THE MICROBIOLOGICAL AND HEALTH IMPACT OF THE BIOSAND FILTER IN THE DOMINICAN REPUBLIC: A RANDOMIZED CONTROLLED TRIAL IN BONAO

Christine E. Stauber

A dissertation submitted to the faculty of the University of North Carolina at Chapel Hill in
partial fulfillment of the requirements for the degree of Doctor of Philosophy in the
Department of Environmental Sciences and Engineering

Chapel Hill
2007

Approved by:

Mark D. Sobsey, PhD

Dana P. Loomis, PhD

Fran A. DiGiano, PhD

Louise Ball, PhD

Rob Quick (CDC), MD

Dale Whittington, PhD

ABSTRACT

CHRISTINE E. STAUBER: The Microbiological and Health Impact of the Biosand Filter in the Dominican Republic: A Randomized Controlled Trial in Bonao
(Under the direction of Mark D. Sobsey)

More than one billion people lack access to improved water supplies and even more lack access to safe water. Many household water treatment technologies have been documented to improve drinking water quality and reduce diarrheal disease. However, other technologies that are being used still lack rigorous evidence on ability to improve water quality and reduce diarrheal disease. One of these technologies is the biosand filter (BSF), an intermittently operated slow sand filter. It is estimated that more than 80,000 BSFs are in use world wide yet there is no rigorous evidence of their ability to reduce diarrheal disease and there is only limited evidence of their ability to improve drinking water. The purpose of this research was to examine the microbiological and health impact of the BSF in the laboratory and in the field. The laboratory research examined the ability of the BSF to reduce viruses and bacteria from water. The field research examined improvements in drinking water quality by the BSF in use in households and the ability of the BSF to reduce diarrheal disease. Based on the laboratory evidence, the BSF can achieve moderate to high reductions of bacteria 90-99% and moderate reductions of viruses (90%). The field study suggested moderate reductions of *E. coli* by the BSF in the field which was 80% on average yet it ranged 0-99.9%. The health impact portion of the field study found a 47% reduction in diarrheal disease in BSF users as compared to non-users. In addition, the health impact study found a weak association between increased contamination in drinking water as measured by *E. coli* and rates of diarrheal disease. The results from this research suggest that the biosand filter may be an effective way to improve drinking water quality and reduce diarrheal disease in the communities studied in Bonao, Dominican Republic.

ACKNOWLEDGEMENTS

First, I would like to thank all of people from the communities of Jayaco and Brisas del Yuna because without their help and patience, none of the field research would have been possible. I also received tremendous support from all of the staff and others that supported the work in the Dominican Republic including our lab manager Douglas Wait. I would also like to thank Dr. Gloria Ortiz and her family for all of their support and assistance while in the country. Gloria's expertise and the help of her family was a significant source of assistance in making the project a reality.

I would like to acknowledge the support of my doctoral dissertation committee: Mark Sobsey, Dana Loomis, Fran DiGiano, Dale Whittington, Doug Crawford-Brown and Rob Quick and Louise Ball's last minute assistance. In addition, I have had a great experience working in the Sobsey lab and have to thank all the members for their help and assistance; especially Mark Elliott. Without Mark, I would not have been able to lift 80lbs. of water to run our lab experiments. I also have to give a mention to the members of team latrine who made playing with fecal matter fun.

Finally, I have to say thank you to my friends and family. I know it seems like this has been a never-ending task but your constant support and understanding has made it easier. Mom and Grandpa, I think of you often. Granny, Dad, Chris and Elaine, Mike and Cindy and Angie, Julie, Nicole and the others, I made it!

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ABBREVIATIONS

BSF – Biosand filter

CI – Confidence interval

DR – Dominican Republic

GBD – Global burden of disease

INAPA – Instituto Nacional de Agua Potable y Aqueductos

IRD – Incidence rate difference

IRR – Incidence rate ratio

LRVEC – Log reduction value for *E. coli*

MDG – Millennium Development Goals

MIT – Massachusetts Institute of Technology

NGO – Non-governmental organization

OR – Odds ratio

PAHO – Pan-American Health Organization

PCA - Principle Components Analysis

POU – Point of use

RCT – Randomized controlled trial

SAL – Single agar layer

SES – Socioeconomic status

SSF – Slow sand filtration

UNICEF – United Nations International Children’s Fund

WHO – World Health Organization

Chapter 1: Overview and Objectives

1.1 Introduction and Background

It is estimated that 4 % of all deaths are a result of the disease burden from inadequate water, sanitation and hygiene and that this accounts for more than 5% of the total disease burden worldwide (Pruss, Kay, Fewtrell, & Bartram, 2002). A recent study suggests that while mortality from diarrheal diseases has dramatically decreased, morbidity has not (Kosek, Bern, & Guerrant, 2003). Furthermore, evidence suggests that 94% of diarrheal disease is attributable to environmental risk factors primarily related to water, sanitation and hygiene (Pruss-Ustun & Corvalan, 2006).

In an attempt to decrease this global diarrheal disease burden, many studies have assessed the effectiveness of interventions in water, sanitation and hygiene. A series of papers in the 1980's and 1990's attempted to examine the large and growing body of evidence on the health impact of these interventions (Esrey, 1996; Esrey, Feachem, & Hughes, 1985; Esrey, Habicht, & Casella, 1992; Esrey, Potash, Roberts, & Shiff, 1991). To determine the most effective interventions, Esrey et al., (1985) reviewed 67 studies and calculated median reductions in diarrheal morbidity for each category of these interventions in water, sanitation and/or hygiene. The results suggested that the median reduction in diarrheal morbidity rates for all types of interventions was 22% and that improvements in water quality contributed the smallest benefit (16%). Later analysis by Esrey and others generally confirmed these findings, as summarized in the analysis of 144 studies (Esrey et al., 1991). In this study, the authors noted that improved water quality was less important than sanitation for reducing diarrheal disease.

While Esrey and others made a major contribution in their review of the published literature of the time, their results suggested that improvements in water quality were not as effective as other interventions at reducing diarrheal disease. Only recently, researchers have reexamined the contributions of different water, sanitation and hygiene measures for their impact on diarrheal disease. Current and more comprehensive and rigorous evidence on interventions to improve water quality suggests that the effectiveness of water quality interventions was underestimated by Esrey and others (L. Fewtrell & Colford, 2005). In a recent meta-analysis on interventions in water, sanitation and hygiene, the authors suggests that improvement in water quality can reduce diarrheal disease morbidity by more than 30%, which is nearly double the original estimate by Esrey et al., (1985).

A comparison of the findings of the two studies is given in Table 1.1 It is interesting to note that, the authors of the 2005 review and analysis of intervention studies suggests that multiple improvements of water, sanitation and hygiene concurrently did not result in increased benefits over improvements achieved for one particular intervention. However, their research and other studies show that there is a renewed interest in the impacts of interventions in water quality on reductions in diarrheal disease burdens. A major difference in the data sets collected in the two studies is that the former included no studies on water quality produced by point-of-use water treatment and its impact on diarrheal disease risks, while the latter study did. The role of household POU treatment on water quality and its impact on diarrheal disease risks is only now being carefully and systematically examined in the developed and developing world.

Table 1.1: Comparison of two studies and the estimated reductions in diarrheal disease of interventions in water and sanitation interventions

Type of interventions Esrey et al. 1985	Median % reduction	Type of interventions Fewtrell et al. 2005	Pooled % Reduction
All types	22%	Multiple	33%
Water availability	25%	Hygiene	37%
Water quality	16%	Water quality	31%
Water quality and availability	37%	Water supply	25%
Excreta disposal	22%	Sanitation	32%

POU Drinking Water Treatment in the Developing World

Nearly 1.1 billion people lack access to improved water supplies worldwide (*The UN Millennium Development Goals (website)*) and even more lack access to microbiologically safe water. In addition, for the population that has to collect and store water, there is risk of contamination and deterioration of water quality. It has been well documented that during collection and storage of household water, the initially safe water often becomes fecally contaminated due to unsanitary storage and handling practices (M.D. Sobsey, 2002; Wright, Gundry, & Conroy, 2004). The United Nations' Millennium Development Goals include the goal to halve the number of the world's people without access to improved water by 2015. A definition of improved and not improved water supplies is provided in Table 1.2. It will take considerable time and money to improve people's access to piped community water supplies, and many people will not be served by such infrastructure by 2015. Practical and innovative in-home treatment technologies can provide an interim solution now.

Table 1.2: Examples of improved /not improved water supplies (adapted from UNICEF)

Improved	Not improved
Household connection Public standpipe Borehole Protected dug well Protected spring Rainwater collection	Unprotected well Unprotected spring Vendor provided water Tanker truck water

The goal of point-of-use (POU) water treatment technology is to empower people who only have access to unsafe water sources to improve the quality of their water by treating it in the home. The concept of expanding POU treatment to people who have access only to poor quality sources of drinking water by treating as much water as they use, in their own home, is a relatively recent development. In 2003 this approach was endorsed by the World Health Organization by incorporating it into the 3rd Edition of the WHO Guidelines fro Drinking-water Quality (WHO, 2003) and facilitating the creation in 2003 of an International Network to Promote Household Water Treatment and Safe Storage (http://www.who.int/household_water/en/). Furthermore, this approach to improving access to safe water has been embraced by the Joint Monitoring Program of the UN and its partners as an approach to achieving the water access target of the Millennium Development Goals WHO-UNICEF, 2006 (*Meeting the MDG Drinking Water and Sanitation Target.*, 2006).

There are a number of different POU technologies, which makes it possible to select among them and adapt them to specific places and populations. These technologies include (adapted from Sobsey 2002):

- Boiling
- Solar disinfection by the combined action of heat and UV radiation
- Solar disinfection by heat alone ("solar cooking")
- UV disinfection with lamps
- Chlorination plus storage in an appropriate vessel
- Combined systems of chemical coagulation-filtration and chlorine disinfection
- Ceramic filtration
- Intermittently operated slow sand filtration or biosand filtration
- Halogenated resin bed (e.g., Lifestraw)
- Inorganic ion disinfectant (e.g., One Drop, a mixture of aluminum, copper, gold, silver, and zinc ions)

Preferred POU technologies are household scale, easy to use, low maintenance, low cost, produce sufficient quantities of water, are effective in removing or inactivating pathogenic microorganisms in water and are proven to reduce diarrheal disease rates in users. These technologies also need to be robust and must fill the need to provide safe drinking water until people have access to safe, piped water, which may be years to decades away for some communities. POU technologies differ in terms of cost, effectiveness against different types of microorganisms, ease of use, maintenance requirements, and quantity of water that can be treated in a given amount of time. All of these factors affect the long-term sustainability and overall effectiveness of these technologies in reducing the global burden of waterborne disease.

Of the technologies listed above, many of them have been documented for their ability to reduce pathogenic microbes in water in laboratory and field studies, and for their ability to reduce diarrheal disease in users (T. F. Clasen, Brown, Collin, Suntura, & Cairncross, 2004; Reller et al., 2003; M. D. Sobsey, Handzel, & Venczel, 2003). In his review of available household water treatment technologies Sobsey (2002) cited improvements in microbial water quality and diarrheal disease reductions for boiling, solar disinfection, and chlorine disinfection, with reductions in disease burden ranging from 9-48% depending on the intervention. In more recent research, Reller et al., 2003, reported a 29% decrease in diarrheal disease rates for household treatment with a combined system consisting of coagulation-filtration and disinfection. Clasen et al., 2004 demonstrated a mean reduction in diarrheal prevalence of 64% when examining the effectiveness of household ceramic filtration in Bolivia. Indeed, a recent meta-analysis by Fewtrell et al., 2005 suggests that household water treatment interventions and improvements in water quality were found to be more effective than previously thought. The researchers documented >30% overall reduction in diarrheal disease risk when examining household POU treatment studies.

The biosand filter is one of the technologies on the list of candidates that has not been well documented for its ability to improve water quality and reduce diarrheal disease. Only limited evidence exists of microbiological effectiveness of the biosand from lab and field studies. Despite the paucity of performance data on microbiological effectiveness and the absence of data on health impact, such as diarrheal disease reduction, the biosand filter is being enthusiastically promoted on many continents. To enhance the current information on the BSF, this project will focus on determining the ability of the biosand filter to treat water to reduce microbial contaminants and to reduce household diarrheal disease.

Traditional Slow Sand Filtration and the Biosand Filter

Slow sand filtration has been employed for more than 100 years to treat small and large community water supplies. Removals of viruses, bacteria and parasites can range from 99-99.99% under mature (ripened) sand-bed conditions (*Slow Sand Filtration*, 1991). While conventional slow sand filtration removes pathogens from water, the effectiveness of the small household scale unit is uncertain because of different operating properties. The biosand filter typically consists of a cement chamber with a spigot as a treated water outlet. The chamber is filled with a column of sand and maintains a layer of water above the sand surface in order to create and maintain the needed biologically active slime layer or "schmutzdecke". Water is poured into the space above the sand, passes through the column, and is collected at the spigot that dispenses water. This spigot is placed at a height that maintains water at a level of several centimeters above the top of the sand bed in order to avoid dewatering of the sand bed.

Limited laboratory evidence exists for reduction of fecal indicator bacteria and parasites by the BSF. A master's thesis documented a 55-day dosing study where fecal coliform removal was an average 96% when the filter was dosed with contaminated pond water (Buzunis, 1995). Researchers at Massachusetts's Institute of Technology (MIT) have performed two laboratory studies on the BSF. In a ripened filter average reduction of fecal indicator bacteria was 99.5% (Lee, 2001). However, in another study the reduction of fecal indicator bacteria was measured over 4 weeks of use and ranged from 52- 97% (Donison, 2004). Reported removals of bacteria seem to be variable and typically lower than expected for slow sand filtration. However, removals of protozoan parasites are high. Palmateer et al. (1999) documented >99% removal of *Giardia* cysts and *Cryptosporidium* oocysts. As these protozoans are at least several-fold larger in size than bacteria, they may be more effectively removed by filtration processes employing beds of sand.

Published field studies suggest that BSFs in Haiti in use for more than one year had 98.5% reductions of *E. coli* (Duke, Nordin, Baker, & Mazumder, 2006). No other field studies are published in the peer-reviewed journal literature on either microbiological performance or ability of the biosand filter to improve health. However, a large body of unpublished literature on the performance of biosand filters from implementing organizations is available. Samaritan's Purse, an international faith-based non-governmental organization (NGO) that has installed more than 75,000 biosand filters worldwide, performed the largest observational study on biosand filters in field use. In this study, they sampled 577 filters on six continents and found an average of 93% (range 81 – 100%) reduction of fecal coliform bacteria (Kaiser, Liang, Maertens, & Snider, 2002).

BSFs are already being used by people in the Caribbean, North and South America, Asia and Africa to treat water in their homes even though detailed laboratory studies of their effectiveness in removing pathogens from water have not been done and there are no rigorous field studies of the effectiveness for reducing waterborne illness in users. Because this POU technology has been inadequately evaluated in the lab or the field for its performance in reducing all classes of pathogens (viruses, bacteria and parasites) and in reducing waterborne disease, a major focus of the project will be to study its microbiological effectiveness in the lab and the field and its health impact benefits in the field Such research is intended to fill crucial gaps in knowledge about its effectiveness in field use.

Importance of the BSF in the Dominican Republic and Opportunities for Field Studies

The Dominican Republic (DR) is an ideal location to evaluate the biosand filter in the field. While the Pan American Health Organization (PAHO) reports decreases in infant mortality in the DR in the last decade, this trend seems to have slowed (PAHO Country Health Profile, 2001). As Kosek et al., (2003) note, the decrease in worldwide mortality due to diarrhea has not been accompanied by a resulting decrease in morbidity, suggesting that diarrheal disease remains a major burden, especially in children. In the DR, communicable diseases, particularly intestinal infectious diseases, are reported to be the cause of >15% of deaths in children ages 1 to 4 in 1994. In 1994, diarrheal diseases represented 4% of all diagnosed deaths and 30.4 % of deaths from communicable diseases (PAHO Country Profile, 2001). In a 2002 survey, diarrheal disease prevalence was cited to be around 20% or 1 in 5 for children under five years of age (*Encuesta Demografica y de Salud: Republica Dominicana*, 2003).

Determining the microbiological effectiveness and health impact of the BSF is a critical need in the Dominican Republic because the biosand filter is already being used by thousands of people in the country. The first filters were first made in the DR in 2000. Since then, almost 8000 filters have been installed. In 2004 alone, over 1,300 filters were installed in various regions of the country with the help of Peace Corps volunteers and local and international Rotary Clubs. Per capita, the DR has one of the highest concentrations of biosand filters in the world, second perhaps only to Cambodia. However, there is no sound scientific evidence to document its effectiveness in reducing the burden of disease in the field, including no such evidence exists from the Dominican Republic.

