand	2	5	57.1	4.3	92.6

 Table 5.7 Summary of average flow rates and turbidities measured during January 2003 Lumbini

 BioSand filter experiments.

5.3.1.5 <u>Turbidity</u>

Turbidity of source water varied considerably. Turbidities of CF1 raw water ranged from 0.0 NTU on January 11 (day 1 of experimentation) to 46.5 NTU on January 15th (day 5, see Appendix D). Similar data were obtained for CF2 raw water, which ranged from 1.8 NTU on January 13th (day 3) to 40.3 NTU on January 16th (day 6). Table 5.7 presents a summary of turbidity data obtained during experimentation.

Both filters were quite effective at removing suspended particles from influent water. Percent turbidity removal for both filters was high -89.3% for CF1 and 93.0% for CF2. The reader is referred to section 2.2 for a brief overview of particle removal mechanisms.

5.3.2 BioSand Pitcher Filter Experiments

5.3.2.1 BioSand Pitcher Filter Conceptualization

The author conceptualized the BioSand pitcher filter in response to the following drawbacks of the concrete BioSand filter:

- BioSand filter owners reported using filtered water for drinking only, or drinking and cooking. No one used filtered water for bathing or washing laundry.
- More than half of the Lumbini concrete household filters had flow rates below the design minimum (20 L/hr). Five out of 9 functioning concrete BioSand filters had flow rates less than 6 L/hr, and 4 of those 5 had flow rates less than 3 L/hr.
- Concrete BioSand filters are currently a relatively expensive technology for poor rural communities; filter cost is estimated at 2,500 Nepali Rupees or \$32 USD (Maitri, 2003).²² Cost estimates provided by Durga Ale (Appendix R) are considerably higher at NRs 3,380 (\$45), possibly because labor charges are included.
- Concrete BioSand filters are extremely heavy and cumbersome.²²
- The BioSand filter technology appears to be well-liked by users in Lumbini district communities. Many individuals expressed an interest in aquiring a filter.

 $^{^{22}}$ A fiberglass version of the BioSand filter was being prototyped during this research period to see if costs could be reduced (see Appendix R) and a lighter material employed. Estimated cost of the fiberglass unit is approximately NRs 2,000 (\$27) – NRs 1,700 for the filter, itself, and NRs 300 for the sand.
A plastic BioSand pitcher filter incorporates the following positive points if proved a viable water purification technology:

- The pitcher filter is designed to provide safe drinking water at the household level, and may compete in a water supply market with concrete BioSand filters used for drinking water purification.
- Flow rates of BioSand pitcher filters are comparable to (and in some cases exceed) flow rates of concrete BioSand filters in the field.
- The pitcher filter is cheap. Materials for prototype construction cost less than 80 Nepali Rupees (approximately \$1 USD) per filter. This price tag is at least 25 times less than that of the concrete full-scale version.
- The pitcher filter is light and easily manageable.
- The BioSand filtration technology is already well accepted by many communities and filter users are relatively comfortable with operating protocol (see section 6.3.4). Many of the same principles of construction, operation and maintenance for concrete BioSand filters apply to pitcher filters. This facilitates pitcher filter introduction, as technical knowledge to be transferred will be minimal.

5.3.2.2 Zone of Biological Activity

The depth of the pitcher filter fine sand layer is approximately four and a half times less than that of the BioSand filter. However, the reduced flow path length is not expected to result in smaller bacterial removal efficiencies as long as the 5 cm supernatant depth is maintained. Bellamy et. al. (1985) showed insensitivity of total coliform and *Giardia* removal to sand bed depths. Buzunis (1995) also found sand layer depth to be inconsequential (in terms of fecal coliform removal) except for the increased headloss

and reduction of flow provided by a deeper sand bed.²³ The depth of the concrete filter's biological layer (i.e., biological removal region) is mainly a function of the depth of water over the sand bed since this controls the rate at which oxygen can be drawn down to the biologically active zone and the depth into the sand oxygen can be supplied. While the well tested concrete BioSand filter has a biologically active zone less than 10 cm in depth, in filters with a more shallow standing water depth the biologically active layer is expected to be deeper (Buzunis, 1995). This would result in a longer contact time with the filter biology and improved filter efficiency.

5.3.2.3 Quantifying Bacterial Removal Efficiency of BioSand Pitcher Filters

E. Coli removal performance of two pitcher filters, a green pitcher filter (GF) and a blue pitcher filter (BF) were compared to each other and to the concrete BioSand filters. Three days of microbial removal efficiency data (from January $16^{th} - 18^{th}$, days 2 - 4 of testing) were obtained during 4 days of experimentation.

Removal efficiency for the GF (with riverbank sand) was quite high on January 16th (97%, see Figure 5.2) and decreased to 80% on January 17th and 18th. Performance of the BF (with dried riverbank sand) was similar – microbial removal efficiency was 98% on January 16th, decreasing to 82% and 59% on January 17th and 18th, respectively. The BF performed slightly better than the GF during the first two days of testing, but had a lower overall efficiency. Average microbial removal for the GF and BF were 86% and 80%, respectively, i.e., the pitcher filter with riverbank sand (GF) removed more biology from source water than the pitcher filter with dried riverbank sand (BF).

These results appear to contradict prevailing theories regarding appropriate filter media. Organic material is thought to provide food for microorganisms at depth in the filter,

²³ The head loss (i.e., pressure drop) that occurs when clean water flows through a clean filter media can be calculated from well-known equations (e.g., the Kozeny equation). The head loss for a clean bed depends on the flow rate, grain size, porosity, sphericity, and water viscosity. As filtration progresses and solids are deposited within the void spaces of the medium, the porosity decreases, and sphericity is altered. Head loss is very dependent on porosity, and reduction in porosity causes the head loss to increase (Water Quality & Treatment, 1999).

whereas in a properly functioning filter, the activity of the microorganisms is limited to the surface of the sand. Though the food is eventually consumed, the process can take months and could result in more contaminated water leaving the filter than going in (Baker, 2002).

Sample size is small, however. Three data points are likely not of sufficient size to account for natural skew or reveal accurate trends. Nevertheless, the data does provide some preliminary insights into pitcher filter viability, confirms the potential of the pitcher system for bench-scale experimentation, and serves as a starting point for a more in-depth investigation (see section 7.2).

Results do support previous findings by Coan and Stoller (2002), who studied dependence of *E. coli* and total coliform bacteria removal in concrete BioSand filters on type of filter media. Experiments utilized four different types of fine sand – riverbank sand from the Amuwa River in Lumbini, Nepal (control), river sand soaked in a 1% chlorine solution for 72 hours, river sand baked in a locally made oven, and sand obtained from a local rock crushing operation. The filter with river sand had the highest removal rates of *E. coli* and total coliform bacteria, followed by burned and crushed sand, and then chlorine soaked sand. Results from these experiments should be interpreted with caution, however, as testing duration was short (11 data points from 3 weeks of experiments) and the authors had difficulty obtaining quality assurance blanks.

Microbial removal performance of Lumbini pitcher filters was comparable to that of the concrete BioSand filters. On January 16^{th} , source water for all filters contained at least 500 cfu/100 mL *E. coli*. Concrete filters 1 and 2 removed 99.8% and 80% of influent *E. coli*, respectively; removal efficiency for the green and blue pitcher filters was 97% and 98%, respectively. Impressively, performance of both pitcher filters surpassed that of CF2 on this day.

Microbial removal efficiency of the BF did decrease to 59% on the last day of testing (January 18th), however, performance of CF2 also declined on this day – to 68% from

92% the previous day. This decline may be attributed to lower quality influent water used on the last day of testing. *E. coli* concentrations in pitcher filter influent water increased by at least 155 cfu/100 mL from January 17th to 18th; an increase of at least 55 cfu/100 mL was observed for CF2.

A larger increase in *E. coli* concentrations for CF1 influent water was observed: concentrations increased from at least 75 cfu/100 mL to at least 413 cfu/100 mL. However, removal efficiency of CF1 increased from 69% to 95% on January 18th. The higher concentrations of *E. coli* in the CF1 January 18th influent water may correspond to higher levels of dissolved organic matter. These organic substances may have stimulated biofilm development and facilitated *E. coli* removal.

In general, microbial removal performance for all filters was high, but variable. Further testing would have been necessary to verify actual trends in microbial removal capacity and determine relative contributions of random variability to data skewing. See section 6.4.1 for a discussion of the effects of source water microbial population density on *E. coli* removal efficiency.

Due to the brevity of the experimentation period, estimates of pitcher filter ripening times were not obtained (see Table 5.5). Subsequent laboratory studies were performed to further investigate ripening periods and microbial removal trends (see section 5.4).

See section 5.5 for a discussion of H₂S presence/absence test results.

5.3.2.4 Duplicate Sample Correlation

Twenty-three samples were analyzed using the membrane filtration method during pitcher filter experimentation. Of these 23, 3 were duplicate samples. Correlation between duplicate pairs was low, at 0.25 (see Table 5.6), however, sample size is small (n=3) and likely does not provide a statistically significant estimate of duplicate correlation. Good correlation between duplicate samples from Lumbini concrete

BioSand filter experiments and MIT experiments, and *E. coli* free quality assurance blanks (see Appendix O) lend credibility to pitcher filter experiment results overall.

5.3.2.5 Flow Rates

Flow rates of pitcher filters averaged between 5 and 6 L/hr (see Table 5.7), approximately one fifth of C1 and C2 concrete filter flows, but comparable to flow rates of Lumbini household BioSand filters (see Table 5.2). Flow of GF decreased from 6 L/hr on January 15th to 5 L/hr on January 18th, a 17% reduction overall (see Figure 5.5). BF experienced a 33% decrease in flow, from 6 L/hr on January 15th to 4 L/hr on January 18th. The observed decrease in flow rate may be attributed to biofilm growth and sedimentation (see sections 2.3 and 5.3.1.4).

5.3.2.6 Turbidity

Pitcher filters appeared equally effective at removing turbidity as concrete filters, and may have even surpassed the latter in turbidity removal capacity. While turbidity removal averaged 91.0% for the GF and 92.6% for the BF (as compared to 89.3% for CF1 and 93.0% for CF2, see Table 5.4), turbidity of pitcher filter source water was approximately 3 times that of concrete filter source water. In summary, pitcher filters were treating water of much higher turbidity content than the concrete filters, but still had approximately identical turbidity removal.