1.2 Objectives

1. **Determine the ability of the biosand filter to reduce *Escherichia coli* and two bacteriophages, MS2 and PRD1, in seeded water when challenged under controlled laboratory conditions.**
2. **Determine the ability of the biosand filter to reduce concentrations of total coliforms and *E. coli* in water in the field in the Dominican Republic.**
3. **Determine the ability of the biosand filter to reduce household diarrheal disease incidence rates in households using the BSF as compared to control households by >15% in users (BSF households) as compared to non-users (households without BSF) in a randomized controlled trial in the Dominican Republic.**

Chapter 2: Literature Review

2.1 Diarrheal Disease and Waterborne Illness

Diarrheal disease is a major cause of morbidity and mortality in young children. Recent estimates suggest that diarrhea accounts for more than 1.6 million deaths annually (WHO, 2006). Mortality from diarrheal disease has decreased over the past four decades yet a recent study on the global burden of the disease suggests that there has not be an accompanying decrease in morbidity (Kosek et al., 2003). The average child in the developing world experiences 3 or more diarrheal disease episodes per year; accounting for more than 4 billion cases of diarrhea annually.

Diarrheal disease is often caused by pathogens that are transmitted through the fecal-oral route. Pathogens transmitted by this route are typically considered enteric pathogens because they can infect the gastrointestinal tract. Once these pathogens are shed into the environment via excreta they are capable of being transmitted in a variety of ways including through contact with contaminated water and person-to-person. Disease transmission by water can classified into four categories: waterborne, water-washed, water-based and water-related (White, Bradley, & White, 2002). Waterborne pathogens are transmitted by ingestion of fecally contaminated water. Water-washed pathogens are transmitted due to a lack of adequate quantity of water for washing and bathing. Water-based pathogens spend parts of their lives in water as essential components of their life cycle. Water-related pathogens are transmitted via an insect vector that breeds in water.

Interventions in drinking water quality to reduce diarrheal disease target primarily waterborne pathogens. Waterborne pathogens comprise a broad range of microorganisms ranging from viruses to bacteria to parasites. A list of pathogens that are common causes of waterborne disease is given in Table 2.1. While in the United States (US) the most common causes of diarrheal disease are viruses (not specifically waterborne), all three pose a significant threat to health and risk of diarrheal disease in less-developed countries (Dennehy, 2005). For example, a recent case-control study in Ecuador documented cases of diarrhea as a result of all three classes of pathogens: *E. coli*, Rotavirus and *Giardia* (Eisenberg et al., 2006).

Table 2.1: Infectious causes of diarrheal disease

Viral	Bacterial	Parasitic
Enteric Adenoviruses	*Escherichia coli*	*Entamoeba hystolitica*
Enteroviruses	*Salmonella* spp.	*Giardia intestinalis*
Rotavirus	*Shigella* spp.	*Cryptosporidium (spp.)*
Calicivirus	*Vibrio cholera*	
Astrovirus	*Campylobacter* spp.	
Hepatitis A/E	*Yersinia enterocolytica*	
	Clostridium difficile	

Source: (Dennehy, 2005; Maier, Pepper, & Gerba, 2000)

Worldwide institution of municipal drinking water and waste water treatment systems in the 20th century resulted in a dramatic decrease in transmission of waterborne pathogens. In the United States, in addition to decreasing the transmission of waterborne pathogens, the implementation of municipal water delivery systems has also affected the etiology of waterborne disease. In the early 20th century, major causes of waterborne disease in the US were cholera and typhoid (bacterial pathogens). In the period 1920-1941, more than 80,000 cases of typhoid were reported; however since 1971 only 282 cases of typhoid have been reported (Craun, Craun, Calderon, & Beach, 2006).

Recent outbreaks of waterborne disease in the US have been attributed to deficiencies in the water distribution system. During 2003-2004, a total of 36 waterborne disease outbreaks were identified. Thirty were associated with drinking water; causing 2760 illnesses and four deaths. The etiologic agents were bacterial, parasitic, viral, and chemical toxin related. Approximately 50% of the deficiencies that caused the illnesses occurred outside of the jurisdiction of the water utility; in distribution systems and in contaminated ground waters (Liang et al., 2006). Other outbreaks in the US are the result of failure of the municipal water system or the use of untreated water supplies such as groundwater.

Municipal treatment systems for drinking water treatment have varying capabilities in dealing with the three main classes of microorganisms. Typical drinking water treatment systems are quite capable of reducing bacteria by 99.999 – 99.9999% yet are less effective against viruses and parasites. Payment and others evaluated bacteria and virus removal in the various stages of water treatment plants in Canada and found all treated water free of indicator bacteria but 7% of the sampled waters tested positive for enteric viruses (Payment, Trudel, & Plante, 1985). Protozoan parasites are also more poorly removed through traditional water treatment processes. *Cryptosporidium* oocysts are highly resistant to chlorine disinfection requiring contact-times of 1000 fold or greater to achieve the same reduction for bacteria (Maier et al., 2000).

The microbial reduction efficiencies of various water treatment processes are listed for the three classes of microorganisms in Table 2.2. As shown in Table 2.2, bacteria are removed or reduced as well as viruses or parasites for most all treatment processes listed.

Also important to note in this table, some processes are less effective against protozoan parasites such as chemical disinfection with free chlorine. Other processes are not as effective at removing viruses such as ultraviolet irradiation. However, with few exceptions, bacteria are reduced by these processes as well or more efficiently as both viruses and parasites with one exception; physical removal via filtration. Filtration is more effective against larger microorganisms such as protozoan cysts or helminth ova. Municipal treatment systems typically employ multi-barrier approaches to address the wide range of waterborne pathogens.

Table 2.2 Reductions of microorganisms by water treatment processes

Treatment process	Viruses	Bacteria	Parasites/helminth ova
Storage (highly dependent on time)	Can reach 90%	Less than 90%	90-99%
Sedimentation (typically low and variable reductions)	Low unless attached to particles: < 90%	Low unless attached to particles: < 90%	Can exceed 90% especially with increased time for settling
Coagulation and Flocculation (dependent upon process parameters)	90-99%	90-99%	90-99%
Slow Sand Filtration (performance highly dependent on sand bed matures)	90-99.9%	90-99.99%	Physical removal can be expected as well as inactivation or reduction 99-99.99%
Rapid Granular Media Filters (typically low reductions; enhanced by pre-treatment with coagulation/flocculation)	<50%	50-90%	50-90% - can exceed 90% but highly variable
Membrane filter	>99.99%	>99.99%	>99.99%
Disinfection by free chlorine (measured in concentration * time) Ct values for 99% inactivation	Polio 1 Ct = 1.7	*E. coli* Ct = 0.04 – 0.6	*Giardia lamblia* cysts Ct = 54 – 192 *Cryptosporidium* oocysts Ct = > 7200
Disinfection by ozone (measured in concentration * time) Ct values for 99% inactivation	Polio 1 Ct =0.2	*E. coli* Ct = 0.006 – 0.02	*Giardia lamblia* cysts Ct = 0.53 *Cryptosporidium* oocysts Ct =3.5
Ultraviolet irradiation (measured in µW – s/cm^2) Dose values are for 90% inactivation	5,000-12,000	1,300 – 3,000	

Sources: (Maier et al., 2000; M.D. Sobsey, 2002)

2.2 The Millennium Development Goals

In developing countries, municipal water systems do not reach all of the populations that need access to them. Billions of people lack access to drinking water that has received any form of improvement. In September 2000, the United Nations (UN) put out the Millennium Declaration. As a demonstration of dedication to human dignity and equality, the UN set a series of development goals intended to decrease global poverty and improve the lives of the billions of people by 2015. The intent is to improve lives of those who still live on less than one dollar a day, and lack access to things that many people take for granted such as improved water and sanitation services. While each Millennium Development Goal (MDG) is targeted to a specific disease burden or facet of poverty such as access empowering women and girls through literacy; the environment and access to water and sanitation play a prominent role in development and in reaching many of the goals stated in the declaration. Specifically, Goal 7 Target 10 is to halve the proportion of people without sustainable access to improved water by 2015 (*The UN Millennium Development Goals (website)*).

At the time of the MDG declaration, more than one billion people lacked access to improved water supplies and double that number lacked access to improved sanitation. Lack of access to water and sanitation services can contribute significantly to the global burden of disease (GBD) and mortality due to diseases such as diarrhea and malnutrition. The World Health Organization (WHO) estimates that diarrheal diseases kill 1.6 million people every year, mostly children under five years of age. Furthermore, while mortality from diarrheal diseases may be decreasing, morbidity has not; with the average child in a developing country under five experiencing 3 or more diarrheal disease episodes per year (Kosek et al., 2003).

Researchers for WHO now estimate that diarrheal diseases account for 4% of the global burden of diseases (Pruss-Ustun & Corvalan, 2006). In addition, lack of access to water and sanitation services also contribute heavily to the disease burden of malnutrition. Recently, in an attempt to estimate how much disease can be prevented by environmental interventions, WHO examined the proportion of the GBD attributable to the environment. The study estimated that 24% of the GBD is due to environmental risk factors and that these risk factors fall disproportionately higher on people in developing countries and in particular on children in developing countries (Pruss-Ustun & Corvalan, 2006). Of the 102 diseases of concern, 85 had environmental contributions. More importantly, diseases such as diarrheal disease and malnutrition were deemed to have significant contributions of risk from the environment and hence potentially significant reductions by environmental interventions. The contribution of risk from the environment is the greatest for diarrheal diseases where the experts hypothesize that 94% of diarrheal disease could be prevented by environmental interventions in areas of water, sanitation and hygiene.

Diarrheal Disease and the Environment: A Historically Important and Recognized Relationship

The relationship between the environment and diarrheal diseases has been investigated by many. The International Water, Sanitation and Hygiene Decade (1981-1990) aimed at improving environmental conditions to limit transmission of pathogens and hence reduce diarrheal diseases. Two reviews from this time period formed the dominant paradigm for focused interventions to reduce diarrheal disease for a period of 10-15 years. In 1985, Esrey and others published a review that summarized the existing literature on environmental interventions to control of diarrheal diseases such as improvements in water, sanitation and hygiene. The authors examined 67 studies from 28 countries in research that spanned three decades (Esrey et al., 1985). The review found that average reduction in diarrheal disease among all interventions was 22% and that improvements in water quality had the least impact on diarrheal disease with a median 16% reduction. The study also found that improvements in both water availability and quality had the highest impact with 37% median reduction in diarrheal disease burden. In a second review, the authors examined the relationship between diarrheal disease and helminthic infections, and improved water and sanitation (Esrey et al., 1991). Again, this review found the lowest reduction of diarrheal diseases due to interventions in water quality (15% median reduction for rigorous studies). These papers (summaries of 144 and 67 studies) suggested that water quality interventions were poor control measures for reductions in diarrheal diseases and other diseases transmitted via the fecal-oral route such as intestinal parasites. For the next five to ten years the reviews were heavily referred to and cited. Those interested in water, sanitation and hygiene as a means for development regarded the work highly and the papers helped shaped the dominant paradigm for development interventions for years (T. F. Clasen & Cairncross, 2004).

2.3 A Change in the Dominant Paradigm

In the last ten years, a growing body of evidence suggests that the impact on diarrheal diseases by interventions in water quality at the household level was underestimated (Arnold & Colford, 2007; T. Clasen, Roberts, Rabie, Schmidt, & Cairncross, 2006; L. Fewtrell et al., 2005; M.D. Sobsey, 2002). One of the main reasons for this difference is that the evidence from earlier research on interventions in water quality focused on improvements in source water quality. However, interventions in source water quality cannot reduce contamination of drinking water that occurs after collection.

For the large majority of the world's population, household drinking water collection and storage is essential. Deterioration of household drinking water quality after collection has been widely documented. Contamination can be introduced after collection due to unclean storage containers and collection equipment as well as during storage. Deterioration of water quality can also be the result of microorganism growth and survival on drinking water container surfaces. In a study in rural Honduras, researchers documented increased concentration of *E. coli* in household stored waters compared to water sampled directly from the source (Trevett, Carter, & Tyrrel, 2004). In South Africa, researchers documented re-growth and survival of total coliforms and sometimes *E. coli* on surfaces of household drinking water containers (Momba & Kaleni, 2002). A meta-analysis of the existing literature revealed that bacteriological water quality deteriorates significantly after collection and that deterioration is proportionately greater in relatively uncontaminated source waters (Wright et al., 2004).

Research on household drinking water contamination after collection and during storage has lead to increased interest in household drinking water treatment and management of the water in the home. This growing body of research focuses on understanding household water management practices and implementing changes to improve water quality at the point-of-use (POU) of the consumer. The topic of household drinking water treatment has been reviewed and the research suggests that improving drinking water quality in the home has significant impacts on the diarrheal disease; a notion that has begun to shift the dominant paradigm of thinking about interventions in water, sanitation and hygiene (T. Clasen, Roberts et al., 2006; L. Fewtrell et al., 2005; M.D. Sobsey, 2002).

In 2002, the WHO published a draft report on various technologies in use around the world for household drinking water treatment (M.D. Sobsey, 2002). The review cited both laboratory and human health evidence to suggest that these technologies provide significant improvements in microbiological water quality and reductions in diarrheal disease. The technologies incorporate a variety of treatment methods such as chemical disinfection, non-chemical disinfection, filtration, and multi-barrier approaches to treating water in the home. Each technology has both advantages and disadvantages. For example, chlorine disinfection provides a residual disinfectant but is not effective against many protozoan parasites. Solar disinfection with combined heat and UV is effective against most pathogens but does not provide a residual disinfectant.

Since the initial review in 2002, two meta-analyses and a Cochrane review critically summarized and statistically analyzed the summary of the effects of intervention in drinking water quality and diarrheal disease (Arnold & Colford, 2007; T. Clasen, Nadakatti, & Menon, 2006; L. Fewtrell et al., 2005; M.D. Sobsey, 2002). A summary of the data is listed in Table 2.3. Table 2.3 is an adaptation of the table by Sobsey 2002 listing various technologies used for drinking water treatment; adapted with information published since his review. It should be noted that the information presented is focused on technologies used to treat microbiological contamination of drinking water and not chemical contamination.

As listed in table 2.3, there are currently five technologies that have been documented in the peer-reviewed literature to improve both microbiological quality of drinking water and reduce diarrheal disease. These technologies are: household chlorination and safe storage

(CDC Safewater system), solar disinfection with combined heat and UV radiation (SODIS), combined systems of chemical coagulation-flocculation and chlorine disinfection (PuR), ceramic filtration (ceramic candle filters and porous clay pots) and boiling. Fewtrell et al., (2005) combined results from 12 household drinking water quality intervention studies in developing countries and found household drinking water treatment reduced diarrheal disease by 35%. In the same meta-analysis, when source water treatment interventions were examined, they were found to reduce diarrheal disease by only 11% (L. Fewtrell et al., 2005).

Table 2.3: Summary of household drinking water treatment technologies

Technology	**Microbial reductions**	**Diarrheal Disease Reduction***	**Acceptability**	**Sustainability**
Boiling with Fuel	Yes, extensive	Yes	High	High, unless fuel is scarce
Solar Disinfection with heat and UV	Yes, extensive for most pathogens	Yes	High to moderate	High to moderate
Free Chlorine and Storage in improved vessel	Yes, extensive for most pathogens	Yes	High to moderate	High to moderate
Chemical Coagulation-Filtration + Chlorine Disinfection	Yes, extensive	Yes	High to moderate	Moderate
Ceramic Filtration - candle filter - porous clay pot	Yes, but limited reduction for viruses	Yes	High	High
Intermittently operated slow sand filtration (BSF)	Yes, but can be moderate or low	N/A*	High	High
Solar disinfection with heat only	Yes, extensive for most pathogens	N/A	High to moderate	High to moderate
UV disinfection with lamps	Yes, extensive for most pathogens	N/A	High to moderate	High to moderate
Onedrop (inorganic chemical mixture)	Yes, extensive for most pathogens	N/A	Not yet in use in the field	High to moderate
Lifestraw (halogenated resin)	Yes, extensive for bacteria	N/A	Moderate	High to moderate
Pureit (carbon block filtration and chlorine disinfection)	Yes, extensive for bacteria and viruses	N/A	High to moderate	High to moderate
Mission Filter (woven yarn filtration and chlorine disinfection)	Yes, extensive for bacteria	N/A	High to moderate	High to moderate

* - Reported in peer-reviewed literature only

In an expanded version of the meta-analysis produced as a report for The World Bank, Fewtrell and Colford examined the combined effects of chemical versus non-chemical household drinking water treatment interventions. The results suggest that chemical treatment was more effective; reducing diarrheal disease by 40% as compared to non-chemical disinfection that was found to reduce diarrheal disease by 29%. However, when the authors excluded one study on non-chemical disinfection, the estimate for non-chemical disinfection was increased to 46% reduction in diarrheal disease (L. Fewtrell & Colford, 2004). It is worthy to note that only one study on household filtration was included in this meta-analysis and the studies on non-chemical disinfection represent research on solar disinfection and boiling.

Clasen et al., 2006 performed a Cochrane review to assess the effectiveness of only interventions in drinking water quality at reducing diarrheal disease (T. Clasen, Roberts et al., 2006). The study summarized 30 trials. The authors examined the studies on multiple sub-group levels (i.e. intervention type, sanitation conditions, etc) and found that household drinking water interventions were more effective than interventions at the source. The report also found that the effectiveness of interventions seemed to be independent of water supply and sanitation conditions. In addition, the authors summarized results based on type of household water treatment method: chlorination, filtration, solar disinfection, or combined flocculation and disinfection. All treatment methods were found to significantly reduce diarrhea with an estimated reduction of 30-50%. However, the report suggests that household filtration offers the most consistent and effective results of the interventions in household drinking water treatment studied.

Recently, a meta-analysis was published summarizing the results from 21 studies all using point-of use chlorine disinfection (Arnold & Colford, 2007). When the results of all studies were pooled together, the estimate for chlorine interventions was a 30% reduction in diarrheal disease. An interesting finding from the meta-analysis was an attenuation of the effect of chlorine intervention on diarrheal disease over time; suggesting more research needs to be performed on the acceptability and sustainability of chlorine interventions (Arnold & Colford, 2007).

Interventions using methods other than chlorine disinfection are of interest for many reasons. Chlorine and solar disinfection do not change the appearance of the water after treatment. In addition, with very turbid water, chlorine and solar disinfection are less effective at reducing pathogens and potentially less effective at reducing diarrheal disease. Filtration methods may combat the problem of very turbid waters; changing the appearance of turbid waters and making them more amenable to additional disinfection processes.

Sobsey reviewed various filtration mechanisms with respect to their potential for use in the home (M.D. Sobsey, 2002). At the time of publication there were limited data on the ability of household filtration to effectively reduce diarrheal disease. Evidence existed on filtration methods to remove larger organisms that are vectors of helminthic and diarrheal diseases and the data show that these can be easily removed through filtration. For example, diarrheal disease caused by *Vibrio cholerae* was reduced when water was filtered through sari cloth because the bacteria associated with zooplankton were filtered out (Huo et al., 1996). However, simple cloth filtration or paper filtration will not remove even the largest parasites if they are not associated with larger particles and these filters only serve to strain out larger particles resulting in very limited removal of most of the waterborne pathogens.

Since the report published by Sobsey in 2002, more evidence exists to suggest household drinking water filtration can improve both microbiological quality and reduce diarrheal disease (T. Clasen, Garcia Parra, Boisson, & Collin, 2005; T. Clasen, Nadakatti et al., 2006; T. F. Clasen et al., 2004; Stauber et al., 2006). More specifically, in the last five years, ceramic filters (candle filters and porous clay pots) have demonstrated reductions in diarrheal disease in multiple locations: 70% in Bolivia, 60% in Colombia and 40% in Cambodia (Brown, 2006; T. Clasen et al., 2005; T. F. Clasen et al., 2004).

Other filtration technologies are being developed and used in developing countries but many of these technologies have limited to no data on either microbiological effectiveness or impact on diarrheal disease. Recently, a laboratory study investigated a water purifier that combines carbon block filtration with disinfection. Researchers found the unit removed 99.9999% of bacteria, 99.99999% of viruses and >99.9% of a surrogate for parasites but it has not yet been tested in the field (T. Clasen, Nadakatti et al., 2006). Other technologies are currently being used in households in developing countries but they lack both rigorous health data and microbiological quality data. For example, a two-bucket system with woven fabric and chlorine disinfection known as the Eagle Spring Mission Filter has been employed in countries like the Dominican Republic. In a small survey by researchers at the CDC, the filters were measured for concentration of free chlorine in finished water and were found to have lower than anticipated levels of chlorine in the finished water. The report did not document removal of *E. coli* but found that many filters installed in homes had broken and were not being used (Lantagne, 2004). A similar filter, the Gift of Water filter, was studied in 120 households in Dumay, Haiti. Households who used the filter were found to have a five point lower incidence of diarrhea and also found to have improved drinking water (Varghese, 2002). Another filtration product under development is the Lifestraw. Composed of a 6 µm mesh filter and a halogenated resin, this personal water purifier filter is being studied for its ability to remove microorganisms but has seen limited use in the field. The Lifestraw and other filtration technologies are still in the development stages and are not being widely employed in communities in developing countries unlike the biosand filter.

2.4 Slow Sand Filtration

The introduction of filtration in the early 20th century in the United States can be cited as a major advancement to improve health and reduce mortality (Cutler & Miller, 2005). In the United States, both slow sand filtration (SSF) and rapid granular medium filtration are employed; however, very few treatment plants use SSF. Slow sand filtration unlike rapid granular media filtration involves both physical removal as well as a biological treatment component. Other differences include no pre-treatment with coagulants or chlorine, extended retention time and different cleaning mechanisms. The differences between slow and rapid filtration are listed in table 2.4.

Due to different operating parameters between slow and rapid media filters, the principal purification mechanisms of SSF are thought to be "the result of straining through the developing filter skin and the top few millimeters of sand, together with biological activity" p. 21 in (*Slow Sand Filtration*, 1991). Biological activity and treatment is unique to the slow sand filter and is thought to be the result of the development of a microbiologically active community in the filter which is thought to be an important part of filter ripening. During the filter ripening process head loss increases, filtration rate decreases and microbiological performance improves. While SSF has been studied for more than 100 years, a clear description of the biological mechanisms in SSF has not been developed. Theories on the biological activity of the filter include: predation, scavenging, natural inactivation, metabolic breakdown, bactericidal effect of sunlight, bactericidal effect of algae, and increased sand stickiness.

Recent research on the mechanisms of slow sand filtration, and in particular on filter ripening, suggests that alum can significantly enhance the ripening process and improve reductions of *E. coli* (Weber-Shirk & Chan, 2007). In their research, the authors dosed a laboratory scale filter with aluminum extract from surface water. They found the extract enhanced ripening of the filter evidenced by increased head loss and improved performance in reducing *E. coli*. An interesting conclusion from this research suggests that the main form of ripening in this experiment was primarily physical-chemical and not mediated by biological processes. This is somewhat contrary to the large evidence base that already exists on slow sand filtration but does suggest that physical mechanisms of removal are as important, if not more important, than the biological component.

Many factors need to be considered when assessing physical mechanisms of removal in slow or rapid filters. The major differences that exist between slow and rapid sand filters have important effects on mechanisms of physical-chemical filtration and particle capture. These are listed in table 2.4 and include: effective size of the media, uniformity coefficient (affects collector size), filtration rate (affects approach rate), the size of the particle being captured (affects mechanisms of collection), temperature (affects viscosity), bed porosity (affects number of collectors), and bed depth and collision efficiency factor (affects chemistry of system and can be enhanced via chemical pretreatment).

Table 2.4 Differences in "typical" design criteria for slow and rapid sand filters

Design Criteria	Slow Filters	Rapid Filters
Filtration Rate	0.1 m/h	10 m/h
Water above top of sand	1.5 m	1.5m
Sand Depth	800 mm	800mm
Retention Time in Sand bed	3.2 h	2 min
Cycle Length	1-6 mo.	1-4 d
Effective size and uniformity coefficient*	ES = 0.2-0.5, UC ≤3	ES = 0.5 – 1.2, UC=1.1 – 1.5

(from Slow Sand Filtration 1991)

Three forces govern the size of particles captured via physical-chemical mechanisms: diffusion, sedimentation and interception (Yao, Habibian, & O'melia, 1971). Based on models by Yao et al., 1971, predicted particle removal in rapid sand filtration is poor for particles of 1 μm but improves for both larger and smaller particles. Removal of particles larger than 1μm increases quickly with increasing size and is governed by sedimentation and interception. Removal of particles less than 1μm increases with decreasing size and is primarily governed by diffusion.

For slow sand filtration, these mechanisms will be similar; however differences in design of the filter will affect particle removal in a variety of ways. For example, Harnoff and Cleasby (*Slow Sand Filtration*, 1991) perform a theoretical exercise in which they compare single collector efficiency between a slow sand filter and rapid sand filter assuming that the grain size of the slow sand filter is ½ the grain size of the rapid sand filter. For the same sized particle, single collector efficiency increases by 4 in the slow sand filter. If the bed depths of the two filters are the same, the number of collectors would be roughly double and collection efficiency would be 8 fold in favor of SSF. Next they examine the effect of approach velocity. They assume approach velocity will be 100 times slower for SSF and this increases single collector efficiency 100 times for the same size particle of the same density. Ultimately, they demonstrate a large increase in deposition efficiency predicated for slow sand filter by mechanisms of sedimentation and diffusion and a modest gain for interception. These gains in efficiency are theoretical and actual gains will be governed by design characteristics of the filters.

In slow sand filtration, particle removal primarily occurs in the top of the filter with some removal within the bed. For physical straining at the surface of a slow sand filter, particle capture is governed by grain diameter. Within bed particle capture is governed by transport and attachment. Transport and attachment have been well characterized in rapid granular media filtration. Particle removal within rapid filter beds includes mechanical processes of interstitial straining and transport into interstices. Transport and attachment are a function of three mechanisms: sedimentation, interception and diffusive transport. Yao et al. 1971 list three models for single collector efficiency for these three mechanisms:

$$\eta_S = V_S/V\ (\rho_S - \rho)g d_P^2/18\ \mu V \qquad (1)$$

$$\eta_I = 3/2(d_P/d_C)^2 \qquad (2)$$

$$\eta_D = 0.9(KT/\mu d_P d_C V)^{2/3} \qquad (3)$$

Where η_S is sedimentation, η_I is interception, η_D is diffusion, V_S is Stokes' settling velocity, V = filtration rate, ρ_S is density of the particle, ρ is density of water, g is acceleration due to gravity, d_P is diameter of particle, d_C is diameter of collector, μ is absolute viscosity, K is Boltzmann constant, and T is absolute temperature.

These equations predict that particle collection is influenced inversely by filtration rate, larger grain size and cold water of higher viscosity. Furthermore, smaller and less dense particles also hinder filtration unless they are small enough to be affected by mechanisms for diffusive transport. However, it is more difficult to determine the effect of particle size on the capture efficiency since all three mechanisms may be effective in removing some particles. In general, larger particles are removed by sedimentation and interception, and very small particles are affected mostly by diffusion.

While many of the mechanisms explained or inferred from the previous research on SSF are important, intermittent operation of the slow sand filter poses additional mechanistic questions. For example, intermittent operation will result in acceleration through the filter bed when operation starts. This may result in shearing and detachment of particles. Furthermore, depending on the length of pause, biological activity may be limited and effects of biofilm or predation may diminish.

2.5 The Biosand Filter

The biosand filter (BSF) was developed in the late 1980's and early 1990's by Dr. David Manz at the University of Calgary, Alberta, Canada. Over the period of 1991 to 1995, the filter was tested and modified both in the laboratory and in small field studies in Nicaragua and Honduras (Buzunis, 1995; Manz, Buzunis, & Morales, 1993). The first biosand filter tested in the field in Nicaragua, shown in Figure 2.1, varies somewhat in design compared to the filter in use today. Since its development, current estimates suggest that more than 80,000 biosand filters have been installed in homes in more than 60 countries serving a population of approximately 500,000 people. Implementing organizations have done substantial amounts of research on the filter in the field, ranging from assessing user acceptance to reductions of indicator bacteria in water; a body of research that began in 1993. The first biosand filters were installed in March 1993 in Nicaragua in a pilot project funded by the Pan-American Health Organization (Manz et al., 1993). A design of that filter is illustrated in figure 2.1. In this project, filters were tested approximately 3 months after installation in July 1993 and were found to be reducing fecal coliforms by 99% or more.

A cross-section of the biosand filter model in use currently is shown in Figure 2.2. The BSF consists of a concrete or plastic chamber filled with sand with an elevated discharge tube that allows the filter to maintain a layer of water above the sand surface and prevents dewatering. The BSF is similar to a conventional slow sand filter (SSF) in that there is typically no pretreatment or backwashing and operation is simple, including gravity-driven rather than pressure filtration.

As in conventional SSFs, the sand bed remains wetted throughout operation and a ripening process occurs, head loss increases, a biological layer may develop and performance improves. However, the BSF does not operate continuously but instead, intermittently wherein a single charge of feed water (typically up to 20 L although multiple daily charges are possible) is made each day. During this charge, the operation is in a declining rate mode of filtration. A portion of the charged water remains in the BSF until the next charge. The time period when water is no longer discharging from the filter is referred to as the idle time.

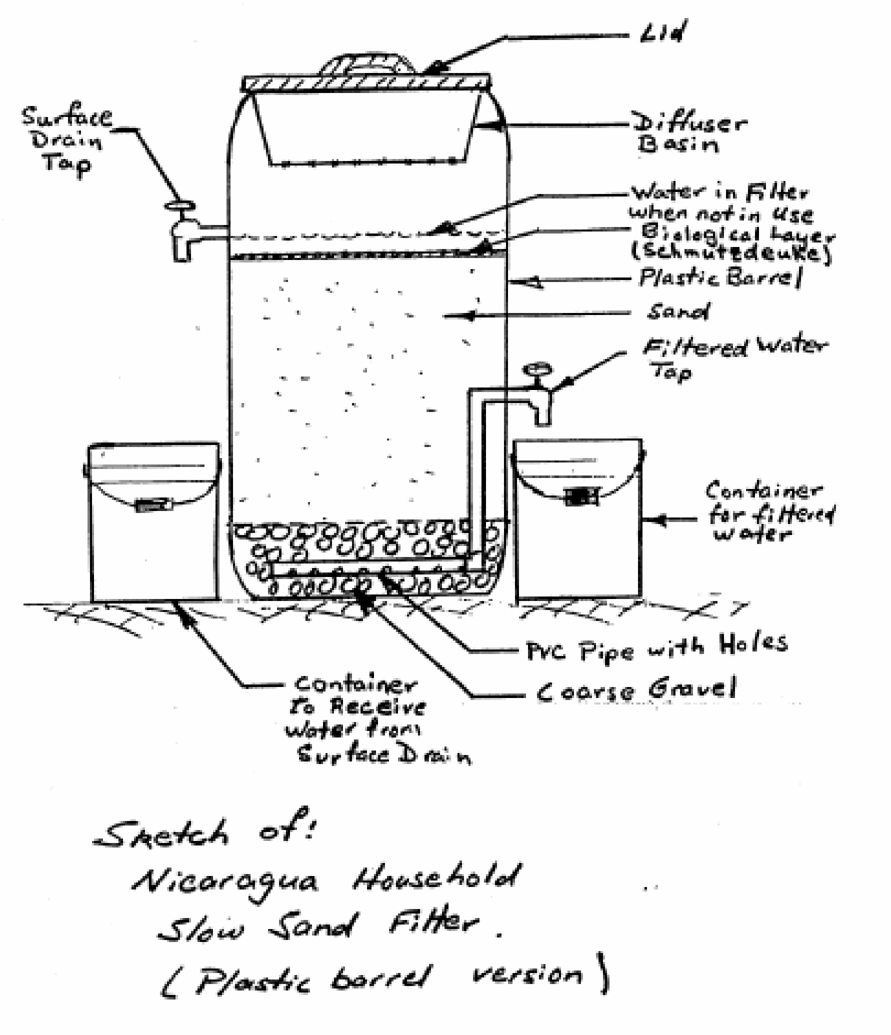


Figure 2.1 First version of the biosand filter tested in homes in Nicaragua in 1993

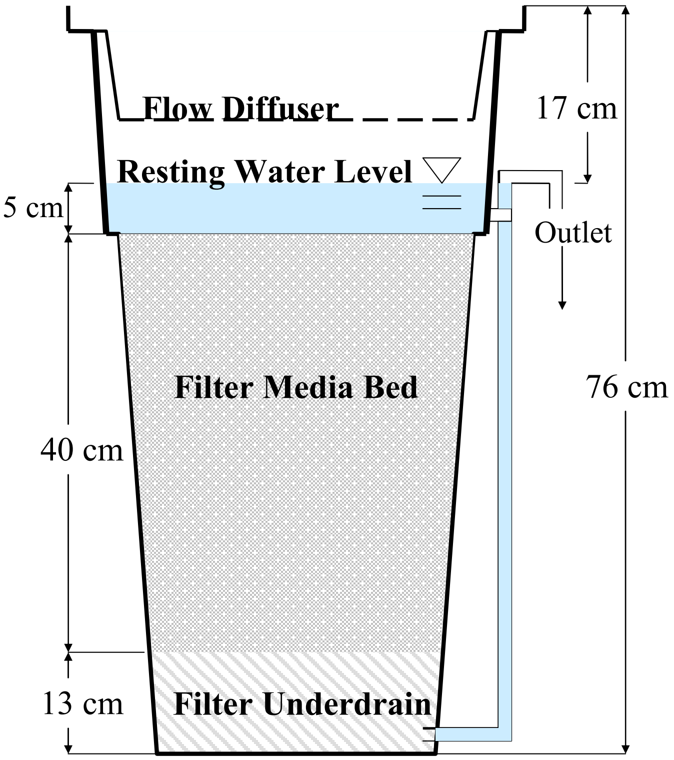


Figure 2.2 Cross-section of the plastic biosand filter (courtesy of Mark Elliott)

As shown in Figure 2.2, dewatering of the filter between charges is avoided by a vertical discharge tube that rises from 2-7 cm above the height of the filter media. The elevated outlet allows the media to remain saturated after a charge has been filtered but water is no longer flowing from the outlet. Another unique aspect of the BSF design is to promote uniform drip flow over the sand surface by use of a plastic or sheet metal diffuser above the filter media. This diffuser prevents the charge of water from disturbing the biolayer.