5.4 MIT BioSand Filter Experiments

5.4.1 Quantifying Bacterial Removal Efficiency

E. Coli removal performance of 2 plastic BioSand pitcher filters, one green (MIT-GF) and one white (MIT-WF, see Figure 2.4), were compared to that of a plastic Davnor filter (see Figure 2.2). Pitcher filters differed by type of plastic casing (the green from an

open-air market in Lumbini, Nepal; the white from an Economy Hardware store in Cambridge, Massachusetts) and holding capacity (0.5 L and 0.3 L for the green and white pitcher filters, respectively). Laboratory work was performed at the Department of Civil and Environmental Engineering at MIT.

For the period of March 7th to April 2nd, filters were challenged with room temperature Charles River water spiked with *E. coli* bacteria. During this time, *E. coli* concentrations in influent water varied from 4 to 345 cfu/100 mL, averaging 87 cfu/100 mL (target concentration was 100 cfu/100 mL, see Appendices L - N).

A generally upward trend in microbial removal efficiency was observed for the period of March 7^{th} – March 21^{st} (days 1 – 15 of experimentation). Removal efficiencies for the green and white pitcher filters increased from 0% and 10% on March 7^{th} to 85% and 62% on March 21^{st} , respectively (see Figure 5.6). These data points correspond to increases in Log_{10} Reduction Values (LRVs) of 0.0 to 0.8 and 0.0 to 0.4, respectively (see Figure 5.7). Removal efficiency for the Davnor filter was 75% on March 8^{th} (LRV of 0.6), but subsequently declined to 50% (LRV of 0.3) the next day. Removal performance gradually increased over the course of the next 12 days to 85% (LRV of 0.8) on March 21^{st} .



Figure 5.6 E. coli removal efficiency data obtained during MIT BioSand filter experiments.



Figure 5.7 *E. coli* removal efficiency data obtained during MIT BioSand filter experiments. Data are presented as log₁₀ reduction values.

On March 22nd, *E. coli* removal efficiencies for all 3 filters dropped significantly. Removal efficiencies for the Davnor and green pitcher filter both dropped from 85% on March 21st to 54% and 18%, respectively, the next day. These values correspond to 36% and 79% reductions in performance, respectively. The white pitcher filter experienced a 53% drop in performance, from 62% to 29% removal.

This drop in performance may have been due to disturbance of the biofilm. Though the author was frequently the only person working in the laboratory, the work space was utilized by department classes at other times. Jostling of the filters and subsequent disturbance of the surface biological community could have caused the drop in performance observed.

For the 11 days following (March 23^{rd} – April 2^{nd}), microbial performance for all three systems remained relatively static; a slight upward trend until March 24^{th} was detected, with subsequent declines in removal efficiency. The Davnor filter showed variable performance around 50% removal, with a high of 57% on the 24^{th} . Microbial removal performance for the green pitcher filter was similar – *E. coli* removal increased from 18%

on the 22^{nd} to 44% on the 24^{th} , then declined to 19% on the 1^{st} . Removal efficiency for the white pitcher filter increased from 29% on March 22^{nd} to 59% on the 24^{th} , then decreased to 28% on the 1^{st} .

On April 3rd and 4th, the filters were challenged with a 1:1 mixture of Charles River water and wastewater obtained from the Deer Island Wastewater Treatment Plant (Boston, Massachusetts). Raw water *E. coli* concentrations for these days were 1813 cfu/100 mL and 1188 cfu/100 mL, respectively, averaging approximately 1500 cfu/100 mL.

Microbial removal performance (% removal) for all filters improved in response to this influent. Removal efficiency for the Davnor increased by 30%, from 54% on April 1st to 77% on April 3rd. These data points correspond to a LRV increase of 50%, from 0.3 to 0.6. Similarly, a 68% increase in removal efficiency (75% increase in LRV) was observed for the green pitcher filter, and a 60% increase (80% increase in LRV) for the white pitcher filter.

Impressively, pitcher filter performance surpassed that of the Davnor filter on April 4th, the last day of experimentation. The green and white pitcher filters both reduced influent *E. coli* concentrations of 1188 cfu/100 mL to 40 cfu/100 mL, a 97% removal rate (LRV of 1.5), compared to a 95% reduction (LRV of 1.3) for the Davnor filter to 60 cfu/100 mL. Further testing would have been necessary to verify that these high removal efficiencies were not transient responses to influent water quality but truly indicative of filter performance.

Based on these data, total ripening times for the Davnor and pitcher filters when challenged with high quality (low turbidity, low organic matter, low nutrients, etc.) Charles River water may be approximated at 30 to 40 days (see Table 5.5).

5.4.2 Duplicate Sample Correlation

Ninety-nine samples were analyzed via the membrane filtration method during BioSand filter experimentation. Of these 99, 15 were duplicate samples. Correlation between duplicate pairs was quite high, at 0.91 (see Table 5.6). High duplicate sample correlation together with *E. coli* free quality assurance blanks (see Appendix P) lend credibility to filter experiment results.

5.4.3 Flow Rates

Flow rates for the Davnor filter averaged 15 L/hr; flow rates for the green pitcher filter were approximately 50% less, averaging 8.1 L/hr. The white pitcher filter had the slowest flow of the 3 filters, approximately 50% less than the green pitcher filter at 4.3 L/hr (see Table 5.8).

Flow rates were high initially and gradually decreased as time passed (see Figure 5.8). Flow of the Davnor filter increased from 17 L/hr on March 7th to 20 L/hr the next day, and subsequently decreased to 13 L/hr on April 4th, a 24% reduction. The green pitcher filter experienced a 45% decrease in flow, from 11 L/hr on March 7th to 6 L/hr on April 4th, and the white pitcher filter experienced a 40% decrease, from 5 L/hr on March 7th to 3 L/hr on April 4th.

Table 5.8 Summary of	average flo	w rates and	l turbidities	measured	during MIT	BioSand	filter
		expe	riments.				

Filter	Volume of water introduced each day (L)	Flow Rate (L/hr)	Turbidity (± 0.1 NTU)		
			Raw Water	Filtered Water	% Removal
Davnor Filter	8.6	15	4	0	100
Green Pitcher Filter	4.6	8.1	4.3	0.7	83.7
White Pitcher Filter	4.5	4.3	4.3	0.6	86.0



Figure 5.8 Flow rate data from MIT BioSand filter experiments.

5.4.4 Turbidity

Turbidity removal for all filters was quite high, with pitcher filter performance slightly less than that of the Davnor filter. Turbidity removal averaged 100% for the Davnor filter, and 83.7% and 86.0% for the green and white pitcher filters, respectively.

5.5 H₂S Test Reliability at Colder Temperatures

5.5.1 Comparison of Results from H₂S Method and Membrane Filtration Technique

The HACH 20 mL H₂S test has a P/A detection limit of 5 cfu/100 mL (IDRC, 1998 in Sullivan, 2002). However, poor correlation was observed between H₂S test results and membrane filtration analyses, even for highly contaminated samples. Ninety-seven H₂S tests were performed, each with a corresponding membrane filtration test. Thirty-seven or 38% of tests gave false negative results, i.e., results showed absence of H₂S bacteria when the corresponding membrane filtration test indicated more than or equal to 5 cfu *E. coli*/100 mL to be present (see Table 5.9). False positive results (i.e., results indicated

presence of H_2S bacterial contamination when enumerative method showed less than 5 cfu *E. coli*/100 mL to be present) were obtained from 4 or 4% of tests.

 Table 5.9 Percentages of H₂S tests resulting in false negative and false positive results when compared to the membrane filtration technique.

	False Negative	False Positive
Percentage of H ₂ S Test Results	38%	4%

Of the 37 false negative H₂S tests, 13 (35%) corresponded to membrane filtration counts of less than 100 cfu *E. coli*/100 mL; 24 (65%) corresponded to counts of greater than or equal to 100 cfu *E. coli*/100 mL (see Table 5.10). Of the 24 tests which underestimated contamination in highly contaminated samples, 14 (58%) corresponded to membrane filtration counts of greater than 400 cfu *E. coli*/100 mL.

Table 5.10 Percentages of false negative H2S test results corresponding to less than & greater than100 cfu E. coli/100 mL as measured by the membrane filtration technique.

	E. coli Concentration		
	less than 100 cfu/100 mL	greater than 100 cfu/100 mL	
Percentage of False Negative H ₂ S Test Results	35%	65%	

5.5.2 Effect of Time Lag between Sampling and Incubation on H₂S Test Results

Poor correlation was obtained between the H_2S method and enumerative technique even when less than approximately 30 minutes elapsed between sampling and incubation. Seventeen out of 37 false negative results (46%) had incubation time lags of less than 30 minutes, while 20 tests (54%) had time lags greater than 30 minutes (see Table 5.11).

Table 5.11 Percentages of false negative H₂S test results with sampling/incubation time lags less than and greater than 30 minutes.

	Time Lag between		
	Sampling and Incubation		
	less than 30 min	greater than 30 min	
Percentage of False Negative H ₂ S Test Results	46%	54%	

Five samples provided true positive results (5 out of 42 contaminated samples, or 12%), corresponding to membrane filtration counts between 10 and >1000 cfu *E. coli*/100 mL. The time lag between sampling and incubation for each of these tests was between 15 and 45 minutes. True negatives were obtained regardless of time period between sampling and incubation – lags between 7 minutes and 20 hours and 20 minutes resulted in true negatives.

5.5.3 Effect of Cold Temperatures on H₂S Test Results

In general, winter temperatures of approximately 10° C (50° F) and lower were thought to significantly decrease the accuracy of H₂S tests as surrogate detectors of fecal coliform contamination in drinking water. Growth of H₂S producers was likely retarded at lower temperatures, slowing production of iron sulfide precipitate even in highly contaminated samples (>1000 cfu *E. coli*/100 mL, see, for example, Appendix D).

These results suggest the unsuitability of H_2S tests for use in colder climates. However, better correlation between H_2S tests and enumerative microbial analyses is expected for ambient operating temperatures between 20 and 44°C (see section 5.5.4).

5.5.4 Comparison with Previous Work

Good correlation has been found between H_2S tests and enumerative bacterial analyses under warmer conditions. For example, Pillai et al. (1999) found good correlation between H_2S and membrane filtration test results at temperatures between 20 and 44 °C; test vials did not require a constant temperature incubator if the room temperature was within this range.