The design of the BSF differs significantly from that of the SSF. The maximum filtration rate of the BSF is up to 100 times greater than for the SSF (1 m/h in contrast to a recommended 0.08-0.4 m/h)(*Slow Sand Filtration*, 1991). The depth of the BSF sand layer is about 50% less than for the SSF (0.4 m compared to a recommended starting depth of >0.8 m for the SSF with a minimum of 0.5 – 0.7 m). The range of particle size of the BSF sand is typically broader than in SSF (e.g., the uniformity coefficient may typically exceed 4.0, compared to a recommended value of <3 for the SSF). In addition, the quality of the sand differs because the BSF is constructed with material that is locally available whereas sands used in most SSF are obtained from a commercial source. To provide quality control on local sand selection, the typical procedure is to measure the initial flow rate of a newly loaded filter following a 20-L charge. If the flow rate falls outside a prescribed range (usually 0.7-1.1 L/min), the particle size is either too small (flow rate is too low) and the sand requires further washing or too large (flow rate is too high) and thus unacceptable for use.

While currently being used in many developing countries, there is relatively little published literature on field or laboratory data from the biosand filter. The most systematic laboratory research to date has been documented in a thesis that examined fecal coliform removals and attempted to determine the extent of oxygen transfer into the biolayer from the standing water (Buzunis, 1995). Buzunis documented average removal of fecal coliform bacteria at 96% when the filter was dosed daily with 25 L of duck pond water. Other laboratory research has been performed by researchers at Massachusetts Institute of Technology (MIT). In their first study, 20 L of river water was added to the biosand filter for 45 days and then the filter was measured for its ability to remove total coliforms during a period of approximately one week of operation. Average reduction of total coliforms was found to be 99.5% (Lee, 2001). Another thesis from MIT performed a 29-day laboratory experiment. In this experiment, filter was dosed daily with 5 L of river/municipal waste water mix and sampled twice each week. In this experiment, the BSF was found to provide an average of 90% (range 52-97%) reduction of thermotolerant coliforms (Donison, 2004). Until 2006, there was only one published paper on the biosand filter; a laboratory study of the filter examining the removal of microorganisms and toxicants via biosand filtration. In this study, the filter was found to remove greater than 99% of protozoan parasites, 83% of heterotrophic bacteria and 50-90% of inorganic and chemical pollutants (Palmateer, Manz, & Jurkovic, 1999). There are no peer-reviewed published laboratory studies to document virus removal but in his report Sattar (1998) documented removals for total and fecal coliforms ranging from 74%- 93% and average Hepatitis A virus removals of 66%. More recently, systematic microbial challenge studies with bacteria and virus under controlled laboratory conditions have been reported and are currently being performed (Elliott et al., 2006; Stauber et al., 2006)

After initial field implementation and laboratory results were favorable, many organizations decided to take up and implement the biosand filter as a household water treatment process. A large body of grey literature exists primarily in the form of reports and theses on the performance of the biosand filter in the field. Both the peer-reviewed and the grey literature are summarized in Table 2.5. As shown in Table 2.5, the biosand filter has received wide study and implementation in many regions of the world; however it demonstrates highly variable microbiological performance and has not thoroughly been evaluated for health impact.

Of the field evidence on the BSF, the largest study was undertaken by Samaritan's Purse Canada. This organization has implemented 75,000 biosand filters worldwide including Kenya, Ethiopia, Mozambique, Brazil, El Salvador, Cambodia, and Vietnam (*Samaritan's Purse Canada Website*). In the evaluation of the BSF, they tested 577 biosand filters in six countries on three continents (Kaiser et al., 2002). The study found an average 93% reduction of fecal coliforms and reported that 98% of the filters were still regularly used. Recently, 107 filters in the Artibonite Valley of Haiti were sampled. This study found an average of 98.5% removal of *E. coli* in filters that had been used on average 2.5 years (Duke et al., 2006). In an additional component to their study, researchers installed new biosand filters in Haiti and measured microbial reductions of *E. coli* over time for a period of three months. In the new set of filters, much lower *E. coli* reductions were found; as low as 73% reduction and increasing to 85% after three months (Baker, 2006). As shown in Table 2.5, field performance of the biosand filter varies greatly with some filters providing apparently negative reductions while other filters provided greater than 99% reduction of *E. coli* and fecal coliforms.

Maertens and Buller assessed the two-week point prevalence of diarrheal disease in households with biosand filters and control households in Ethiopia. Biosand filter users reported 82% less occurrences of diarrheal disease compared to the control households (Maertens & Buller, 2006). However, control households had significantly different surface water sources for drinking water, were located in separate villages, and represent potentially very different risk factors for diarrheal disease. The Centre for Affordable Water and Sanitation Technology assessed diarrheal disease in their longitudinal study in the Artibonite Valley in Haiti. Households were monitored for diarrhea one month prior to biosand filter installation and then three months following in bi-weekly household visits. Their results suggest an improvement of diarrheal health but they do not provide an estimate of that level of reduction (CAWST, 2006).

The majority of implementation assessments have asked users to rate their health status since installation of the biosand filter; which invariably is always better. To date there has been no rigorous scientific study on the ability of the biosand filter to reduce diarrheal disease in users as compared to non-users; and the lack of this evidence is a major barrier to scaling up implementation.

Table 2.5: Summary of peer-reviewed and grey literature on the BSF

Reference	**Summary of Study**	**Average Reduction (Sample Size)**
(Manz et al., 1993) Field study in Nicaragua	Four BSFs installed and sampled two months later.	99.5% for fecal coliforms (n = 3)
(Buzunis, 1995) Laboratory study in Canada	Tested filter for 2.5 months; dosed daily with environmentally contaminated surface water.	96% (range 99.7-91.1%) for fecal coliforms during sampling on days 10-42.
(Sattar, 1998) Laboratory study in Canada	Filter dosed with 60 L of water with high algal content; then dosed for 28 days with 20 L of untreated surface water. Hepatitis A virus dosed onto filter; other bacteria measured were naturally occurring.	89.8% for total coliforms (n= 4) 87.4% for fecal coliforms (n = 4) 66 % for Hepatitis A virus (n = 3)
(Palmateer et al., 1999) Laboratory study in Canada	Filters dosed with surface waters until biofilm formed (~ two weeks). Chemicals and microorganisms were dosed. One-time dose of 10^6 *Cryptosporidium* and 10^5 *Giardia* then sampled. Naturally occurring bacteria measured.	>99.999% for Giardia (n=1) 99.98% for Cryptosporidium (n=1) 83% for heterotrophic plate counts bacteria (n = 5) 50-99% reduction of organic and inorganic chemicals

(Kaiser & Chang, 1998) Field study in Vietnam	100 filters installed in Ha Tay province in the community of Lai Yen.	95.8% for fecal coliforms (n= 38)
(Snider, 1998) Field study in Western Kenya	25 filters installed in Londiani, Western Kenya	93% for *E. coli* (n = 25)
(Lee, 2001) Field study in Nepal, Laboratory study at MIT	39 filters sampled in Nepal. In laboratory at MIT, BSF studied for 2 months. Filter dosed daily for 45 days with 20 L surface water prior to sampling. Naturally occurring bacteria sampled.	99.5% for fecal coliforms in laboratory study (n=5)
(Mol, 2001) Field study in Kenya	110 filters installed in Machakos District in Eastern Kenya.	93% *E. coli* (n=110)
(Kaiser et al., 2002) Field study in Honduras, Nicaragua, Mozambique, Kenya, Cambodia and Vietnam	Evaluated the BSFs in six countries; tested 577 filters and interviewing users. 94.6% - 100% of users said BSF improved health of their household. 98.4% still used the filter and 88.5% on a daily basis.	93% for fecal coliforms (n=577) Honduras 100% Nicaragua 99% Mozambique 98% Kenya 94% Cambodia 83% Vietnam 81%
(Lantagne, 2004) Field visit in Dominican Republic	Visited 10 BSFs in Playa Oeste, Puerto Plata, Dominican Republic. Filtered water sampled.	Filtered water positive for total coliforms but not *E. coli*.
(Donison, 2004) Field study in the Dominican Republic and laboratory study at MIT	45 BSFs in Dominican Republic were visited. Laboratory study at MIT, filters dosed with 5 L of a 1:10 mix of waste water to river water for 29 days; sampled twice each week.	In 5 communities, 80% for total coliforms, and in two communities <0% for total coliforms. 90% *E. coli* in laboratory study (range 52-97%) (n = 7)
(Maertens & Buller, 2006) Field study in Ethiopia	> 500 filters installed in the Oromia region, Liben Woreda district, Ethiopia. 50 BSF households and 50 control households were interviewed. Filters test for bacterial reductions. Control households reported higher two week point prevalence of worms, skin infections, vomiting, and diarrhea.	98.6% for total coliforms (n=50) 97.3% *E. coli* (n=50) 85% for turbidity (n=50)

(Earwaker, 2006) Field study in Ethiopia	57 BSFs from the Oromia region Liben Woreda district of Ethiopia. 39 filters were sampled.	87.9% for *E. coli* (n=39)
(Duke et al., 2006) Field study in Haiti	107 households with BSFs installed in Artibonite Valley, Haiti were interviewed and water samples were taken. Filters had been in use for an average of 2.5 years.	98.5% for *E. coli* (n = 92) (10 samples omitted)
(Baker, 2006; CAWST, 2006) Field study in Haiti (longitudinal component of study listed in Duke et al., 2006)	80 households received BSFs and were followed for 3 months for water quality and diarrheal disease. Diarrheal disease was assessed prior to and after filter installation and compared with households who had received filters > 2 years prior. Most indicators of diarrheal disease improved after installation of filter.	76% *E. coli* (n = 80 filters but sampled repeatedly)
(Stauber et al., 2006) Field study in Dominican Republic and laboratory study at UNC	Two laboratory BSFs were dosed daily with 40 L of surface water inoculated with *E. coli.* Filters sampled over 3-6 weeks. 55 BSFs in Bonao, Dominican Republic had been installed 4-11 months prior to sampling.	94% for *E. coli* in laboratory studies. 93% for *E. coli* in filters in the field (n=55)
(Elliott et al., 2006) Laboratory study at UNC	4 experiments with daily dosing volumes of 20 or 40 L surface water inoculated with *E. coli* and viruses (coliphage and echovirus type 12).	73.6% was initial reduction for *E. coli* reduction. Improved to 97.5% after 30 days. Similar results for coliphage 69% initially then improving to 90% after 30 days. 95% for Echovirus
(CAWST) Summary of all lab and field tests (website)*	-Family Bible Fellowship in Guatemala and El Salvador. 31 field tests performed in 2002. -Global outreach student's association in Guatemala in 2001. -Biosand water filter project in Nicaragua in 1999 -Samaritan's Purse in Brazil in 1998	-83.1% for *E. coli*, 89.16% for coliforms (n=31) -99.6% for coliforms (n=3) -79.9% for fecal coliforms (64.4-95.0%) -99.7% for fecal coliforms, 98.64% for *E. coli* (n=55)

* - These studies were summarized by the authors in the reference and not by this author

2.6 The History of Biosand Filtration in the Dominican Republic

The biosand filter was introduced into the Dominican Republic through a filter technician training program in October 2000 promoted by the non-governmental organization (NGO) Add Your Light, the Canadian Embassy, and the Rotary Foundation in Calgary. Fourteen technicians were trained how to make and install the biosand filter by Dr. David Manz. The technicians, supported by international non-governmental organizations, began to build filters and sell or supply them (with subsidies) to families in the DR. By January 2002, there were 1000 filters in the DR and the implementation program was growing. In the next three years, 3000 filters were made and installed in the DR. Most of the filter work was located near or around where the filter makers worked and lived or in Puerto Plata, Dajabon and other areas near the northern coast of the island. The developed implementation program which had only limited regional distribution of filters made the DR an ideal location with which to perform the type of rigorous scientific study needed to document diarrheal disease reduction as a result of filter use.

Recent estimates from the Joint Monitoring Program of the WHO and United Nations Children and Environment Fund (UNICEF) cite that approximately 98% of the urban and 60% of the rural population have access to improved water (*Joint Monitoring Programme Coverage Estimates: Improved Drinking Water*, 2006). This estimate includes a significant portion of the population relying on bottled water as an improved source of drinking water. While many in the population have access to a piped source within 15 minutes of the home, the piped supplies are not typically reliable. They provide intermittent flow and are recognized to be of poor water quality. Due to these factors, households are forced to store water in the home and/or purchase bottled water if possible.

In addition to poor water quality, diarrheal disease continues to be a burden to the population. A 2002 Demographic and Health Survey cite that 14% of all children are suffering from diarrhea during a two-week survey and that the burden in those between six months and 24 months surpasses 20% (*Encuesta Demografica y de Salud: Republica Dominicana*, 2003). Increased disease burden above national average was also found in four Provinces in the country: Bahoruco 24%, Barahona, 24%, Independencia 29% and Monsenor Nouel 22%. The high burden of diarrheal disease in these communities (as compared to the other provinces) suggests specific risk factors that can be controlled such as water quality. These places represent ideal location in which to implement household water treatment by the biosand filter; especially since very few filter implementation projects have taken place in these regions.

Limited research has been performed in the Dominican Republic about diarrheal disease and household drinking water treatment. However, boiling water is typically encouraged because of poor piped water quality (McLennan, 2000). This study cites high biomedical knowledge of diarrheal disease prevention practices but suggests less than 50% of interviewed care givers practice water treatment. Perhaps giving households another option in drinking water treatment can increase the percentage of care givers practicing diarrheal disease prevention strategies.

Chapter 3: Reduction of *E. coli* bacteria and MS-2 and PRD-1 Viruses by the Biosand Filter under Controlled Conditions

3.1 Introduction

One of the most promising POU filtration technologies is the biosand filter (BSF), a household-scale, intermittently operated slow sand filter. Although the ability of traditional slow sand filtration (SSF) to reduce pathogens in water is well-documented, the effectiveness of the BSF unit in reducing waterborne microbes is uncertain because it has different design and operating properties from conventional SSFs. Traditional slow sand filters operate continuously at constant head and flow rate and the upper layer of sand is periodically replaced when it becomes clogged. However, the BSF is operated intermittently, head and flow rate vary and the upper few centimeters of sand containing the schmutzdecke are not replaced but rather cleaned periodically by agitation and decanting of the released contaminants and excess biological growth.

Only limited evidence of the ability of the biosand filter to reduce waterborne microbes in laboratory or field studies has appeared in the peer-reviewed scientific literature. Duke et al., (2006) documented 98.5% bacterial reduction efficiency in filters that had been in use for 2 years or more in a field study in Haiti. Palmateer et al., (1999) documented >99% reduction of *Giardia* cysts and *Cryptosporidium* oocysts and 65-90% reductions of indigenous fecal coliform bacteria in a laboratory study. To date, no studies have been published on reductions of *E. coli* by the BSF in under controlled laboratory conditions. As *E. coli* is the recommended fecal bacterial indicator of drinking water quality and is less prone to variability and uncertainty caused by the diversity and re-growth of fecal coliform bacteria, studies on its reduction by the BSF are much needed. In addition, no published studies have documented the ability of the biosand filter to remove viruses from water.