Similar results were found in previous field studies by MIT Nepal Project researchers. For example, Sullivan (2002) compared H₂S test performance of duplicate incubated and non-incubated samples at approximately 70°F (21°C). The correlation between these sets of tests was 84 percent and non-incubated samples were only slightly less likely to yield negative results than incubated samples, 45 percent positive among non-incubated samples versus 48 percent positive among incubated samples (Sullivan, 2002). Lukacs (2002) also found high correlation between H₂S P/A and fecal coliform membrane filtration tests; correlation for levels of fecal coliform contamination of >15 cfu/100 mL, >5 cfu/100 mL and 0 cfu/100 mL was 1.0, 0.88 and 0.82, respectively. Lee (2001) observed moderate correlations between H₂S tests and *E. coli* and total coliform assays (0.72 and 0.64, respectively).²⁴

 $^{^{24}}$ Lee (2001) assessed *E. coli* and total coliform removal efficiencies of concrete BioSand filters in the central Palpa region and Nawalparasi district of Nepal.

6 Discussion

6.1 Defining Access

Access to safe water remains an urgent human need in many countries. Tremendous human suffering is caused by diseases that are largely conquered when adequate water supply and sewerage systems are installed. It is the poor – the Nepali woman drawing water from an open sewage channel or the Bangladeshi child washing household utensils in a pool also used as a latrine – who bear the brunt of risks from contaminated water (World Bank, 1992). Poor women are particularly affected. It is primarily women who bear the daily burden of hauling heavy buckets long distances to meet the domestic water needs of their families (Crow, 2001).

The differences in access to safe water by income exist both within and across countries. The rural poor are more likely to rely directly on rivers, lakes, and unprotected shallow wells for their water needs and are least able to bear the cost of simple preventative measures such as boiling water to make it safe for drinking (World Bank, 1992). Waterborne disease is estimated to be killing one child every 8 seconds (Crow, 2001).

Large-scale water distribution networks often fail to provide users with adequate access to safe water. Old and deteriorated networks, poor operation and maintenance, insufficient cost recovery and low tariffs are usually put forward to describe water supply deficiencies. Where access to water does exist, services may be highly unreliable: water is not provided around the clock, low pressure, sudden breakdowns, seasonal variations, bad quality of water, etc (Zerah, 2000).

6.2 **Providing Access**

Because water is pivotal for health and livelihoods, insufficient access to water may be a significant cause of poverty and conflict (Crow, 2001). The need for a simple, inexpensive and effective water treatment technology is great. However, the academic

and research community in the industrialized world frequently focus their interest on high tech, centralized and expensive solutions to water problems that are not relevant to the millions of people who are most in need. This is especially true in the water area, where the kinds of new technologies (e.g., expensive reverse osmosis systems, ultra-violet sterilizers, etc.) being researched in many developing countries – using up precious skilled manpower and financial resources – bear little relationship or relevance to local needs and resource endowment (Munasinghe, 1990). Developed specifically to address local needs and with much local support, BioSand and potentially pitcher filtration technology have much to offer the developing world as a purveyor of safe household drinking water.

6.3 Discussion of Results from Lumbini Survey of Concrete BioSand Filters

6.3.1 Microbiological Results from Membrane Filtration Assays

Results from microbial analyses of Lumbini concrete BioSand filters are mixed. Of the 9 operating BioSand filters, 2 (BSF 5 & 6 – Khambe) were successfully removing 99% of *E. coli* from highly contaminated raw water, while 3 (BSF 2 & 3 – Sekhuwadand and BSF 7 – Sonbarshi) were contaminating relatively clean source water. BSF 7, specifically, is likely a significant health threat, adding more than 1000 cfu/100 mL to relatively clean raw water (0.5 cfu/100 mL) of low turbidity (2.8 NTU). Four filters (BSF 1 & 4 – Sekhuwadand, BSF 8 – Ramawa-pur and BSF 9 – Mujhana) were treating high quality source water with high bacterial removal efficiencies.

One day of testing for each of the Lumbini household BioSand filters, which was all the time afforded for that activity, is insufficient to adequately characterize BioSand filter performance, however. The Sonbarshi school filter (BSF 7; the worst result obtained), which significantly contaminated clean raw water may or may not consistently contaminate its filtrate. It may be that children in the school contaminated the outflow spout when they handle it, and filtered water is only contaminated upon exiting the filter. This hypothesis could only be verified (or disproved) by cleaning the spout and

evaluating subsequent filter performance on multiple occasions. It is unfortunate that this was not possible to do during the limited time available to the author.

6.3.2 Microbiological Results from H₂S Tests

Results from field experiments suggest the unsuitability of presence/absence H₂S tests for use in colder climates (i.e., temperatures at or below 10°C or 50°F), as very poor correlation was observed between H₂S and membrane filtration test results, even for highly contaminated samples (e.g., greater than 500 cfu *E. coli*/100 mL). Thirty-seven of the 97 H₂S tests performed (each with a corresponding membrane filtration analysis), or 38%, gave false negative results, i.e., results showed absence of fecal contamination when the corresponding membrane filtration test indicated more than or equal to 5 cfu *E. coli*/100 mL to be present.

These findings raise questions as to the validity of using H_2S producing bacteria as an index for drinking water pathogens of concern in colder climates. Current research by the World Health Organization (WHO, 2002) is investigating the utility of using heterotrophic bacteria counts as an alternative measure of drinking water safety.²⁵

Even so, numerous studies have found good correlation between H_2S tests and enumerative bacterial analyses under warmer conditions (see Ratto et al., 1989; Kromoredjo and Fujioka, 1991; Kaspar et al., 1992; Castillo et al., 1994; Venkobachar et al., 1994; Martins et al., 1997; Rijal and Fujioka, 1998; Genthe and Franck, 1999; Pillai et al., 1999; and Sullivan, 2002), and many researchers recommend the H_2S method as a reasonable approach for detecting fecal contamination in drinking water. In addition, the tests may be ideal for facilitating community involvement in the monitoring and management of drinking water supplies and treatment systems because of their simplicity and ease of interpretation. Because test results are based simply on the observable

²⁵ Heterotrophs are broadly defined as microorganisms that require organic carbon for growth. They include bacteria, yeasts and moulds. A variety of simple culture-based tests which are intended to recover a wide range of microorganisms from water are collectively referred to as "heterotrophic plate count" or "HPC tests" procedures (WHO, 2002).

formation (or lack thereof) of a black precipitate, even poorly educated (e.g., illiterate) individuals may learn to successfully use and interpret the results of H_2S analyses.

As with any analytical tool, the H_2S test's limitations (e.g., non-fecal sources of hydrogen sulfide producing false positives, cold-induced false negatives, etc.) should be thoroughly understood before applying the method to analysis of any particular water supply.

6.3.3 Social Acceptability

While this work focused on a technical evaluation of BioSand and pitcher filter system performance, a brief note on social acceptability is appropriate. Lumbini district communities appear to be interested in and accepting of the BioSand technology. Many villagers expressed an interest in aquiring BioSand filters, and filter users were generally eager to learn (or be reminded of) cleaning and maintenance protocol. In addition, many individuals reported improved health and reduced illness following filter introduction. Section 7.1.4 discusses recommendations for a health-based evaluation of project success.

6.3.4 Issues with Knowledge Transfer

As with the 10 household BioSand filters in Lumbini villages, sand preparation for field experiments was performed by Durga Ale (see Figure 4.6), a local technician trained in concrete BioSand filter construction. Subsequent flow rates of the experimental filters (less than 10 L/hr) and non-uniform grain sizes in the surface sand layer indicated improper sand preparation techniques (see section 4.2.1.1).

These findings raise questions as to the integrity of many of the Lumbini household filters, as Durga Ale was responsible for sand preparation (and concrete shell molding) for all 10 filters. Lukacs (2002) notes that, "Durga Ale sifted and washed the filter media in his Nawalparasi workshop prior to filter installation. Ale's method of preparation is

unknown at this time." As Durga Ale incorrectly prepared the filter media for the author's experimental filters, it is likely that sand in some of the village filters was also prepared incorrectly. This might explain the marked reduction in flow rates of new filters observed by Lukacs in 2002, and the low flow rates measured by the author in 2003. However, flow rate reductions were also observed for filters commissioned according to CAWST instructions. This would suggest that clogging is an issue which needs to be addressed as a maintenance concern.

6.4 Discussion of Results from Field and Laboratory Experiments on Full-Size BioSand Filters

6.4.1 Dependence of Ripening Period on Source Water Quality

A strong correlation was observed between biofilm maturation periods and source water quality. Lower quality influent water (high turbidity, high levels of organic matter and nutrients, high levels of bacteria, etc.) facilitated biofilm ripening. For example, the concrete BioSand filters studied at IBS (which were challenged with highly turbid, stagnant pond water) were removing 95% and 50% of influent *E. coli* after 8 days of testing; removal efficiency for the pitcher filters was 80% and 59% after only 3 days. Conversely, both the plastic Davnor filter and pitcher filters (challenged with higher quality Charles River water) were removing less than 55% of influent *E. coli* after 26 days of testing.

Weber-Shirk and Dick (1997) note that filters with high quality source water may not achieve effective particle removal if raw water contains few particles, organic matter, bacteria, etc. The long ripening times for the MIT filters as compared to the Lumbini filters (30 - 40 days vs 8 - 10 days, respectively) support the idea that source water organic matter and microbial content strongly influence biofilm maturation. In the present case, the high quality Charles River water (with low levels of dissolved organic matter and naturally occuring microbes) did not contain sufficient nutrients to support elevated populations of microorganisms in the supernatant. It was only when challenged

with an organic rich, low quality (high microbial content) influent that significant bacterivore growth in the biologically active zone was possible. This would suggest the viability of a low tech approach to accelerating biofilm ripening²⁶ consisting of using low quality source water initially to facilitate bacterivore and surface film growth, and subsequently using higher quality water for daily use. More detailed research in this area is needed to determine the influent bacterial and turbidity levels necessary to accelerate ripening but minimize threats to public health.

6.4.2 Bacterial Removal Efficiencies

Results from field and laboratory experiments on concrete and Davnor BioSand filters support the use of this technology to provide households with safe drinking water. Data strongly suggest that fully ripened BioSand filters will significantly improve the quality of influent water, reducing turbidity by at least 90% (to less than 5.0 NTU²⁷) and influent *E. coli* concentrations by at least 95%.

6.5 Feasibility of BioSand Pitcher Filter

6.5.1 Bacterial Removal Efficiencies

In general, results from field and laboratory experiments on BioSand pitcher filters are encouraging and suggest the viability of pitcher filters as a household water purification system. *E. coli* removal rates of pitcher filters were comparable to those of the concrete and Davnor BioSand filters. Average *E. coli* removal efficiencies of pitcher and concrete BioSand filters from the last three days of field testing (January $16^{th} - 18^{th}$) were virtually identical. Removal efficiencies for the blue and green pitcher filters were 80 and 86%, respectively, as compared to 81 and 87% for concrete filters 1 and 2, respectively.

²⁶ Jellison et al. (2000) investigated the use of synthetic polymers to enhance ripening of slow sand filters and facilitate bacterial removal. A successful ripening agent, polymer Pol-E-Z 652, was determined to function by agglomerating particles in the raw water and hastening their removal at the filter surface so as to develop the surface film.

²⁷ WHO guideline for turbidity in drinking water is less than 5.0 NTU (WHO, 2002a).

Laboratory pitcher filter *E. coli* removal efficiencies were comparable to, and in some cases exceeded plastic Davnor BioSand filter removal rates. For example, both the green pitcher filter and the Davnor filter had removal efficiencies of 85% (\log_{10} reduction value of 0.8) after 15 days of experimentation (on March 21st). Removal efficiency of the white pitcher filter for this day was somewhat less – 62%. When challenged with dilute wastewater on day 28 (April 3rd), pitcher filters reduced influent *E. coli* concentrations of 1188 cfu/100 mL by 97% (1.5 log₁₀ reduction units), to 40 cfu/100 mL. Removal efficiency for the Davnor filter was slightly less – 95% removal to 60 cfu/100 mL (\log_{10} reduction value of 1.3).

6.5.2 Turbidity Removal

Field pitcher filters appeared equally effective at removing turbidity as concrete filters; like the concrete filters, pitcher filters successfully reduced influent turbidity to less than 5.0 NTU. Impressively, field pitcher filters were challenged with water that was approximately 3 times as turbid as concrete filter raw water (57 NTU vs 19 NTU), yet approximately identical turbidity removal was observed (91% and 93% removal for green and blue pitcher filters vs 89% and 93% removal for concrete filters 1 and 2, respectively).

Turbidity removal for laboratory pitcher filters was also high, at 84% and 86% removal for the green and white pitcher filters, respectively (to less than 1.0 NTU). However, source water for laboratory filters was of low turbidity to begin with (approximately 4.3 NTU vs 57.1 NTU for field tests).

6.5.3 Flow Rates

Flow rates of field and laboratory pitcher filters were generally between 4 and 8 L/hr. These flows are comparable to those of several Lumbini household concrete filters (of the

9 functioning BioSand filters visited, 5 had flow rates less than 6 L/hr). While less than the ideal flows of concrete and plastic BioSand filters (20 - 40 L/hr), pitcher filter flows are significantly higher than those of alternative low-tech household water filters. Flow rates of ceramic filters, for example, are generally between 1 and 4 L/hr (see Dies, 2003).

6.5.4 Pitcher Filter Viability

Comparable microbial and turbidity removal performance between pitcher filters and concrete CAWST and plastic Davnor filters support the use of a pitcher system testing platform to model the commercially available BioSand technology. Pitcher filters might also be used as an interim measure until a household mobilizes funds for a larger capacity water filter (e.g., a concrete BioSand filter), however, further testing and field verification in this area is needed.

Limitations of the pitcher filter system also deserve consideration. Due to its small size, the pitcher filter biofilm may be at a greater risk of disturbance (i.e., from jostling) than the heavier concrete or plastic BioSand filter. Disturbances to the supernatant may decrease microbial and turbidity removal effectiveness. Secondly, holding capacity of the pitcher filter (approximately 0.5 L as currently designed) is approximately twenty times less than that of the concrete BioSand filter. Filtration times for larger volumes of water will thus be increased.

7 Conclusions and Recommendations

7.1 Potential of Lumbini BioSand Filter Pilot Project

7.1.1 Need for Consistent Monitoring Efforts

While field and laboratory experiments on concrete CAWST and plastic Davnor BioSand filters strongly suggest that fully ripened BioSand filters will significantly improve the quality of influent water, further testing of the Lumbini household filters is appropriate. One day of testing for each filter, which was all the time afforded for that activity, is insufficient to adequately characterize BioSand filter performance. Regular, repeated samplings of source water, filtered water and water in collection buckets still needs to be performed on these pilot household units.

Dry and monsoon season sampling rounds are also recommended, as water sources may vary seasonally with respect to levels of bacterial contamination (Maitri, 2003). Seasonal sampling will better gauge filter effectiveness when challenged with variable quality source water.

Similarly, while Lumbini district wells generally tested free of *E. coli* contamination and showed low levels of total coliform bacteria to be present, one day of sampling is insufficient to adequately characterize well water quality. Regular monitoring should be performed to verify which wells pose actual threats to human and environmental health. For example, the Bhagawanpur well (B16) which tested at 28 cfu *E. coli*/100 mL may or may not be a health threat if this contamination is transient. Consistent monitoring efforts are needed to verify all findings.

Additionally, wells testing free of *E. coli* during the dry season may not be so during the monsoons. During the rainy season it is likely that fecal contamination reaches wells which were previously clean. Repeated sampling would help to identify those wells

which vary seasonally in terms of fecal contamination, and are therefore a seasonal threat to public health.

7.1.2 Need for Consistent Sampling Protocol

The objective of sampling a groundwater-fed source is to collect groundwater that is representative of the quality of water supplied by that source at a point in time for analysis (Taylor, 2003). Groundwater sampling efforts (e.g., of handpumps) must attempt to ensure representative sampling of subsurface aquifers.

In some cases, well purging may be appropriate to extract stagnant bore water whose chemical, physical and/or biological properties do not accurately represent the source in entirety. For sources which are not continuously flowing, it is recommended that a consistent volume of water be pumped prior to sampling wells, and, critically, that this value be recorded as part of well water surveillance. Implementation of a consistent purge volume will help to improve comparisons of well water quality data between sites and regions (Taylor, 2003). However, higher quantity well purging may not always be appropriate, e.g., in regions where groundwater resources are scarce.

Sampling programs should also include consistent sterilization protocol. For example, if users are suspected of introducing contamination at the source, paired water samples should be obtained (a) as collected by the users, and (b) collected in a manner that carefully avoids contamination at the spout, e.g., after flaming the spout and collecting into a sterilized container (Carter, 2003).

7.1.3 Need for Simple and Effective Monitoring Tool

Because of their simplicity and ease of interpretation, tests for the presence or absence of hydrogen sulfide (H_2S) producing bacteria may be ideal for encouraging stakeholder (i.e., user) participation in water quality and treatment system monitoring. Numerous studies

have found good correlation between H_2S tests and enumerative bacterial analyses under warmer conditions (see section 6.3.2), however, findings from this work suggest the unsuitability of the method for use in colder climates (i.e., temperatures at or below 10°C or 50°F). Further investigations of temperature effects on the reliability of H_2S tests to accurately detect fecal contamination in drinking water are appropriate. Specifically, studies conducted in a controlled laboratory setting are recommended.

7.1.4 Need for Community Health Data

While microbial data suggest the viability of the BioSand filtration technology, health outcome data showing disease reduction in intervention communities are necessary if this technology is to gain international and scientifically credible acceptance. Ideally, a study design should include a control group not employing the intervention so as to provide a direct measure of the proportion of household and community illnesses (e.g., diarrhea) reduced using the water treatment technology.

Standard epidemiological methodology would suggest the following possible study design: Choose two communities of, say, 50 families each, with essentially the same demographic and population features (age, gender, race, socio-economic status, household size, etc.). Monitor their water quality and diarrheal illness rates for a sufficient time period to establish that they do not differ. These would be considered baseline studies. Then, choose one set of 50 families as the intervention community and the other as the control community, and implement the intervention on the community selected. Continue to monitor water quality and diarrheal illness in both communities. Collect enough data to be able to show statistically significant differences in water quality and in diarrheal illness rates in community members. The unit of observation can be the household, but the study design is really based on the outcome for the two communities (Sobsey, 2003).

7.1.5 Need for Women's Involvement in the Management of Water Systems

Lukacs (2002) observed that during the January 2002 BioSand filter installation, brief tutorials were given to male filter owners but not their wives. When questioned later about filter operating procedures it was common for women to say, "my husband knows, but he is not here" (Lukacs, 2002), even though the women were primarily responsible for household water supply. Their traditional roles in water provision are the obvious rationale for involvement of women in the introduction of improvements to water supply and in concurrent arrangements for operation, maintenance and health education (Van Wijk-Sijbesma, 1985). As key stakeholders in the BioSand technology and the domestic managers responsible for its operation, the Lumbini district women must be included in the management of the BioSand project.

One way to facilitate women's involvement in water system management might be to train several IBS women motivators in the specifics of BioSand technology. As fully trained technicians they might then transfer this knowledge to female filter owners who, in turn, could use this knowledge to increase the beneficial health impact of the project. In addition to filter manufacturing and commissioning, women motivators (or other trained technicians) might be given the specific task of visiting filters and observing flow rates. Flow rates are one indicator (if only a crude one) of filter performance, and indicate when cleaning and maintenance should be performed. Thus a system of regular monitoring and feedback could be initiated in which women play an integral role in maintaining their household's water purification system.

7.2 Future Developmental Work for the BioSand Pitcher Filter

While results from field and laboratory experiments suggest the viability of pitcher filters as a household water purification system and BioSand testing platform, further investigation is appropriate to more fully develop the technology.

Areas meriting future investigation are as follows:

- What is the optimal holding capacity for the pitcher filter? That is, to what extent does decreasing the depth of the fine sand layer (and thus increasing liquid holding capacity) affect microbial removal?
- Because of its small size, the pitcher filter biofilm may have a greater risk of disturbance (i.e., from jostling) than the heavier concrete or plastic BioSand filter. To what extent do slight disturbances affect microbial removal performance of the pitcher filter system?
- To what extent does increasing or decreasing the distance between the supernatant surface and the diffuser plate affect microbial removal performance?
- How is microbial removal performance of the pitcher filter affected by pause times? Are these effects comparable to those experienced by concrete filters?
- How feasible is a low-tech accelerated ripening approach consisting of challenging pitcher filters with lower quality water (e.g., dilute wastewater) prior to daily use? What sorts of time-frames should be considered for this method (days, weeks, etc.)?