The goal of the laboratory research was to evaluate the maximum reduction efficiency of the biosand filter by controlling: frequency of dosing, source water quality, and constant concentration of microorganisms. These conditions differed significantly from existing research which evaluated the BSF under fluctuating source water quality conditions and/or dosing conditions such as in households. The objective of this research was to document, under controlled laboratory conditions, the reduction of *E. coli* bacteria from feed water seeded with a consistent input level of this bacterium. In a second experiment, the reduction of two bacteriophages (MS-2 and PRD-1) was also documented.

3.2 Methods

Laboratory Filter Preparation

Plastic filter units, 60-L capacity, were obtained from Davnor Water Treatment Technologies Ltd. (Alberta, Canada). In an attempt to prepare the laboratory filters as they are prepared in the field, all media materials were prepared according to field instructions for the concrete biosand filter. Crushed granite gravel was purchased locally and sieved through three mesh screens to prepare filter media of the appropriate size (mean diameter of ≤ 1 mm), in accordance with current field practice, to provide an initial flow rate of 0.7 – 1.1 liters per minute (L/min). Filters were loaded with 5 cm of under-drain gravel, 5 cm of medium size gravel, and 40 cm of sand. During loading, the empirical pore-volume of the filters was measured by loading the filter to saturation. After loading the filter, the initial flow rate was measured by filling the upper filter chamber full and measuring the time it took to filter 500-mL of water.

Constant Dosing, Filtration Experiments

Two filtration experiments with constant daily dosing volumes of water were conducted. In the first experiment, a filter was dosed for 17 days with 40 L/day of lake (reservoir) water seeded to achieve an initial concentration of 10^5 colony-forming units per mL (CFU/mL) of *E. coli* B. In the second experiment, a filter was dosed daily for 43 days with 40 L of lake water seeded to achieve an initial concentration 10^2 CFU/mL of *E. coli B* and 10^2 plaque forming units per mL (PFU/mL) of both MS-2 and PRD-1. The lower concentrations of *E. coli* and coliphages of this second experiment were considered more typical of the concentrations of these enteric microbes found in fecally contaminated water, compared to the much (1000-fold) higher initial concentration of *E. coli* used in the first experiment.

In both experiments, raw influent surface water was collected from the local drinking water treatment plant (Orange Water and Sewer Authority, Orange County, NC, USA) at weekly intervals and stored at 4 ºC until one day prior to dosing. Then, 40 L of water was allowed to come to room temperature (approximately 25 ºC) prior to dosing onto the filters. For both runs, the filters were dosed daily with 40 L of water that was seeded to the initial target concentrations of the test microorganisms stated above.

E. coli, MS-2, PRD-1 Filter Dosing

A pure culture of *E. coli* strain B (ATCC No. 11303) was grown to log phase in shaker culture flasks of tryptic soy broth at 36°C, as described in EPA Method 1602 (EPA 2001). After reaching log phase, the culture was cooled to approximately 4ºC, serially diluted in phosphate buffered saline and spread plated onto MacConkey agar (Becton-Dickinson, Franklin Lakes, New Jersey). Plates were incubated at 36 ºC for 24 hours and resulting colonies were counted to express the *E. coli* concentration as CFU/mL.

Log-phase cultures were stored for up to 7 days at 4ºC and maintained stable concentrations of viable *E. coli*. Cultures of *E. coli* were prepared weekly during the two dosing experiments. For daily dosing, the culture was serially diluted in lake water immediately prior to seeding to prepare a stock suspension and this stock was dosed into lake water to achieve the desired *E. coli* concentration in water to be dosed onto the filter.

Stocks of bacteriophages MS-2 and PRD-1 were grown, enumerated by double agar layer procedure (EPA, 2001) and stored at -80ºC. Aliquots of each stock were thawed each week, serially diluted ten-fold in phosphate buffered saline and stored at 4ºC for up to 7 days. Aliquots of this dilution were then seeded into feed water to achieve desired coliphage concentration for each daily charge of seeded water.

Water Analysis Methods

During water analysis, one 500 mL sample was drawn from the 40 L seeded lake water prior to dosing onto the filter as a composite of influent water. In addition, the first 30 L of filtered water was collected and a 500 mL sample was drawn as a composite of filtered water. *E. coli* in composite influent water samples from 40-L daily doses, composite filtered water samples (from the first 30-L of filtered water) and samples of the seeded influent water from the day prior to sampling, were quantified by membrane filtration on MI agar BBL™ (Becton-Dickinson, Franklin Lakes, NJ) using EPA Method 1604 (EPA, 2002). MS-2 and PRD-1 in water were assayed using the single agar layer method (EPA Method 1602, EPA, 2001). Turbidity and pH were measured using a turbidimeter (Model 2100N, Hach, Loveland, CO.) and pH meter (Model 215, Denver Instruments, Denver, CO.). In both experiments, composite samples of total volume introduced and total volume filtered were taken on day 0, day 1 and then at approximately weekly intervals throughout the length of the filter challenge study.

To examine the effect of time spent in the filter, a composite of the first 15 L of filtered water (which is water that was retained in the filter bed from the dose of seeded influent water of the preceding day) was sampled and analyzed on Days 42 and 43 of the second experiment. Log_{10} reductions of *E. coli*, MS-2 and PRD-1 were calculated as $\log_{10}$ influent water concentration minus $\log_{10}$ filtered water concentration, as shown in the following equation:

$$\text{Log}_{10}\text{ reduction} = \text{Log}_{10}\text{ influent concentration} - \text{Log}_{10}\text{ filtered water concentration} \qquad (1)$$

The term log10 reduction was used because an infectivity method was used to determine the concentrations of the microorganisms in the water. Because an infectivity assay was used, only a reduction can be determined since the absence of the microorganisms does not mean that it is not present but that it is not able to be cultured.

In addition to monitoring *E. coli* reductions based on analysis of composite filtered samples, in the second experiment we monitored filtered water concentrations of *E. coli* after various volumes (0, 2, 5, 10, 15, 20, 25, and 30 L) of the daily 40 L water dosing were filtered. These values were monitored on three sampling days: day 35, day 42 and the day the biolayer was punctured accidentally during sampling (day 45).

3.3 Results

Filter Flow Rate

The flow rates of filters over the course of the filter runs of the two laboratory experiments are summarized in figure 3.1. During both experiments, the filter flow rates declined significantly over time. The initial flow rate during experiment #1 was 0.67 L/min, and by Day 17 it had declined to 0.09 L/min. Experiment #2 began with an initial filter flow rate of 0.9 L/min, and it declined to 0.2 L/min by Day 25. These declines are thought to be due to filter ripening or maturation and the development of the biologically active surface layer or “schmutzdecke” typical of slow sand filters. Initial filter flow rates were different in the two experiments due to media preparation and filter packing. However, it is noteworthy that the rate of decline in filter flow rate is similar for both experiments, suggesting that filter ripening was occurring at similar rates in both experiments.

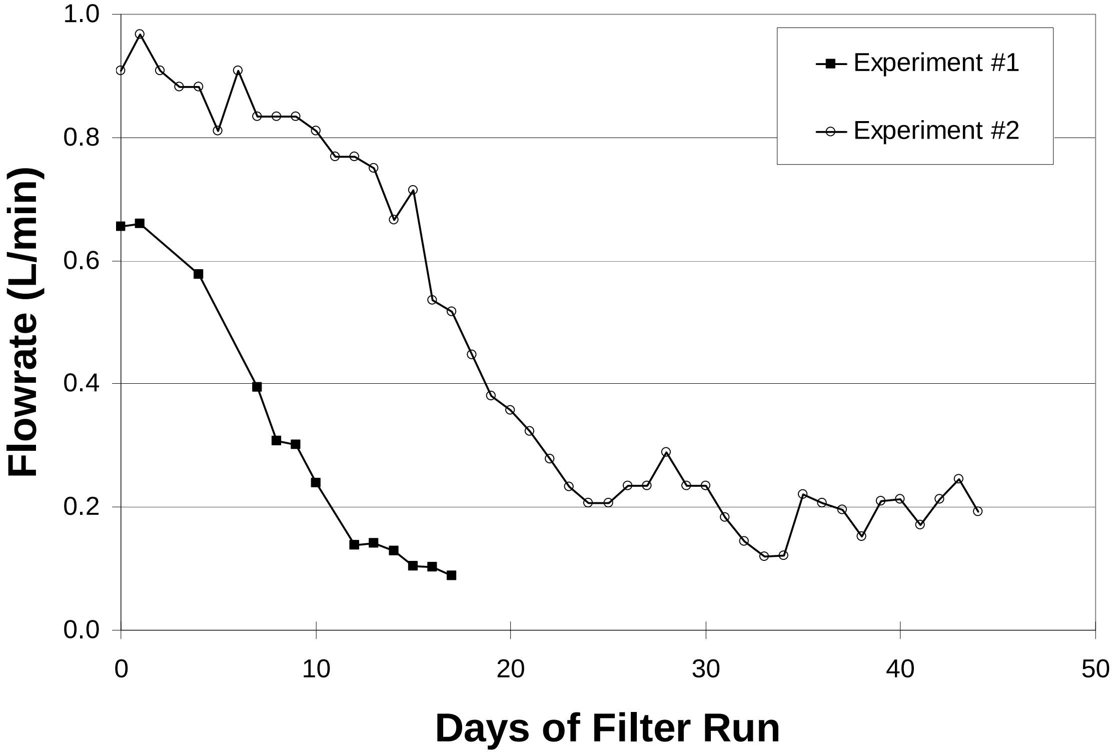


Figure 3.1 Flow rate following initial 20-L charge of a 40-L dose for BSFs over time

Reduction of *E. coli*, MS2 and PRD-1 in Composite Samples

In experiments 1 and 2, geometric mean reductions of *E. coli* by the biosand filter were 97% and 91%, respectively. In both experiments, the lowest *E. coli* reductions were found during initial days of filter dosing. The minimum *E. coli* reduction in the first experiment was 1.2 $\log_{10}$ (93%) measured on day 4, and in the second experiment it was 0.43 $\log_{10}$ (or 63%) on day 3. Maximum *E. coli* reductions typically occurred towards the end of the filter dosing experimental period. Maximum *E. coli* reduction in the first experiment was nearly 2.0 $\log_{10}$ (or 99%), reached on day 17, and maximum *E. coli* reduction in the second experiment was 1.9 $\log_{10}$ (or 98.9%), reached on day 42. The improvement in *E. coli* reductions to about 98% during the length of the filter run is shown in figure 3.2 for both experiments.

Coliphages MS-2 and PRD1 were dosed to the BSF in experiment #2. Over the entire experimental period, virus reductions were much lower than bacteria reductions, with maximum reductions of 78% and 87% for MS-2 and PRD-1 respectively. Therefore, compared to *E. coli* bacteria, reduction of the two coliphages was relatively poor for both virus types and remained below 1 $\log_{10}$ (90%) for the duration of the experiment. However, virus reductions increased over time in the filter run in similar fashion as for *E. coli* (as shown in Figure 3.3), again suggesting improved performance as the filter ripened.

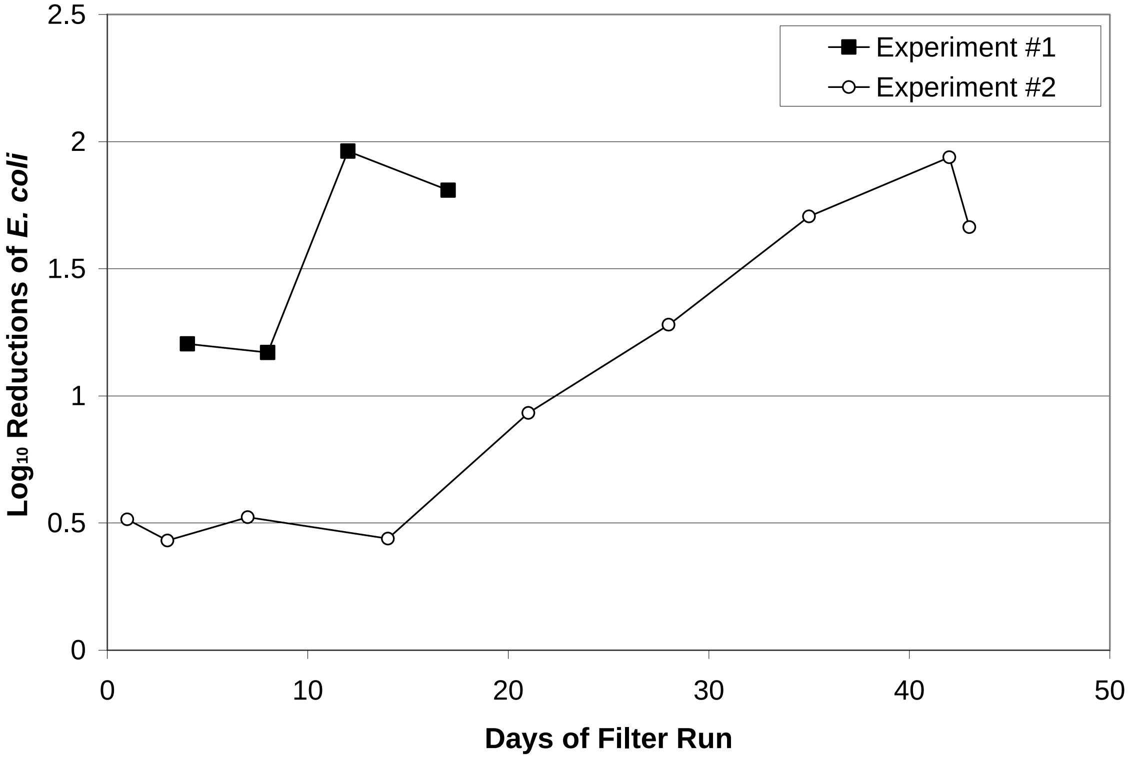


Figure 3.2 Composite $\log_{10}$ reductions of E. coli from dosed water as a function of time in BSF experiments

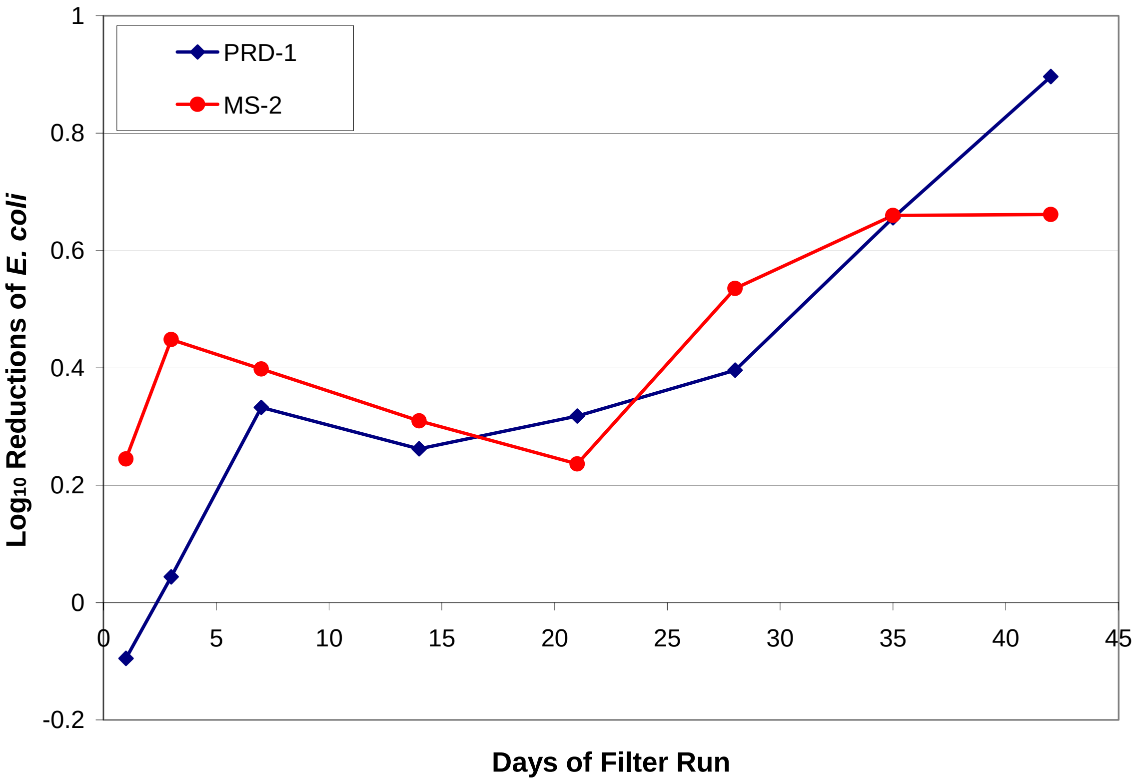


Figure 3.3 Composite $\log_{10}$ reductions of coliphages MS-2 and PRD-1 from dosed water as a function of time in experiment #2

Average $\log_{10}$ reductions for all three test microorganisms were calculated and compared for the period from day 3 to day 42 of experiment #2. The average values for *E. coli*, MS-2 and PRD-1 were: 1.0, 0.46 and 0.41 $\log_{10}$, respectively. The average $\log_{10}$ reduction for *E. coli* was significantly different when compared to MS-2 and PRD-1 as determined by repeated measures ANOVA ($p < 0.05$). However, the average $\log_{10}$ reductions for MS-2 and PRD-1 were not significantly different from one another.