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9 Appendices

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Appendix A. IBS Village Maps – Lumbini, Nepal

(modified from Sullivan, 2002)

The following maps were hand-drawn by IBS staff. Legends symbols vary from map to map. The general legend symbols are given in English and transliterated Nepali below.

Map Legend

Building or Structure - garr A Temple - mandir R Tree - ruk Road - bato Drainage Channel - nali River - nadi Ca Pond, Surface Water Source - pokari Open Well - kuwa \bigcirc Tubewell - boring Handpump - nul F Farmland or Field - kith BioSand Filter - filter













सीनवर्षी- SONBARSHi



	Date			Age			
Filter	Visited	Village	Owner	(yr)		GPS	Comments
BSF1	1/6/2003	Sekhuwadand		1	N E	27 [°] 29.384' 083 [°] 13.723'	inside house in cool, dark comer; good condition ^e ; D&C ^b ; 1 house (~20 people) ^c ; CF ^d = 2 <i>l</i> mnth; M ^e = 1 woman; RW ^f = hand pum g G ^g
BSF2	1 <i>/</i> 6/2003	Sekhuwadand		1	N E	27 [°] 29.396 083 [°] 13.738	good condition; 2 houses (13 people); CF = 4 <i>i</i> mnth; M = several people; post filter ^h = less giardiasis incidence
BSF3	1 <i>/</i> 6/2003	Sekhuwadand	Ram Chandra	1	N E	27 [°] 29.374 083 [°] 13.747	good condition; 3 houses (18 people); D&C M = 1 man; post filter = less sickness; casing reads "Group A Non Disinfection Finnida"
BSF4	1 <i>/</i> 6/2003	Sekhuwadand	Kushγlia	1	N E	27 [°] 29.348 083 [°] 13.754'	blue; good condition; D&C CF = 4/mnth; M = 1 woman
BSF5	1/6/2003	Khambe	Pusspalata	1	N E	27°26.986 083°14.913	casing intact; corner missing from diffuser plate; 1 house; RVV = hand pump
BSF6	1 <i>/</i> 6/2003	Khambe		1	N E	27 [°] 26.978 083 [°] 14.942	good condition; 10 people; CF = 4 <i>i</i> m nth
BSF7	1/8/2003	Sonbarshi	School	1	N E	27 [°] 30.837' 083 [°] 18.844'	good condition; 20 people; used intermittently, RW = Artesian well (~300 ft) spout
BSF8	1/8/2003	Ramawa-pur	Kanchan	1	N E	27 [°] 31.149 083 [°] 18.581'	spout stopped w/straw, good condition; D; 2-3 houses; CF = 2/mnth; M = 1 woman; RW = hand pump (186 ft); post filter = less sickness
BSF9	1/9/2003	Mujhana	School	1	N E	27°26.454 083°17.863	spout stopped w/strawis.t. standing water > 5 cm; good condition; RW = local hand pump
BSF10	1/10/2003	BuddhaNagar	IBS	1	N E		in IBS kitchen; good condition; not in use

⁹good condition = concrete casing intact, cover and diffuser plate in place, diffuser plate intact ⁹D&C = filtered water is used for drinking (D) and cooking (C) only, not washing (W) or bathing (B) ⁹number of houses or individuals served by filter ⁹CF = cleaning frequency

Filter maintained (M) by 1 woman RW = raw water source [®]GI = filtered water is not chlorinated ^hpost filter = following filter introduction

Well	Date Visited	Туре	Village	Depth (ft)	Age		GPS	Comments
DW9	1/6/2003	Artesian	Dhodahawa	350	4	N E	27°29 D19' D83°13.960'	
B14	1/9/2003	IBS hand pump	8 hagawanpur	200	4	N E	27°26 .492' 083° 15 .392'	
B15	1/9/2003	IBS hand pump	Bhagawanpur	230	4	N E	27°26.530' 083°15.351'	
B16	1/9/2003	IBS hand pump	Bhagawanpur	190	4	N E		
L1	1/9/2003	IBS hand pump	Lamtihawa	200	2	N E	27°25,931' 083°17.558'	plain opposite school
L2 (broken)	1/9/2003	IBS hand pump	Lamtihawa	180	2	N E	27°25,970' 083° 17,562'	
MB	1/9/2003	IBS hand pump	Mujahana	230	4	N E	27°26.440° 083°17.620'	
BUD1"	1/1 4/2003	local hand pump	BuddhaNagar			N E	27°28 276' 083°17,176'	junction of highway and main street
BUC2	1/1 4/2003	local hand pump	BuddhaNagar			N E	27°28 294' 083°17.176'	on left [®] in front of IBS clinic entrance
BUD3	1/1 4/2003	local hand pump	Buddha Nagar			N E	27°28 280' 083°17.232'	on right ^e btwn restaurant & house w/pink porch, green walls

Well	Date Visited	Туре	Village	Depth(ft)	Age		GPS	Comments
BUD4	1/1 4/2003	local hand pump	BuddhaNagar			N	27°28.271' 083°17 297'	n ear utility pole on right ^e in front of mud-walled bouse, brick bouse
BUD5	1/1 4/2003	local hand pump	Buddh a Nagar			NE	27°28.289' 083°17.329'	on right [®] in btwn 2 mud-walled houses
BUD6	1/1 4/2003	local hand pump	Buddh a Nagar			N E	27°28.291' 083°17.370'	on right ^e in btwn brick building & mud-walled house
BUD7	1/1 4/2003	local hand pump	Buddh a Nagar			N E	27°28.327' 083°17.419'	across from power station on left ^e adj.to narrow building w/white walls & blue doors
BUD_SK Siddarth Kumar	1/1 4/2003	private hand pump	Buddh a Nagar			N E	27°28.298' 083°17 256'	inside house
BUD_CK Chandra Kala	1/1 4/2003	private hand pump	Buddh a Nagar			N E	27°28.273' 083°17.141'	in front of house where we ate dinner
MUH4	1/16/2003	IBS hand pump	Muhuwari	195	1	N E	27°26.434' 083°13.675'	
MUH5	1/16/2003	IBS hand pump	Muhuwari	191	1	N E	27°26.486' 083°13.613'	
MUH6	1/16/2003	IBS hand pump	Muhuwari	150	3	NE	27°26.432' D83°13.661'	
MUH7	1/16/2003	IB S hand pump	Muhuwari	195	3	NE	27°26.465' 083°13.614'	
MUH8	1/16/2003	IB S hand pump	Muhuwari	203	3	N E	27°26.404' 083°13.686'	

^aall local Budda Nagar hand pumps along main street ¹when facing away from Butwal-Kathmandu highway

Date	FlowRate (L/hr) ^c	Turb	idity (± 0.1	I NTU) ^c	Microbial Measurements ^a									
		Raw Water	Filtered Water	% Removal	Raw ¹ H _z S Tests (P/A)	Water E.Coli (cfu/100 mL)	Filtered H _z STests (P/A)	E. Coli (cfu/100 mL)	% Removal E. Coli ⁵	Log Reduction Value [∎]				
1/11/03	41	0.0	0.0	100.0	А	0	А	> 100						
1/12/03	25	5.9	0.9	84.7	А	O	Р	> 95						
1/13/03	27	1.3	3.4	-161.5	А	0	А	O						
1/14/03	24	1.2	2.3	-91.7	А	.0	А	O						
1/15/03	19	46.5	10.9	76.6	А	> 100	А	> 75	25	0.1				
1/16/03	18	44.1	3.3	92.5	А	> 500	А	> 100	80	0.7				
1/17/03	21	30.3	4.7	84.7	А	> 75	А	23	69	0.5				
1/18 <i>/</i> 03	21	24.0	0.7	97.1		> 413		21	95	1.3				
average	25	19.2	3.3	89.3 ⁴										

*Where duplicate samples were obtained, results are presented as mean values

Where microbial measurement results are presented as bounded values, the lower limit was used to calculate removal efficiency

"Where multiple tests were performed, results are presented as mean values

In bold = E. Coli rich source water introduced

*Calculation excludes data points with negative values of % removal

[•]Log Reduction Value (LRV) = log _{(raw} water E. Coli concentration/filtered water E. Coli concentration)

Where filtered water E. Coli concentrations are 0 cfu/100 mL, a value of 1 cfu/100 mL was used to compute LRV.

1 LRV= 90 % reduction

2 LRV= 99 % reduction

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3 LRV= 99.9% reduction

4 LRV= 99.99 % reduction

D ate	Flow Rate (L/hr) ^c	Turbidity (± 0.1 NTU) ^c			Microbial Measurements ^a									
					Ra	տ մ	/ ater	Filtere	d W	í ate r	% Removal E. Coli ^b	Log Reduction Value		
		Raw	Filtered	% Domenter	H _Z S Tests		E.Coli	H _Z S Tests		E. Coli				
1/11/03	61	6.0	0.8	86.7	A (1770)	>	20	A A	>	500 mL)				
1/12/03	21	14.0	0.5	96.4	P		0	P		0				
1/13/03	31	1.8	3.9	-116.7	A		0	A		0				
1/14/03	28	7 .8	0.0	100.0	A		Ô	A		0	•••			
1/15/03	22	35.5	4.5	87.3	A	>	100	A		0	99	2.0		
1/16 <i>/</i> 03	22	40.3	2.9	92.8	A	>	500	A		0	99.8	2.7		
1/17 <i>/</i> 03	20	30.3	2.8	90.8	A	>	100	A		8	92	1.1		
1/18/03	20	14.0	0.4	97.1		>	155		>	50	68	0.5		
average	28	18.7	2.0	93.0			214							
W iere dipli W iere micro W iere mith n bold – E. Calcitatione Log Redictio W iere filtere 1 LRV – 90% 2 LRV – 99%	cate sam ples u oblaim easurem ple tests were Colincisourc excludes data ou Value (LRV) dwater E. Col e reduction reduction 3% reduction	vere obtali estresit performed ewateris obitswiti solitswiti solitswiti solitswiti solitswiti solitswiti	ed, res lits sare prese , res lits are trodiced legathe ua www.ater E. ations are D	are presenter n bed as bonne presented as nessor % ren Coll concentr c11/100 m L, :	d as mea uai dedualles, th s mea uailes nouai natio (Yiliteredu a uaile of 1 cf	vate	verim Itwas use r E. Colicouceu J m L was used	d to calc∎late tratio∎) to com p∎te LR1	rem 0 /.	uai efficie i cy				

Appendix E. Summary of data from January 2003 Lumbini BioSand filter experiments – Concrete Filter 2

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Well	Turbidity (± 0.1 NTU)	HACH H ₂ S Tests ^a								
		Time Sampled	Time Incubated	Timelar	Test Result	Test Result				
DW9		14:27	1/7/2003 8:53	18:26	241115 A	401115 A				
B14	0.0									
_	0.0									
B15	0.0	13:00	17:00	4:00	A	A				
B16	0.2									
L1	2.0	14:55	17:00	2:05	А	А				
L2										
МЗ	0.0	15:20	17:00	1:40	А	A				
BUD1	0.0	15:10	16:20	1:10	А	А				
BUD2	0.4	15:00	16:20	1:20	А	А				
BUD3	1.5	14:45	16:20	1:35	А	А				
BUD4	0.0	14:40	16:20	1:40	А	A				
BUD5	0.0	14:30	16:20	1:50	A	А				
BUD6	2.4	14:20	16:20	2:00	А	А				
BUD7	1.9	14:10 14:10	16:20 16:20	2:10 2:10	A A	A A				
BUD_SK	1.1	14:50	16:20	1:30	A	А				
BUD_CK	0.0									
MUH4	0.4	12:50	15:30	2:40	А	Α				
MUH5	2.4	12:40	15:30	2:50	А	А				
MUH6	0.9	12:20	15:30	3:10	А	А				
MUH7	0.7									
MUH8	0.6	13:00	15:30	2:30	А	А				

Appendix F. Data from wells tested during January 2003 in Lumbini district

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^aSample volumes are 20 mL.

^bUnless otherwise specified, sample volumes are 100 mL.

Well	Sample Name		Millip	ore Membran	e Filtration ^D	
		Time Sampled	Time Incubated	Time Lag	E. Coli (cfu/100 mL)	Total Coliform (cfu/100 mL)
DW9		14:27	1/7/2003 11:41	21:14	0	15
B14		12:30	18:30	6:00	0	55
B15		13:00	18:05	5:05	4	>1004
B16		12:54	18:19	5:25	28	>628
L1	L1 - 100 mL L1 - 10 mL	14:55 14:55	17:30 17:42	2:35 2:47	0 10	>100 160
L2						
МЗ	M3 - 100 mL M3 - 5 mL	15:20 15:20	17:00 17:10	1:40 1:50	0 0	>20 0
BUD1		15:10	15:23	0:13	0	1
BUD2		15:00	15:23	0:23	0	140
BUD3		14:45	16:30	1:45	0	0
BUD4		14:40	17:15	2:35	0	3
BUD5		14:30	16:25	1:55	0	50
BUD6		14:20	17:15	2:55	4	239
BUD7	BUD7 - 100 mL BUD7 - 100 mL	14:10 14:10	16:05 16:35	1:55 2:25	0 0	25 30
BUD_SK		14:50	16:00	1:10	0	0
BUD_CK					0 ^c	280 [°]
MUH4		13:00	15:00	2:00	0	85
MUH5		12:50	15:12	2:22	0	4
MUH6		12:20	15:12	2:52	0	1
MUH7		12:40	15:35	2:55	0	2
MUH8		13:10	14:45	1:35	1	33

^aSample volumes are 20 mL. ^bUnless otherwise specified, sample volumes are 100 mL. ^cSampled by Xanat Flores

Appendix G. Sol	ar Radiation ir	l Lumbini, Nepal	during January	2003
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Day	UV-A Radiation	Observations
	Whr/m2	
1/6/2003	832	Cloudy
1/7/2003	891	Cloudy
1/8/2003	443	Very Cloudy (battery)
1/9/2003	1449	Cloudy and little sun at 16hrs
1/10/2003	2166	Very foggy morning, sunny afternoon
1/11/2003	544	Very foggy
1/12/2003	794	Foggy morning, cloudy afternoon
1/13/2003	695	Foggy and very cloudy
1/14/2003	813	Cloudy and foggy
1/15/2003	2559	Cloudy and sunny after 12hrs
1/16/2003	796	Very foggy anf cloudy
1/17/2003	1154	Cloudy and dim sun afternoon
1/18/2003	3150	Cloudy and sun after 13hrs
1/19/2003	523	(Half day) Very foggy and then sun





Appendix H. Instructions for BioSand Pitcher Filter Construction

Materials

plastic pitcher, 5 inch diameter ¹/₄ inch diameter plastic tubing gravel, coarse sand, fine sand metal tie (flexible wire) phillips-head screwdriver or hand/power drill pliers knife or razor blade

Instructions

- 1. Cut slits in 4 inches of plastic tubing. Slits should be $\sim \frac{1}{4}$ in. apart and span one third the circumference of the tubing.
- 2. Use screwdriver or drill to make ¹/₄ in. wide holes ³/₄ 1 in. apart in the pitcher lid (diffuser). There should be approximately 27 holes in the diffuser plate.
- 3. Make a hole at base of pitcher filter. The hole must be just large enough to squeeze tubing through. If the hole is too wide the filter will leak.
- 4. Use pliers to pull 4 in. of slitted plastic tubing into pitcher.
- 5. Wash gravel, coarse sand and fine sand according to the recommendations of Ritenour (1998).
- 6. Install a 1 in. layer of gravel covering the intake tubing.
- 7. Add water to cover gravel layer.
- 8. IMPORTANT: Always add sand to water. Add water as needed so that surface of sand layer is always covered with water.
- 9. Add 1 in. of coarse sand.
- 10. Add 4 in. of fine sand.
- 11. Use metal tie to attach plastic tubing to filter handle. The outflow spout should be placed such that at rest, there is a 2 in. (5 cm) layer of standing water.
- 12. Install diffuser plate. Always pour water through diffuser plate.

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Filter	Flow Rate (L/hr)		Turbidity (± 0.1 N	TU)
		Raw Water	Filtered Water	% Removal
BSF1	5.6	3.9	1.7	57.7
BSF2	1.0	360.0	0.8	99.8
BSF3	14.5	15.0	1.7	88.7
BSF4	24.1	176.0	2.3	98.7
BSF5	2.8	179.0	2.3	98.7
BSF6	2.4	1.2	1.8	(-50.0)
BSF7	2.4	2.8	1.3	53.6
BSF8	37.5	3.4	2.1	38.2
BSF9	34.9	5.0	2.0	60.0
BSF10	0.0	N/A	N/A	N/A

Appendix I. Summary of data from Biosand filters tested during January 2003 in Lumbini district

^aTime lag between sampling and incubation exceeded 2 hours

^bValue does not include result obtained after 2 day lag between sampling and incubation.

^cWhere duplicate samples were obtained, results are presented as mean values ^dWhere microbial measurement results are presented as bounded values, the lower limit was used to calculate removal efficiency

^eLog Reduction Value (LRV) = log_{10} (raw water E. Coli concentration/filtered water E. coli concentration)

Where filtered water E. Coli concentrations are 0 cfu/100 mL, a value of 1 cfu/100 mL was used to compute LRV.

1 LRV = 90% reduction

2 LRV = 99% reduction

3 LRV = 99.9% reduction

4 LRV = 99.99% reduction

Filter				Microbial M	easurements ^c			
		Raw Wate	r		Filtered Wat	er		Log
	H ₂ S Tests	E. Coli	Total Coliform	H ₂ S Tests	E. Coli	Total Coliform	% Removal	Reduction
	(P/A)	(cfu/100 mL)	(cfu/100 mL)	(P/A)	(cfu/100 mL)	(cfu/100 mL)	E. Coli ^d	Value ^e
BSF1	A ^a	2.5	10	A ^a	0.5	1.5	80	0.7
BSF2	A ^a	0	110	A ^a	>10	>90	(-1000)	-1
BSF3	A ^a	0 ^b	1.5 ^b	Aª	>400 ^b	>800 ^b	(-40000)	-2.6
BSF4	A ^a	0	20	A ^a	0	15	N/A	N/A
BSF5	Aª	>1000	>2000	Aª	10 ^b	>1010 ^b	99	2
BSF6	Aª	>110	>110	A ^a	0	>120	99	2
BSF7	Aª	0.5	29	Aª	>1000	>1032.5	(-199900)	-3.3
BSF8	A	0	5.3	Р	0	>1000	N/A	N/A
BSF9	Aª	1	101	A ^a	0	1	(100)	N/A
BSF10	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A

^aTime lag between sampling and incubation exceeded 2 hours

^bValue does not include result obtained after 2 day lag between sampling and incubation

^cWhere duplicate samples were obtained, results are presented as mean values

^dWhere microbial measurement results are presented as bounded values, the lower limit was used to calculate removal efficiency ^eLog Reduction Value (LRV) = log₁₀(raw water E. Coli concentration/filtered water E. Coli concentration)

Where filtered water E. Coli concentrations are 0 cfu/100 mL, a value of 1 cfu/100 mL was used to compute LRV.

1 LRV = 90% reduction

2 LRV = 99% reduction

3 LRV = 99.9% reduction

4 LRV = 99.99% reduction

Date	F low R ate (L/hr) ^c	Turt	oidity (± 0.	1 NTU) ^C		Microbial Measurements ^a						
					Raw	W ate T	21	Filtered	Filtered Water			Log
		Raw Water	Filtered Water	% Removal	H _Z S (ests (P/A)	(cf	E. Coli u/100 mL)	Hz5 Tests (P/A)	E. Coli (cfu/100 mL)		% Removal E. Coli ^b	Reduction Value ^E
1/11/2003				••								
1/12/03			*									
1/13/03												
1/14/03												
1/15/03	6	80.0	0.6	99.3	Р	>	1000	A				
1/16/03	6	60.0	8.2	86.3	А	>	500	Р		15	97	1.5
1/17/03	6	59.5	7.5	87.4	А	>	500	P/A ^d	>	100	80	0.7
1/18/03	5	29.0	4.3	85.2		>	655		>	132	80	0.7
average	6	57.1	5.2	91.0			664					

Appendix J. Summary of data from January 2003 Lumbini BioSand filter experiments – Green pitcher filter – riverbank sand

Where duplicate samples were obtained, results are presented as mean values

Where microbial measurement results are presented as bounded values, the lower limit was used to calculate removal efficiency

Where multiple tests were performed, results are presented as mean values

^dResults from multiple tests

"Log Reduction Value (LRV) = log a(raw water E. Coli concentration/litered water E. Coli concentration)

Where filtered water E. Coli concentrations are 0 cfu/100 mL, a value of 1 cfu/100 mL was used to compute LRV.