In order to evaluate the effect of ripening on filter performance for microbe reductions, the results from the BSF run during experiment #2 were divided into two time periods: an unripened and a ripened period. The distinction between unripened and ripened was based on the change in filter flow rate. Prior to day 21, filter flow rate was > 0.2 L/min.

After day 21, the filter flow rate remained at 0.2L/min. Based on this distinction, average $\log_{10}$ reductions were calculated for the first four sampling points and second four sampling points, classified as the unripened and ripened period respectively. These data are presented in table 3.1. Based on repeated measures ANOVA, statistically significant differences were found between unripened and ripened periods for reductions of *E. coli* and PRD-1; with reductions found to be significantly higher in the ripened period. However there was not a statistically significant difference in the reductions of MS-2 during the two time periods.

Table 3.1: Effect of ripening on reductions of *E. coli* MS-2, PRD-1 in experiment #2

Filter operating period (days of analysis):	**Mean *E. coli*** LRV* (%)	**Mean MS-2** LRV (%)	**Mean PRD-1** LRV (%)
Entire experimental period (days 3, 7, 14, 21, 28, 35, 42)	1.0 (90)	0.46 (65)	0.41(61)
“Unripened” period; first 4 sampling days (days 1, 3, 7, 14)	0.48 (67)	0.35 (55)	0.14 (28)
“Ripened” period; second four sampling days (days 21, 28, 35, 42)	1.46 (97)	0.52 (70)	0.57 (73)

* - LRV = log reduction value

Effect of Water Volume Filtered and Contact Time

In table 3.2 and table 3.3 are shown the $\log_{10}$ reductions of *E. coli* and MS-2 and PRD-1 in 15 L composite samples of initial filtrate from a daily water dose of 40 L. In Table 3.2 are shown the reductions on days 42 and 43 of the second experiment. *E. coli* reduction was approximately 0.3 $\log_{10}$ higher in the 15 L composite samples (2.2 and 2.0 $\log_{10}$) when compared to 30-L composite samples taken the same day (1.9 and 1.7 $\log_{10}$). Table 3.3 suggests a similar trend for the coliphages with an increase of 0.5 $\log_{10}$ reduction higher in the 15-L composite samples (1.1 and 1.5 log10 for MS-2 and PRD-1, respectively) compared to the 30-L composite samples (0.66 and 0.9 log10 for MS-2 and PRD-1, respectively) on day 42 of the second experiment. The reductions of *E. coli*, MS-2 and PRD-1 were greater for water that remained in the pores of the filter overnight, compared to the water that came through the filter afterwards and was only in the filter for several hours during the day of dosing. Water that passes through the filter during a dosing and did not stay in the filter overnight has lower $\log_{10}$ reductions for all three test microorganisms compared to dosed water that stayed in the filter overnight.

Table 3.2: Effect of volume filtered on *E. coli* reduction by BSF in experiment #2

Water Volume Filtered	***E. coli* (Day 42)** LRV (%)	***E. coli* (Day 43)** LRV (%)
15-L Composite; overnight water	2.2 (99.4%)	2.0 (98.9%)
30-L Composite; same day water	1.9 (98.8%)	1.7 (97.8%)

Table 3.3: Effect of volume filtered on *E. coli*, MS2 and PRD-1 reduction by BSF in experiment #2 on day 42 of dosing

Water Volume Filtered	***E. coli*** LRV (%)	**MS-2** LRV (%)	**PRD-1** LRV (%)
15-L Composite; overnight water	2.2 (99.4%)	1.1 (92.2%)	1.45 (96.4%)
30-L Composite; same day water	1.9 (98.8%)	0.66 (78.2%)	0.90 (87.3%)

The effect of volume filtered was further investigated by collecting grab samples of water for *E. coli* analysis at multiple times and their corresponding filtrate volume points during one filter charge. The *E. coli* reduction profile of these samples versus pore volume is illustrated in figure 3.4. In this figure, the filter watered *E. coli* concentration/influent water *E. coli* concentration is shown for various pore volumes filtered corresponding to three time points during experiment #2: day 35, day 42 and during a day 43. The reduction of *E. coli* is greater in the water that remained in the pores of the filter overnight compared to water that passed through the filter later on the same day. The water that passed through the filter during the same day of the charge experienced lower *E. coli* reductions compared to water that stayed in the filter medium pores overnight. In addition, as the BSF ripened the reduction of *E. coli* improved over the entire filtration run; all portions of filtrate water had increased reductions of *E. coli* during day 42 as compared to day 35.

On the day where the biofilm was accidentally punctured (day 43), the extent of *E. coli* reduction is less for the water passing directly through the filter the day of filtration compared to water that stayed in the filter overnight or compared to the water that passed directly through from the previous sampling time point (day 43). This finding suggests that the mechanism for *E. coli* reduction was perhaps *E. coli* straining out by the biolayer (“schmutzdecke”) and associated antagonistic biological effects, which could not occur very much or at all after the biolayer was punctured.

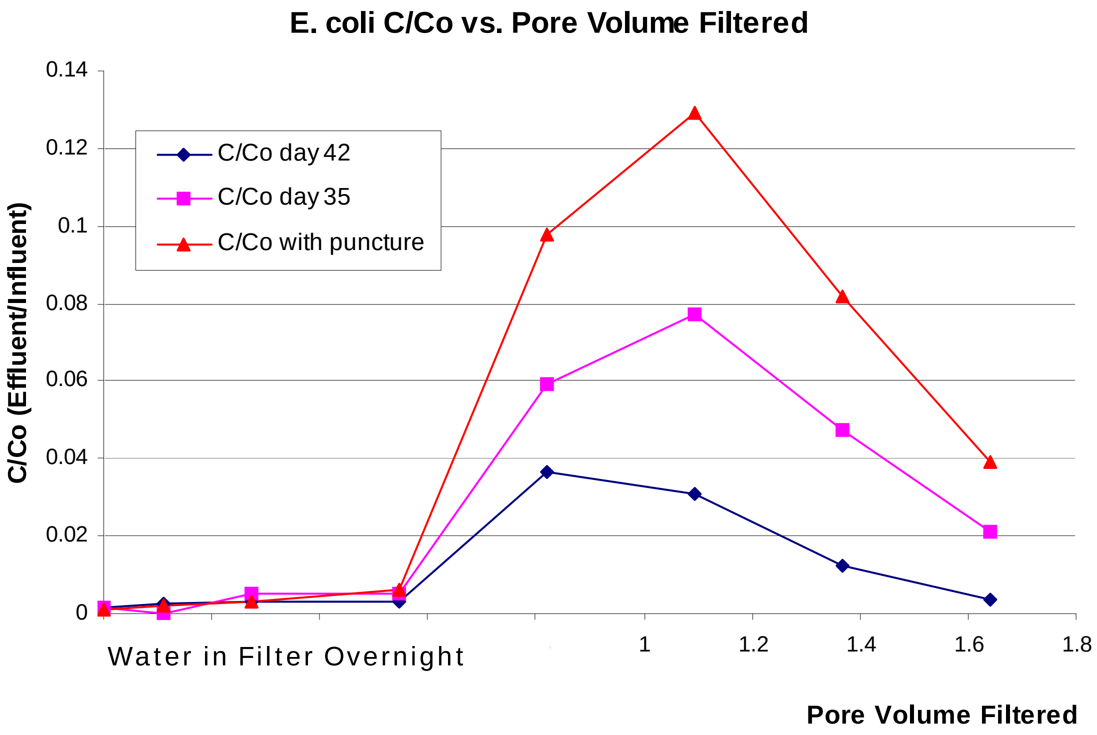


Figure 3.4 Effect of water volume filtered and overnight retention of water in the filter medium (filter medium pore volume) on E. coli reduction for different dosing days of experiment #2

3.4 Discussion

There was considerable difference in *E. coli* reductions by biosand filters in the laboratory, depending on the day of the filter run and the extent of filter ripening at that time. *E. coli* reductions ranged from 63% in the early days of the run when the filter was not ripe to 99% in the later days of the run when the filter was ripe in both experiments. Coliphage reductions also differed according to the day of the filter run and the extent of filter ripening. For PRD-1 initial removal was as low as 10% and maximum reduction was as high as 87% later in the run. A similar pattern was seen for MS-2; however the difference was not as great. Initial reduction of MS-2 was 43% and increased to 78% later. Overall, for both coliphages, reduction was less compared to that of *E. coli*. For both *E. coli* and PRD-1, reductions from water by BSF filtration were significantly greater after filter ripening compared to the unripe filter. This performance difference of the ripened and unripened filter was not as apparent for MS-2.

The changing results for *E. coli* and coliphage reductions as a function of filter operating time as well as the declines in filter flow rate over time suggest that some form of filter maturation was occurring over the period of filter use (figures 3.3, 3.4). The filter maturation could be both biological in the form of a biolayer and physical-chemical where additional particle deposition can enhance straining and particle capture. While it is likely that both of these mechanisms are at work, the experimental design did not distinguish between biological and physical-chemical ripening.

The observed reductions of bacteria and viruses of by the biosand filter are considerably less than those previously observed for a typical SSF (99+ %) (Hendricks, et al. 1991). Further studies of microbial reductions in relation to filter flow rate and the development of functional biological activity in the BSF are needed to more clearly determine the basis of the performance differences between a conventional SSF and the BSF, and the effects of ripening and flow rate on the performance of both types of filter in reducing microbes

The lower filtrate concentrations, and hence greater reductions of *E. coli*, MS-2 and PRD-1 in the first 15 L of filtered water from a 40 L daily dose, suggest the potential importance of water and microbe retention time in the filter bed in contributing to enhanced microbial reductions. Because the empirical pore volume of the filter is approximately 18 L, the initial 15 L of filtrate from a dosing after an overnight period of no filtrate flow are likely to have been residing in the filter during the period in between feed water dosing. Therefore, this initial 15 L of filtrate had the longest period of contact with the filter bed and the associated exposure to the biological activity within it. It is hypothesized that biological activity contributes to microbial reductions, as do additional time for microbial adsorption to and sedimentation near filter media. However, these physical, chemical and biological processes potentially contributing to microbial reductions by filtration deserve more investigation, as they appear to have important implications for filter use and management practices.

Conclusions

The results from laboratory experiments in which typical household biosand filters were dosed with 40 L of microbe-seeded water per day gave average *E. coli* reductions of 94% and average coliphage reductions of 62%. However, *E. coli* reductions ranged from a maximum 98-99% in ripened (biologically mature) filters to as low as 63% initially in unripened filters. Coliphage reductions also improved over time in the filter run but not as much as for *E. coli* bacteria. Further studies are needed to better determine the factors contributing to the changes in bacterial and viral reductions by biosand filters as they ripen and as flow rates decline with increasing time of filter use. Such studies could aid in identifying design features and operating conditions for optimized performance in reducing microbes in water. Such laboratory research can help inform the needs for research and demonstration of performance under field use conditions in order identify and implement best management practices for optimizing performance of filters used in the field

Chapter 4: Evaluation of the Biosand Filter in a Six-Month Field Trial in Bonao, Dominican Republic

4.1 Introduction

More than a billion people lack access to improved water supplies and many more lack access to microbiologically safe water. There is an increasing interest in improving water quality and access by utilizing household water treatment and safe storage at the point of use of the consumer. Recent evidence suggests that point of use drinking water treatment can improve the microbiological quality of drinking water and reduce diarrheal disease (Arnold & Colford, 2007; T. Clasen, Roberts et al., 2006; L. Fewtrell & Colford, 2004). The field evidence of successful performance by various interventions to treat water at point of use has also been paralleled by the recognition and development of a growing number of available technologies to treat drinking water in the home.

Filtration technologies to purify water at the point of use are among the available alternatives. Unlike chemical disinfection or solar disinfection, household filtration can not only improve the microbiological quality of the water, it can also improve the appearance and taste of the drinking water. The need for filtration technology is particularly important in areas where the accessible sources have high levels of turbidity. High levels of turbidity decrease the efficiency of both solar and chemical disinfection, and they make the water unappealing to the consumer.

One disadvantage of filtration technologies is the lack of a residual disinfectant and the potential for post-treatment contamination. It is due to the possibility of recontamination that there is an interest in combining both filtration and disinfection technologies to treat water at the point of use.

Currently, the household filtration system being used in developing countries that has been documented the most for performance is ceramic microfiltration. This technology has been characterized for its ability to improve microbiological water quality and reduce diarrheal disease (T. Clasen et al., 2005; T. F. Clasen et al., 2004). However, these filters treat relatively small volumes of water, typically 10 L per batch, and their filtration rates are relatively slow at about 1-3 L/hour. Other filtration technologies besides ceramic filters also are being used in developing countries, and some have the advantage of treating larger volumes of water at faster flow rates. One of these is the biosand filter (BSF), an intermittently operated slow sand filter that produces about 1L/min. It has been used in households around the world for more than ten years and recent estimates suggest that there are more than 80,000 BSFs globally serving more than 500,000 people (Duke et al., 2006).

The biosand filter is unique compared to many of the other currently available treatment technologies because laboratory evidence indicates that performance improves with use due to development of a biologically active surface later or schmutzdecke. (Buzunis, 1995; Stauber et al., 2006). Other technologies have been known to break over extended periods of use or to experience decreased effectiveness and decreased levels of usage after extended periods of time (Arnold & Colford, 2007). There have been no rigorous field studies to date documenting improved performance of newly installed filters over time in use or their sustainability. However, there is some evidence of continued use years after implementation. Duke and others reported 98.5% reduction of *E. coli* in 107 households in Haiti where the BSF had been implemented for more than two years (Duke et al., 2006).

The BSF is similar to a conventional slow sand filter (SSF) in that there is typically no pretreatment or backwashing and operation is simple, with gravity-driven rather than mechanical pressure filtration. As in conventional SSFs, the sand bed remains covered with water throughout operation. Upon start-up, a ripening process occurs, during which a biolayer (or schmutzdecke) forms, head loss increases and filtration performance improves. Unlike a SSF, the BSF does not operate continuously but instead, intermittently, with periodic charges of feed water (typically up to 20 L each) each day.

4.2 Objective

The main objective of the current research was to document reductions of *E. coli* and total coliforms from water by BSFs newly installed in approximately 75 households in two communities of Bonao, Dominican Republic. Performance of the filters was followed for a period of six months, with sampling at approximately two-week intervals. Information gathered in interviews during periodic household visits was used to determine if filter ripening occurred and what factors or conditions were related to improved filter performance. Multivariate linear regression models were developed to characterize filter performance. These results were also compared to those of other BSFs that had been installed in and around the study site for periods of four to nine months and to newly made BSFs operated in the UNC laboratory.

4.3 Methods

Household Selection

A cross-sectional study was performed from June to August 2005 in two communities in Bonao, DR. The households from this study were then asked to participate in the randomized controlled trial of the biosand filter. Requirements for inclusion in the study were: no BSF previously in household, at least one child under five years of age and willingness to participate. One week prior to randomization, all households were assigned a unique number and random numbers were generated using a random number generator program in Excel to identify the ~50% of the households that were selected to initially receive the BSF (the intervention). In February 2006, 81 households were selected to receive the BSF.

Filter installation and follow-up evaluations

During February 2, 2006 to February 8, 2006, 81 biosand filters were installed in the households that had been randomly selected to receive filters. All filters were made and installed by filter technician Jose Rivas of Dajabon, DR. Households received the BSF, a safe storage container and instruction on use of the BSF during filter installation. Initial filter flow rate was measured by filling the upper chamber of the filter and measuring the elapsed time to filter the first 1L of water. If the flow rate was outside of the range of 0.7 – 1.1 L/min., the filter sand was re-installed. Filter flow rate was outside of the range for approximately 10% of filters installed in February 2006. That 10% of filters were re-installed to achieve acceptable flow rates. Filters were visually inspected during weekly household visits and households were asked via structured questionnaire to report problems with the filter. Water samples were collected from households at 11 time points during a six month period after filter installation. Filter flow rates were measured three additional times after installation, specifically at 6, 17 and 23 weeks after installation.