1 LRV= 90 % reduction

2 LRV= 99 % reduction

3 LRV= 99.9% reduction

4 LRV= 99.99% reduction

Date	Flow Rate (L/hr)°	Turbidity (± 0.1 NTU) ^c			Microbial M easurements ^a								
					Raw	Wate	er	Filtered	l Water		100		
		Raw	Filtered		H₂S Tests		E Coli	H ₂ S Tests	E Coli	% Removal E	Reduction		
		Water	Water	% Removal	(P/A)	(cfi	1/100 mL)	(P/A)	(cfu/100 mL)	Coli ^b	Value ^e		
1/11/03											1 4/40		
1/12/03													
4/4 2/0 2													
1113103													
1/14/03													
1/15/03	6	80.0	0.7	99.1	Р	≻	1000	А					
1/16/03	5	60.0	9.1	84.8	А	>	500	P/A [₫]	10	98	1.7		
1/17/03	5	59.5	4.2	92.9	А	>	500	А	> 90	82	0.7		
1/40/00		20.0	20	00.7			0.5.5			50	<u>.</u>		
1/18/03	4	29.0	3.0	89.7			000		> 269	59	U. 4		
average	5	57.1	4.3	92.6		<u> </u>	552						

^aWhere duplicates amples were obtained, results are presented as mean values

^bWhere microbial measurement results are presented as bounded values, the lower limit was used to calculate removal efficiency

Where multiple tests were performed, results are presented as mean values

^dResults from multiple tests

^ELog Reduction Value (LRV) = log₁₀(raw water E. Coli concentration/filtered water E. Coli concentration)

Where filtered water E. Coli concentrations are 0 cfu/100 mL, a value of 1 cfu/100 mL was used to compute LRV.

1 LRV = 90% reduction

2 LRV= 99% reduction

3 LRV= 99.9% reduction

4 LRV = 99.99% reduction

· · ·

Date	Volume of Water Introduced (L)	Flow Rate (Ưhr) ^c	Tur	oidity (± 0.1	NTU)°	E. Coli Concentrations (cfu/100 mL) ^a				
			Raw Water	Filtered Water	% Removal	Raw Water	Filtered Water	% Removal E. Coli ^b	Log Reduction Value [®]	
3/7/03	5.0	17	10.5	0.0	100	> 225 ^d				
3/8/03	7.0	20	8.0	0.0	100	≻ 258ª	65	75	0.6	
3/9/03	9.0	20	13.7	0.0	100	> 115 ^d	58	50	0.3	
3/10/03	8.0	18	3.0	0.0	100	55	24	56	0.4	
3/11/03	7.0	19	3.8	0.0	100	37	17	54	0.3	
3/12/03	10.0	17	2.0	0.0	100	52	17	67	0.5	
3/13/03	9.0	17	3.4	0.1	97	37	15	59	0.4	
3/14/03	8.0	15	3.5	0.0	100	123	75	39	0.2	
3/15/03	7.0	15	1.4	0.5	64	22	6	73	0.6	
3/16/03	7.0	15	4.0	1.0	75	117	35	70	0.5	
3/17/03	10.0	14	1.5	0.1	93	4 ^a	2	50	0.3	
3/18/03	11.0	17	2.8	0.7	75	ga	2	78	0.7	

.

Date	Volume of Water Introduced (L)	Flow Rate (L/hr) ^c	Tu	rbidity (± 0.1	NTU)°	E. (Coli Concentrati	ons (cfu/100 ml	_) ^a
			Raw Water	Filtered Water	% Removal	RawWater	Filtered Water	% Removal E. Col ^p	Log Reduction Value ^e
3/19/03	8.0	16	1.0	2.0	-100				
3/20/03	10.0	14	2.4	1.0	58	8	2	75	0.6
3/21/03	7.0	15	3.4	0.0	100	13	2	85	0.8
3/22/03	8.0	15	1.8	0.1	94	28	13	54	0.3
3/23/03	9.0	11	1.7	0.1	94	47	28	40	0.2
3/24/03	10.0	14	3.1	0.4	87	98	42	57	0.4
3/25/03	10.0	12	2.6	0.2	92	28	14	50	0.3
3/26/03	9.0	15	1.9	0.1	95	115	55	52	0.3
3/27/03	10.0	14	1.5	0.3	80				
3/28/03	10.0	14	2.3	0.2	91				
3/29/03									

Date	Volume of Water Introduced (L)	Flow Rate (Ưhr) ^c	Ти	rbidity (± 0.1	NTU)°	E. (E. Coli Concentrations (cfu/100 mL) ^a			
			Raw Water	Filtered Water	% Removal	RawWater	Filtered Water	% Removal E. Col∤	Log Reduction Value ^e	
3/30/03										
3/31/03	10.0	14	0.6	0.0	100					
4/1/03	8.0	15	7.3	1.0	86	345	160	54	0.3	
4/2/03	8.0	14	4.7	0.5	89					
4/3/03	8.0	14	10.9	1.2	89	1813	420	77	0.6	
4/4/03	8.0	13	14.3	0.9	94	1188	60	95	1.3	
average	8.6	15	4	0	100					

Where duplicate samples were obtained, results are presented as mean values

Where microbial measurement results are presented as bounded values, the lower limit was used to calculate removal efficiency

Where multiple tests were performed, results are presented as mean values

[#]Estimate based on highest concentration of E. Coli in filtered water

[•]Log Reduction Value (LRV) = log_{in}(raw water E. Coli concentration/filtered water E. Coli concentration)

Where filtered water E. Coli concentrations are 0 of u/100 mL, a value of 1 of u/100 mL was used to compute LRV.

1 LRV = 90% reduction 3 LRV = 99.9% reduction

2 LRV = 99% reduction 4 LRV = 99,99% reduction

In bold = 1:1 mixture of Deer Island wastewater: Charles River water introduced

•

Date	Volume of Water Introduced (L)	Flow Rate (L/hr)°	Turbidity (± 0.1 NTU)°				E. Coli Concentrations (cfu/100 mL) ^a				
			Raw Water	Filtered Water	% Removal	Raw Water		Filtered Water	% Removal E. Coli ^b	Log Reduction Value ^e	
3/7/03	5.0	11	10.5	0.8	92	>	225 ^d	225	0	0.0	
3/8/03	5.0	10	8.0	1.3	84	≽	258 ^d	258	0	0.0	
3/9/03	5.0	10	13.7	1.9	86	≻	115 ^d	108	6	0.0	
3/10/03	5.0	10	3.0	0.7	77		55	50	9	0.0	
3/11/03	5.0	11	3.8	0.4	89		37	34	8	0.0	
3/12/03	6.0	9	2.0	0.2	90		52	25	52	0.3	
3/13/03	5.0	10	3.4	1.9	44		37	22	41	0.2	
3/14/03	5.0	9	3.5	0.5	86		123	103	16	0.1	
3/15/03	5.0	8	1.4	0.1	93		22	15	32	0.2	
3/16/03	4.0	8	4.0	0.7	83		117	71	39	0.2	
3/17/03	5.0	8	1.5	0.0	100		4 ^d	4	0	0.0	
3/18/03	6.0	8	2.8	0.0	100		9ª	9	0	0.0	

Date	Volume of Water Introduced (L)	Flow Rate (L/hr) [¢]	Turb	idity (± 0.1	1 NTU)°	E. Coli Concentrations (cfu/100 mL) ^a					
<u></u>			Raw Water	Filtered Water	% Removal	Raw Water	Filtered Water	% Removal E. Coli ^b	Log Reduction Value ^e		
3/19/03	4.0	8	1.0	0.0	100						
3/20/03	4.5	9	2.4	0.0	100	8	4	50	0.3		
3/21/03	4.0	5	3.4	0.0	100	13	2	85	0.8		
3/22/03	4.3	8	1.8	0.4	78	28	23	18	0.1		
3/23/03	4.5	6	1.7	0.3	82	47	32	32	0.2		
3/24/03	4.5	8	3.1	0.5	84	98	55	44	0.3		
3/25/03	5.5	. 8	2.6	0.6	77	28	24	14	0.1		
3/26/03	4.3	8	1.9	0.3	84	115	77	33	0.2		
3/27/03	4.7	7	1.5	0.3	80						
3/28/03	3.4	8	2.3	0.2	91						
3/29/03											
3/30/03											

Date	Volume of Water Introduced (L)	Flow Rate (⊔'hr) ^c	Turk	oidity (± 0.1	1 NTU)°	E. C	Coli Concentrati	ons (cfu/100 m	L) ^a
_			Raw Water	Filtered Water	% Removal	Raw Water	Filtered Water	% Removal E. Coli ^b	Log Reduction Value [®]
3/31/03	5.0	6	0.6	0.0	100				
4/1/03	3.5	7	7.3	2.1	71	345	280	19	0.1
4/2/03	4.5	7	4.7	1.8	62				
4/3/03	3.0	7	10.9	1.8	83	1813	750	59	0.4
4/4/03	3.0	6	14.3	1.3	91	1188	40	97	1.5
average	4.6	8.1	4.3	0.7	83.7		1		

^aWhere duplicate samples were obtained, results are presented as mean values

^bWhere microbial measurement results are presented as bounded values, the lower limit was used to calculate removal efficiency

Swhere multiple tests were performed, results are presented as mean values

^dEstimate based on highest concentration of E. Coli in filtered water

^ELog Reduction Value (LRV) = log₁₀(raw water E. Coli concentration/filtered water E. Coli concentration)

Where filtered water E. Coli concentrations are 0 cfu/100 mL, a value of 1 cfu/100 mL was used to compute LRV.