Drinking Water Sampling and Analysis

At approximately two-week intervals, households were asked to provide samples of drinking water being used in the home. Staff collected approximately 500 mL water samples in sterile Whirlpak® bags. Households were asked to provide a sample of water prior to filtration (feed water), water directly from the BSF outlet and stored BSF-treated water, as well as any water receiving additional treatment after BSF treatment. All water samples were stored on ice and kept cool until processing, which was within 8 hours of collection. Water samples were analyzed for total coliforms and *E. coli* using the Colilert™ Quantitray 2000 system from IDEXX (Westbrook, Maine). Sample water pH, turbidity, and free and total chlorine were also analyzed using the following methods: pH with a Sension1 meter, turbidity with a turbidimeter 2100p and free and total chlorine by the colorimetric method using a pocket colorimeter II (all meters for pH, turbidity and chlorine analysis were provided by Hach, Loveland, CO).

Additional biosand filters that were not part of the RCT

In addition to the longitudinal sampling of RCT biosand filters that were sampled during the period of February to August 2006, an additional 106 biosand filters were sampled once during the period of March 2005 and June 2006 and analyzed for reduction of total coliforms and *E. coli*, turbidity, and for pH. All households in the additional sampling of BSF had received training in filter use from an implementing organization. These implementing organizations received training from the implementing organization that installed the BSFs from the RCT in the longitudinal portion of the study. The additional 106 filters were installed in five locations in the center of the Dominican Republic; near Bonao and the two communities of the longitudinal field study.

Data Collection and Analysis:

All data were entered into Excel or EpiInfo and imported into Stata 8.0 and GraphPad for analyses that could not be performed in Excel or EpiInfo. Log_{10} reduction of *E. coli* and total coliforms were calculated by the following formula:

$$\text{Log}_{10} \text{ reduction} = \text{Log}_{10} \text{ influent concentration} - \text{Log}_{10} \text{ filtered water concentration} \qquad (1)$$

Water quality analyses leading to $\log_{10}$ reduction calculations were done 11 times during the study: specifically at 1, 4, 6, 8, 10, 13, 16, 18, 20, 22, and 24 weeks after filter installation. Data were also collected to determine turbidity reduction, change in pH, filtration rate, and average frequency of filter use. These variables were evaluated for graphical presentation and also in a multivariate linear model in Stata 8.0. The main focus of the multivariate linear regression was to determine what variables were potential predictors of improved filter performance in reducing *E. coli* from water.

4.4 Results

Performance of RCT Filters over a Six-Month Period

Of the filters initially installed in February, 2006, a total of 75 remained and were accessible in August 2006. Six households stopped participating in the study: two because they did not want to participate; two because they were unavailable during scheduled household visits and two moved from the study area. A total of 671 sets of samples were analyzed to measure for reductions of *E. coli* and total coliforms by the filters. The number of complete sets of water samples from each household ranged from 2 to as many as 11. A sample set was considered complete if *E. coli* was measured both prior to and after BSF treatment.

In figure 4.1 and 4.2 are the histograms of $\log_{10}$ reduction of *E. coli* and total coliforms respectively. The ability of the biosand filter to reduce the concentration of *E. coli* and total coliforms based on $\log_{10}$ difference between influent and effluent concentration varied and appeared to be relatively normally distributed with the exception of the large portion of samples that were indicated to have no reduction (i.e., $\log_{10}$ reduction = 0). Of the ~25-30% of samples that demonstrated < 0.3 $\log_{10}$ reduction of E. coli, 107 of these samples had < 1/100mL *E. coli* in the water prior to filtration. The largest proportion of samples with *E. coli* $\log_{10}$ reductions < 0.30 occurred in the first two months of filter installation: February and March 2006.

In addition to a large proportion of the samples that demonstrated no reduction of *E. coli* or total coliforms, a proportion of samples showed negative reductions or an increase in concentration of *E. coli* or total coliforms. Negative reductions can be the result of fluctuations in source water quality, flushing of organisms from the filter and/or the sign of a poorly functioning filter. For example, initially, reductions of total coliforms were negative during the first week of filter sampling. This was likely due to the organisms being flushed out from the sand during the installation process. In addition, changes in water quality can result in seemingly negative reductions by the filters because the water that is within the sand pores is of a different quality that the water that was being poured into the filter during household visits.

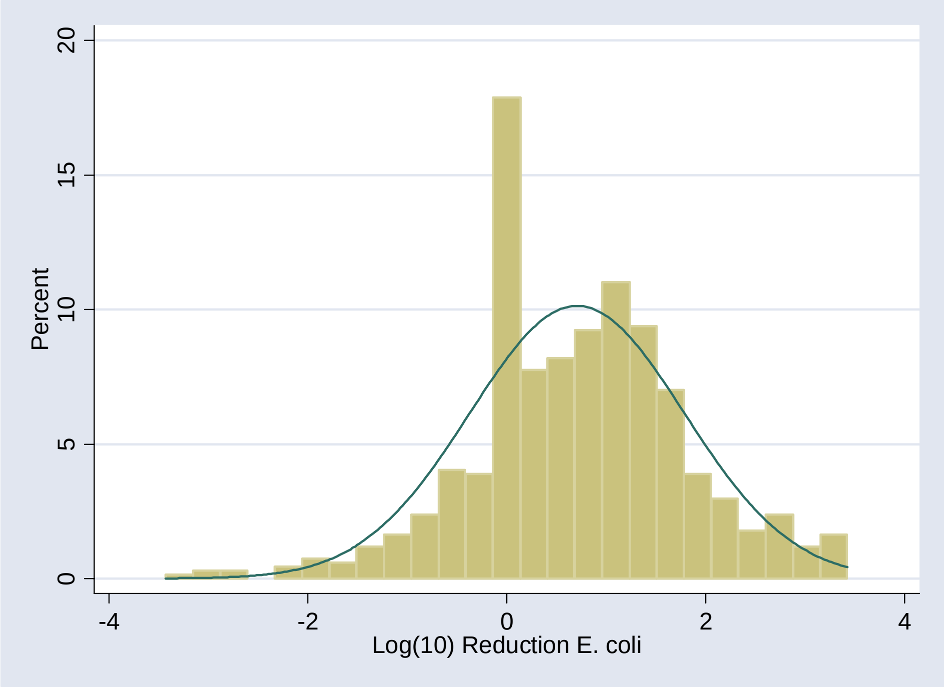


Figure 4.1 Histogram of log$_{10}$ reduction of E. coli during six- month study in Bonao, Dominican Republic

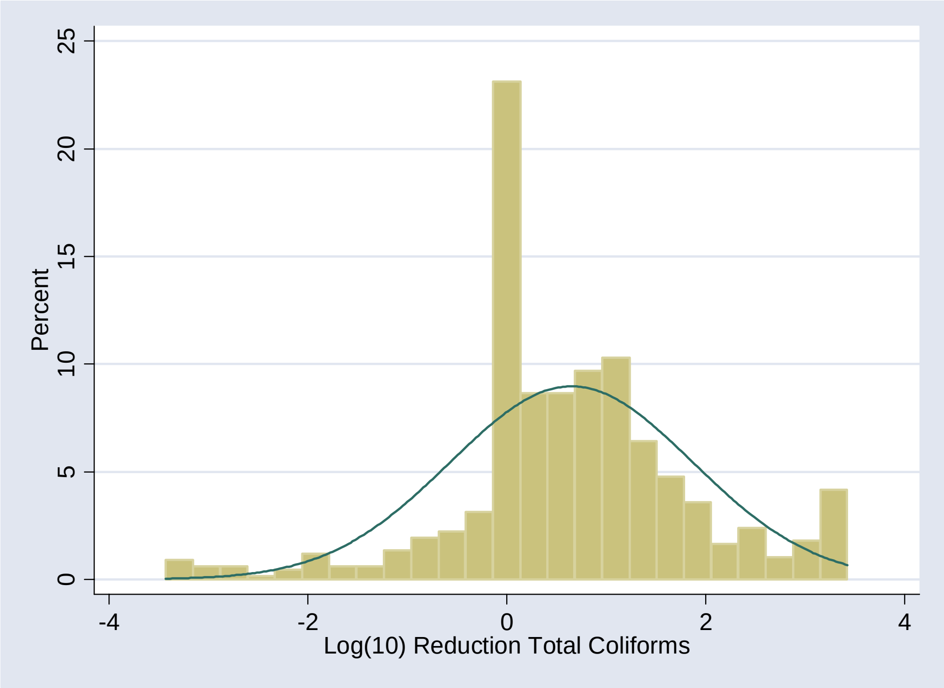


Figure 4.2 Histogram of log$_{10}$ reductions of total coliforms during six- month study in Bonao, Dominican Republic

Average reductions of *E. coli* and total coliforms were 0.71 $\log_{10}$ (80% reduction) and 0.65 $\log_{10}$ (78% reduction) respectively. As shown by the histograms, *E. coli* and total coliforms reductions varied widely from apparently negative, <0 $\log_{10}$, to as great as 3.4 $\log_{10}$ (>99.9% reduction). Geometric mean *E. coli* MPN/100mL was 25 and 5 for unfiltered and BSF-treated water, respectively. Average turbidity reduction was 11%, but this also ranged widely, from <0 to 98%. Average unfiltered water turbidity was 1.7 NTU while average BSF-treated water turbidity was 0.9 NTU, representing a 47% reduction when comparing average unfiltered and filtered water turbidities. Water pH changed during filtration, averaging pH 7.6 in samples prior to BSF treatment and average pH of filtered water was 7.8, representing a slight increase in pH.

Effect of Cumulative Filtration Time on BSF Performance

In order to evaluate the effect of cumulative filtration time in use on BSF performance, the $\log_{10}$ reductions of *E. coli,* total coliforms and percentage reductions of turbidity were examined at each of the 11 different days during filter operation. The average reductions at each sampling point for *E. coli*, total coliforms and turbidity are listed in table 4.1 Geometric mean reductions of *E. coli*, total coliforms and turbidity were the lowest one week after installation: 0.29 log10, -0.27 log10, and -28% respectively. The highest $\log_{10}$ reductions for *E. coli* and total coliforms were achieved in the 22nd week after installation, with 1.1 (92% reduction) and 0.98 (89% reduction) $\log_{10}$ reductions, respectively. The highest reduction in turbidity was 25% achieved in the 13th and 16th week after installation. Reductions for *E. coli* and total coliforms generally improved over time but fluctuated throughout. Turbidity fluctuated throughout the study period and also improved over time but to a lesser extent than that for the bacteria.

Table 4.1 Reductions of *E. coli*, total coliforms and turbidity for 11 sampling periods during six-month study in Bonao, Dominican Republic

Week after installation	**# of Observations**	**Geometric Mean Log_{10} Reduction of *E. coli***	**Geometric Mean Log_{10} Reduction of Total Coliforms**	**Average % removal NTU**
1	65	0.29	-0.27	-29
4	68	0.31	0.37	-12
6	65	0.93	1.0	8.3
8	63	1.0	0.91	18
10	61	0.68	0.39	-7.2
13	61	0.58	0.71	25
16	57	0.83	0.96	**25**
18	63	0.89	0.80	22
20	63	0.62	0.69	11
22	48	**1.1**	**0.98**	22
24	59	0.71	0.80	24
Total	**671**	**0.71**	**0.65**	**11**

There was considerable variation in bacterial and turbidity reductions among filters at each sampling point of the six month study. The variation in *E. coli* reductions and turbidity reductions for each sampling point are presented in figure 4.3 and in figure 4.4 respectively. Median $\log_{10}$ *E. coli* reduction increased during the first three sampling periods (initial six weeks after installation) but did not appear to further increase linearly thereafter but rather fluctuated up and down. The range of *E. coli* reduction spanned two $\log_{10}$ or more for every sampling point; suggesting great variations in performance among filters. While not shown, total coliform reductions demonstrated a similar pattern where initially low $\log_{10}$ reductions improved in the early weeks but then fluctuated and gave a wide range of $\log_{10}$ reductions among individual filters.

The range of turbidity reductions also spanned many orders of magnitude. Turbidity reductions appeared to increase over time but due to the large variation, the linear regression does not fit well to the data. Turbidity reductions did improve over time yet were highly influenced by the unfiltered water turbidity. Average unfiltered water turbidity was low < 5NTU. While unfiltered water turbidity was low, average filtered water turbidity was < 1 NTU. Initial poor turbidity removals are also likely to be the result of flushing of fine particles from the prepared sand media.

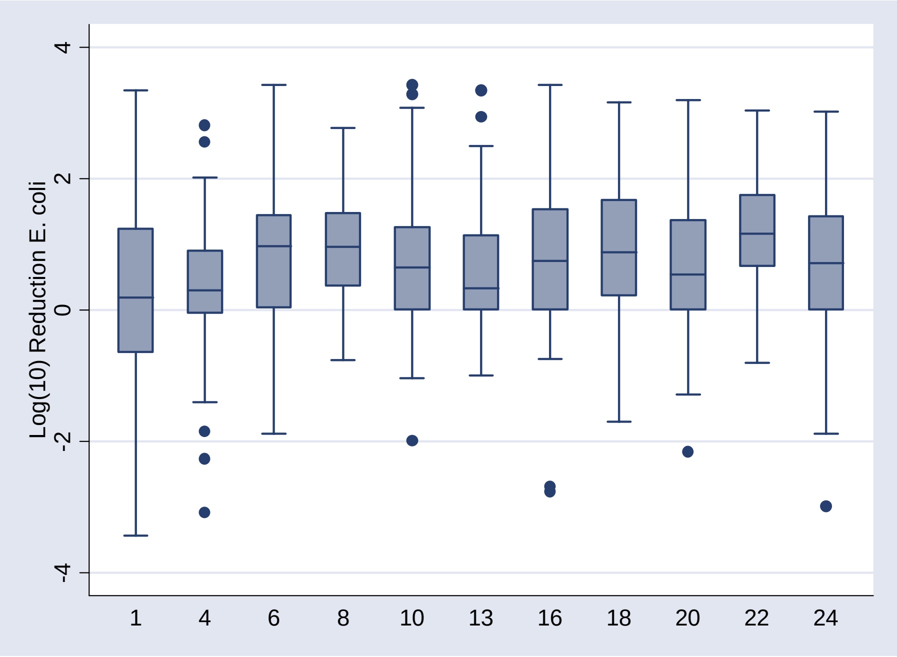


Figure 4.3 Box plots of $\log_{10}$ *reductions of E. coli over 24 weeks of study*

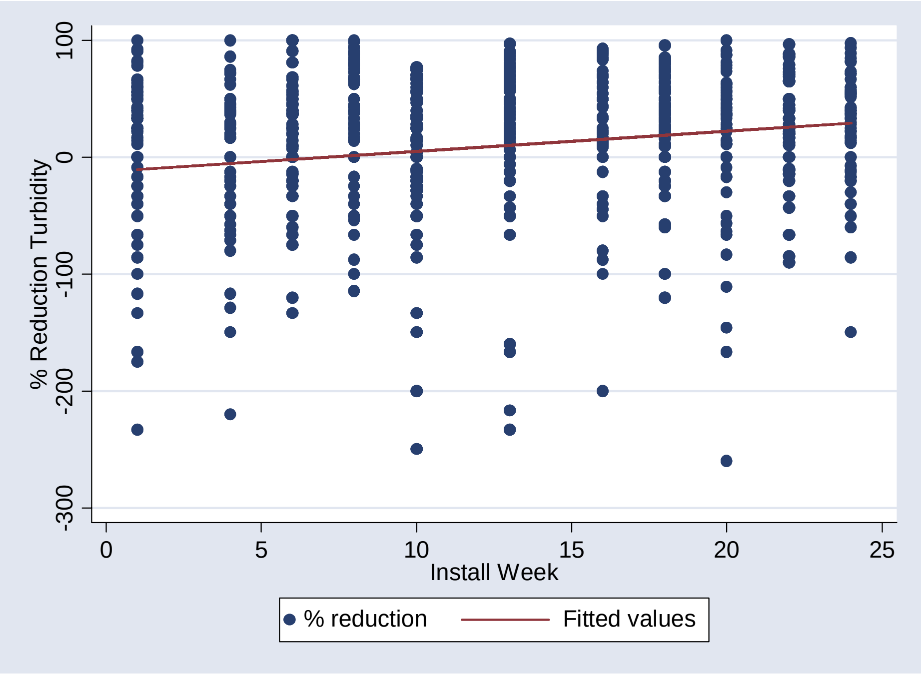


Figure 4.4 Turbidity reductions in BSFs over 24 week sampling period
Solid circles represent turbidity reductions and the dashed line is the fitted linear regression.