1 LRV = 90% reduction 3 LRV = 99.9% reduction

2 LRV = 99% reduction 4 LRV = 99.99% reduction

In bold = 1:1 mixture of Dieer Island wastewater:Charles River water introduced
Date	Volume of Water Introduced (L)	Flow Rate (L/hr)°	Turbidity (± 0.1 NTU)°			E. Coli Concentrations (cfu/100 mL) ^a				
			Raw Water	Filtered Water	% Removal	Raw Water		Filtered Water	% Removal E. Coli ^b	Log Reduction Value ^e
3/7/03	5.0	5	10.5	1.7	84	×	225 [₫]	203	10	0.0
3/8/03	5.0	7	8.0	1.6	80	>	258 ^d ·	192	26	0.1
3/9/03	5.0	6	13.7	1.2	91	>	115 ^d	115	0	0.0
3/10/03	4.7	6	3.0	1.2	60		55	50	9	0.0
3/11/03	5.0	5	3.8	0.9	76		37	26	30	0.2
3/12/03	6.0	5	2.0	0.6	70		52	36	31	0.2
3/13/03	5.0	5	3.4	0.5	85		37	25	32	0.2
3/14/03	4.5	5	3.5	0.6	83		123	110	11	0.0
3/15/03	4.0	4	1.4	0.1	93		22	15	32	0.2
3/16/03	4.0	5	4.0	0.8	80		117	67	43	0.2
3/17/03	5.0	5	1.5	0.2	87		4 ^d	3	25	0.1
3/18/03	5.0	4	2.8	0.0	100		9ª	6	33	0.2



.

Date	Volume of Water Introduced (L)	Flow Rate (L/hr) ^c	Turbidity (± 0.1 NTU)°			E. Coli Concentrations (cfu/100 mL) ^a			
			Raw Water	Filtered Water	% Removal	Raw Water	Filtered Water	% Removal E. Colf	Log Reduction Value ^e
3/19/03	5.0	4	1.0	0.0	100				
3/20/03	4.5	3	2.4	0.1	96	8	4	50	0.3
3/21/03	3.3	4	3.4	0.2	94	13	5	62	0.4
3/22/03	5.0	5	1.8	0.3	83	28	20	29	0.1
3/23/03	4.5	3	1.7	0.2	88	47	23	51	0.3
3/24/03	4.8	3	3.1	0.5	84	98	40	59	0.4
3/25/03	5.0	4	2.6	0.5	81	28	14	50	0.3
3/26/03	4.3	4	1.9	0.5	74	115	72	37	0.2
3/27/03	4.7	3	1.5	0.1	93				
3/28/03	3.4	3	2.3	0.0	100				
3/29/03									
3/30/03									

Date	Volume of Water Introduced (L)	Flow Rate (L/hr) ^c	Turbidity (± 0.1 NTU) ^o			E. Coli Concentrations (cfu/100 mL) ^a				
			Raw Water	Filtered Water	% Removal	Raw Water	Filtered Water	% Removal E. Coli ^b	Log Reduction Value [®]	
3/31/03	5.0	3	0.6	0.0	100					
4/1/03	3.5	4	7.3	1.5	79	345	250	28	0.1	
4/2/03	4.5	4	4.7	0.9	81					
4/3/03	3.0	4	10.9	1.4	87	1813	550	70	0.5	
4/4/03	3.0	3	14.3	1.0	93	1188	40	97	1.5	
average	4.5	4.3	4.3	0.6	86.0				<u></u>	

Where duplicate samples were obtained, results are presented as mean values

^bWhere microbial measurement results are presented as bounded values, the lower limit was used to calculate removal efficiency

 ${}^{\rm S}$ where multiple tests were performed, results are presented as mean values

^dEstimate based on highest concentration of E. Coli in filtered water

^ELog Reduction Value (LRV) = log m(raw water E. Coli concentration/filtered water E. Coli concentration)

Where filtered water E. Coli concentrations are 0 cfu/100 mL, a value of 1 cfu/100 mL was used to compute LRV.

1 LRV = 90% reduction 3 LRV = 99.9% reduction

2 LRV = 99% reduction 4 LRV = 99.99% reduction

In bold = 1:1 mixture of Deer Island wastewater:Charles River water introduced

Appendix O. Quality assurance blanks obtained during January 2003 in Lumbini district

Date	Millipore Membrane Filtration ^b								
	Time Sampled	Time Incubated	Time Lag	E. Coli (cfu/100 mL)	Total Coliform (cfu/100 mL)				
1/6/2003	19:33	19:36	0:03	0	0				
1/7/2003	7:19	8:00	0:41	0	0				
1/7/2003	10:20	11:19	0:59	0	0				
1/7/2003 (1 mL)	15:11	15:17	0:06	0	0				
1/8/2003	15:02	15:09	0:07	0	0				
1/8/2003	15:30	15:38	0:08	0	3				
1/9/2003	17:15	17:22	0:07	0	0				
1/12/2003	8:20	8:30	0:10	0	0				
1/13/2003	12:45	12:50	0:05	0	10				
1/14/2003 ^c	16:30	16:35	0:05	0	0				
1/15/2003	13:20	13:25	0:05	0	0				
1/16/2003	16:45	17:25	0:40	0	1				
1/17/2003	17:35	17:45	0:10	0	0				

^aSample volumes are 20 mL.

^bUnless otherwise specified, sample volumes are 100 mL.

^cTest performed by Xanat Flores

Date	HACH H ₂ S Tests ^a								
	Time Sampled	Time Incubated	Time Lag	Test Result 24 hrs	Test Result 48 hrs				
1/6/2003	20:30	1/7/2003 8:53	12:23	Α	A				
1/6/2003	20:30	1/7/2003 8:53	12:23	Α	А				
1/7/2003	8:30	8:53	0:23	Α	А				
1/8/2003	14:32	14:45	0:13	Α	Α				
1/9/2003	16:45	17:00	0:15	Α	А				
1/12/2003	8:15	8:15	0:00	Α	А				
1/14/2003	15:30	16:20	0:50	А	А				
1/16/2003	17:15	17:17	0:02	А	A				

^aSample volumes are 20 mL.

^bSample volumes are 100 mL.

Appendix P. Quality assurance blanks obtained during laboratory experiments at MIT

		Millipore Membrane Filtration ^a
Date	E. Coli (cfu/100 mL)	Total Coliform (cfu/100 mL)
3/7/2003	0	0
3/8/2003	0	0
3/9/2003	0	0
3/10/2003	0	0
3/11/2003	0	0
3/12/2003	0	0
3/13/2003	0	0
3/14/2003	0	0
3/15/2003	0	0
3/16/2003	0	1
3/17/2003	0	0
3/18/2003	0	0
3/19/2003	0	0
3/20/2003	0	0
3/21/2003	0	0
3/22/2003	0	3
3/23/2003	0	5
3/24/2003	0	0
3/25/2003	0	0
3/26/2003	0	0
4/1/2003	0	6
4/3/2003	0	0
4/4/2003	0	0

^aSample volumes are 100 mL.

Appendix Q. Morbidity Statistics for Lumbini District Villages (August 2001 – August 2002)

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(obtained from	International	Buddhist	Society	health	clinic.	BuddhaNaga	ar. Nepal)
N C C C C C C C C C C)		·

	Leucorrhea	Amoebiasis	Diahhrea	Gastritis
Sonbarsha	102	130	85	120
Sonbarshi	70	55	65	84
Ramwapur	110	80	75	62
Bichauwapur	67	45	48	82
Sujandihawa	188	165	103	131
Bhagawanpur	156	140	133	110
Dhodawa	175	95	58	53
Ramwapur	112	94	62	33
Shivgadiya	132	75	46	47
Mehilwari	75	54	36	19
Sekhuadad	16	28	20	12
Lankapur	18	22	32	15
Mujhana	183	146	92	77
Laxmipur	110	72	41	47
Manauri	156	195	53	91
Mahuwari	108	82	41	23
Bhujaihiya	141	120	70	60
Lamtihawa	137	78	43	27
Ekla	116	127	78	81
Khambe	97	76	28	38
Bhagatpurwa	42	59	30	23
Tenuhawa	130	115	49	32
Mahilwar	89	77	67	47

Appendix Q continued Illness Symptoms

(from conversation between Susan Murcott and Rajess Yadav, Community Health Assistant at IBS Clinic, January 2003)

Illness	Symptoms	Medicines Prescribed
Leucorrhea	backache	Tus. Metronidazole – 7 days
(from virus	white discharge	200-404 Ital Tidis
Tricomonussis)	general pain and body ache	Tus B. Complex
	tingling sensation on hands and feet	
	loss of appetite	
	no fever	
Amoebiasis	body ache	Tus. Metronidazole
	abdominal pain near umbical region	Tus. Antispasmodic
	mucus in stool	Antacid syrup
	constipation	
	nausea but no vomiting	
	small fever	
	2-3 times toilet	
	indigestion	
Diahhrea	watery discharge	Icy Riyer Lented
(from bacteria,	feel thirsty	Jeevanja Pouod (ORS)
food poisoning)	skin cramps	Tus. Metronidazole
	sunken eyes	Tus. Novfloxain
	whole body pain	
	fever	
	nausea/vomiting	
	bloody stool	
Gastritis	nausea	Tus. Ranitidine
(from smoking,	gastric pain	Antacid syrup
chewing tobacco,	indigestion	
spicy food, drug	constipation	
addiction)	loss of appetite	
	heart burn	

Appendix R. Cost break-down of concrete BioSand filter

Sn	Description	Unit	Quantity	Rate	Amount	Remark
1	Cement	Bag	1	300.00	300.00	
2	Sand	Bag	2	150.00	300.00	
3	Aggregate	Bag	2	150.00	300.00	
4	Pipes	Μ	1	150.00	150.00	
5	Mould	Ls	1	200.00	200.00	
	transport					
6	Bucket	No	2	200.00	400.00	
7	Diffuser	Pc	1	250.00	250.00	
8	Cover	Pc	1	200.00	200.00	
	plate					
9	Fitting	No	2	200.00	400.00	
10	Laborers	No	2	100.00	200.00	
11	Oil	L	1/2	200.00	100.00	
12	Repairs	L		100.00	100.00	
13	Sand	NO	2	100.00	200.00	
	Washing					
14	External	LS	1	80.00	80.00	
	Paint					
15	Misc	LS	1	200.00	200.00	
				Total	3380	
	Exchange	lusd	=74 Rs	In	45.00	
	rate			USD		

(from Durga Ale, Lumbini, Nepal)