In addition to parameters measured in water prior to and after BSF treatment, filters were also tested for flow rates and their changes over time. Average flow rate at installation was 0.94 L/min and after 23 weeks of operation, average flow rate was 0.83 L/min; a statistically significant difference ($p < 0.001$ by two sample t-test). The fractional change in filter flow rate was expressed as flow rate at time t (Q_t) divided by the initial flow rate (I) or Q_t/Q_i. The fractional change in filter flow rate was an average of 0.91, but ranged from a low of 0.11 to a high of 1.2, for individual filters. In addition to physical, chemical and biological performance measurements, weekly data were collected on household BSF practices, including filter use, based on estimated number of times of use per week. Average household filter use was 4 times each week, but ranged from once a week to seven times a week (or daily). These responses on filter use were averaged for each household and used in later analysis of BSF performance.

Flow rate measurements over time demonstrated a decrease compared to the initially measured rate. A box plot of filtrate flow rate at the four time periods it was measured is presented if figure 4.5. As shown in the figure, flow rate decreased over time, especially by week 23. Flow rate varied among filters at each sample time and the extent of variation appeared to increase over the 6-month period of filter operation, as indicated by the increased span of plot whiskers. This increased variation was caused by higher flow rate reductions in a portion of the BSFs compared to others in the study. A few BSFs were documented to have higher flow rates as compared to initial rates which could be due to possible initial flushing of fines and an increase in flow rate as compared to initial flow rate. However, many BSFs experienced considerable flow rate reduction, to as low as 1/10th the original flow rate. While some filters demonstrated decreased filter flow rates, the reduction in flow rate was minimal compared to reductions seen in laboratory studies.

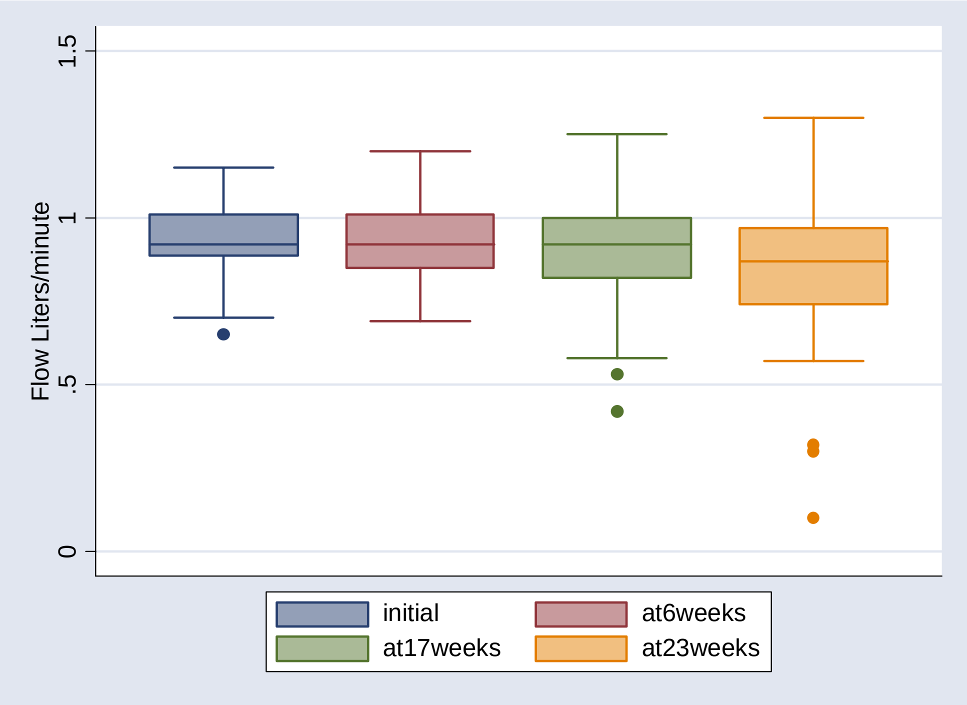


Figure 4.5 Flow rate of BSFs over time during six-month field trial in Bonao, Dominican Republic

To evaluate whether or not $\log_{10}$ *E. coli* reductions increased over time, a linear regression model was created examining the effect of time (measured in weeks after installation) on the $\log_{10}$ reductions of *E. coli*. The linear regression suggested that there is a slight increase in *E. coli* reductions with increasing time after installation. The coefficient from the linear regression for weeks after installation was 0.016 ($\log_{10}$ reduction/week). This suggests a 0.016 log10 increase in *E. coli* reduction per week after installation. Based on the ANOVA analysis, this value was considered a significant predictor ($p = 0.004$) of $\log_{10}$ reduction values; however the r^2 value was only 0.10. A graph of *E. coli* $\log_{10}$ reduction values over the sampling weeks (as dots) for the 11 observation periods and showing the linear regression prediction (as a dashed line) is illustrated in figure 4.6. Because the actual values at each sampling time vary greatly and the r^2 value of the regression line is small, the effect of time since installation on filter flow rate is modest when examined in this way for individual observations for all of the filters at once.

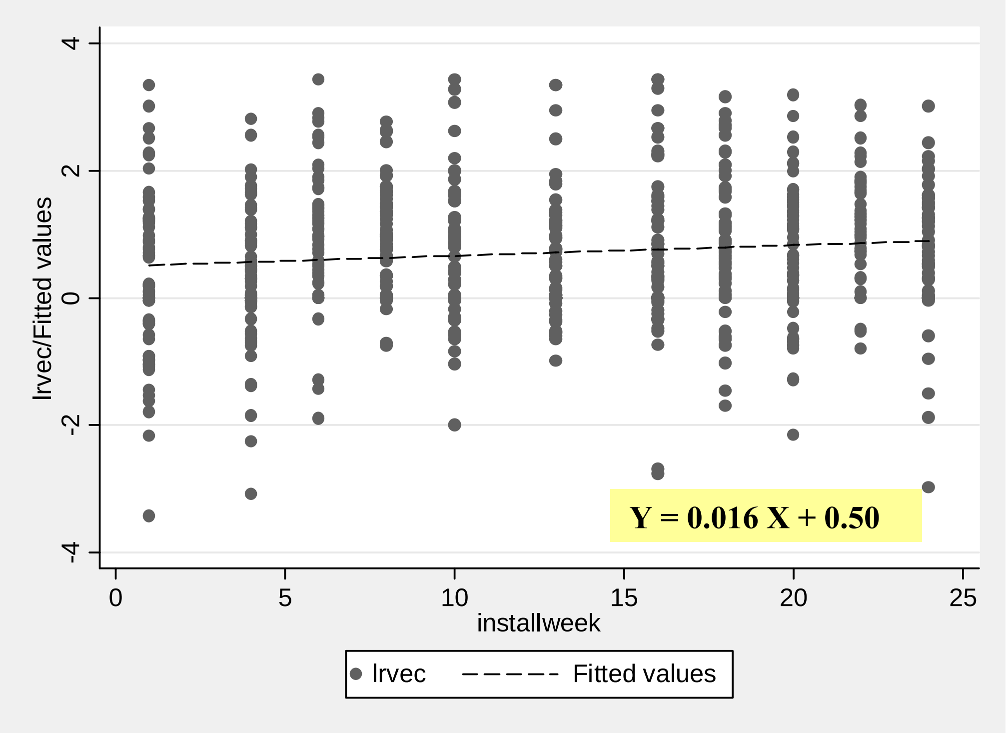


Figure 4.6 Graph of values of $\log_{10}$ reduction E. coli and linear fit to data

Univariate and Multivariate Regression Models

In order to further explore what factors play a significant role in biosand filter performance, a number of variables were examined both graphically and through linear regression analyses to determine the ability of the variable to predict filter performance where filter performance is based on reduction in *E. coli* concentrations. The variables that were included in the analysis were: influent levels of *E. coli*, turbidity reduction, flow rate reduction, and average frequency of use (or doses per week).

Influent concentrations of *E. coli*

Influent concentrations of *E. coli* were measured for each weekly set of $\log_{10}$ reductions of *E. coli*. To evaluate the relationship between, $\log_{10}$ influent concentrations of *E. coli* and $\log_{10}$ reductions of *E. coli*, a linear regression model was fitted (figure 4.7). There are three important boundaries to note on figure 4.7. One is the line indicating the detection limit of $\log_{10}$ *E. coli* reduction, as the top boundary line of the scattered dots representing individual $\log_{10}$ *E. coli* reductions of filters. This line results from the lower detection limit of the *E. coli* assay when no *E. coli* were detected in a sample and the resulting calculation of *E. coli* reduction based on the difference between *E. coli* in the feed water and the BSF filtrate is a greater than the value. The upper and lower boundaries of the vertical data points for $\log_{10}$ *E. coli* reduction capture the detection limit of the *E. coli* assay. The upper detection limit was 2419.6 *E. coli*/100mL (3.4 $\log_{10}$) and the lower detection limit was 1 *E. coli*/100mL (0 $\log_{10}$).

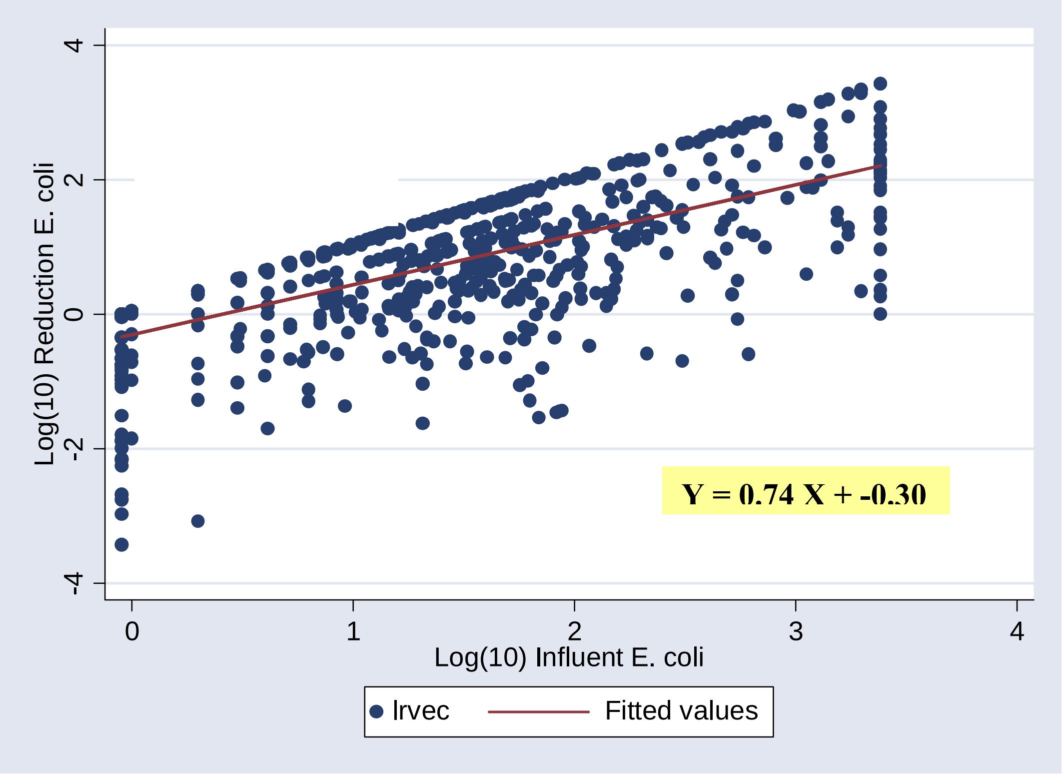


Figure 4.7 Scatter plot of $\log_{10}$ influent E. coli/100 mL and $\log_{10}$ reduction of E. coli (LRVEC) and fitted values.

The results of these analyses as shown in figure 4.7 suggest that there is a linear relationship between $\log_{10}$ influent concentrations of *E. coli* and $\log_{10}$ reductions of *E. coli*. From the ANOVA, the coefficient for linear relationship between $\log_{10}$ influent *E. coli* and $\log_{10}$ reductions of *E. coli* was 0.74, ($p < 0.001$), $r^2 = 0.48$. The ANOVA analysis suggests that for every unit change in $\log_{10}$ *E. coli* influent there is a 0.73 $\log_{10}$ increase in reductions of *E. coli*.

Effect of other variables on $\log_{10}$ reductions of *E. coli*

Similar to the graphical analysis and linear regression analyses performed for the relationship between weeks after installation and influent concentrations of *E. coli*, three other variables were analyzed as predictors of $\log_{10}$ *E. coli* reduction: change in filter flow rate (measured as Qt/Qi), frequency of weekly filtration (as number of filter doses per week) and reductions in turbidity. The coefficients for each variable in the univariate linear regression and the coefficients for the multivariate linear regression with all of the variables included are listed in table 4.2. In addition, the p-value for each coefficient is listed. The p-value indicates whether or not the coefficient is significantly different from zero and indicates whether or not that variable helps to predict the outcome. However, it is possible to have a p-value that indicates the coefficient is significantly different from zero and have a relatively low r-squared value. Therefore, r^2 values were also included in the table.

Initially, all 671 observations for *E. coli* $\log_{10}$ reductions were included and analyzed to determine the relationship between this reduction and flow rate reduction, frequency of use and % reduction in turbidity. In the univariate analysis of the independent variables mentioned above, weeks after filter installation, $\log_{10}$ influent concentration of *E. coli*, and %

reduction in turbidity appeared to be significant predictors of $\log_{10}$ reduction values of *E. coli*. However, when all of these variables were fitted in the multivariate model, average filtration frequency (weekly filter doses) and $\log_{10}$ influent *E. coli* concentrations were the only variables which remain as significant predictors of $\log_{10}$ E. coli reductions (based on an a priori $p < 0.05$ cutoff). The relationship suggests a 0.05 decrease in $\log_{10}$ reduction values as frequency of filtration increases and 0.76 increase in $\log_{10}$ *E. coli* reductions for every unit change in $\log_{10}$ influent concentrations of *E. coli*.

Table 4.2 Linear regression coefficients for independent variables as predictors of $\log_{10}$ *E. coli* reductions during a six-month study of installed BSFs in two communities of Bonao, Dominican Republic

Variable	**Univariate linear regression coefficient (p value, r^2)**	**Multivariate linear regression coefficient (p value, r^2)**
Weeks after installation	0.016 (0.004, 0.013)	0.0057 (0.177, 0.50*)
Log_{10} influent *E. coli*	0.74 (< 0.001, 0.48)	0.77 (<0.001, 0.50)
Fraction of initial flow rate	-0.13 (0.561, <0.01)	0.33 (0.054, 0.50)
Average weekly filtration frequency	0.032 (0.179, <0.01)	-0.05 (0.005, 0.50)
% Reduction in turbidity	0.0018 (0.001, 0.017)	0.000 (0.957, 0.50)

* - Multivariate model r^2 is the model fit when all are included in the model and not measured for each variable individually

Effect of independent variables on: final $\log_{10}$ reductions at 24-weeks after installation and average $\log_{10}$ reduction values for each filter

Because $\log_{10}$ reductions of *E. coli* fluctuated throughout the study, the relationship between filter performance and the variables of interest was evaluated at two additional measures of filter performance: during the last week of observation (24 weeks after BSF installation) and for average $\log_{10}$ performance of each filter. Linear regression analysis was done for each variable individually as well as in a multivariate analysis that included all of the variables of interest for each performance measure. The results are presented in tables 4.3 and 4.4 for the last week and the average performance respectively.

Table 4.3 Linear regression coefficients for independent variables as predictors of $\log_{10}$ *E. coli* reductions at the final sampling time of the study, 24 weeks after installation of BSFs in two communities of Bonao, Dominican Republic

Variable	**Univariate linear regression coefficient (p value, r^2)**	**Multivariate linear regression coefficient (p value,r^2)**
$\log_{10}$ influent *E. coli*	0.74 (< 0.001, 0.49)	0.70 (<0.001, 0.49)
Fraction of initial flow rate	0.14 (0.85, <0.01)	0.49 (0.420, 0.49)
Average weekly filtration frequency	0.16 (0.036, 0.08)	0.086 (0.15, 0.49)
% Reduction in turbidity	-0.0022 (0.46, <0.01)	-0.0027 (0.22, 0.49)

Table 4.4 Linear regression coefficients for independent variables as predictors of average $\log_{10}$ *E. coli* reductions for each BSF from six-month study in two communities in Bonao, Dominican Republic

Variable	**Univariate linear regression coefficient (p value, r2)**	**Multivariate linear regression coefficient (p value)**
Average influent $\log_{10}$ *E. coli*	0.61 (< 0.001, 0.52)	0.58 (<0.001, 0.56)
Fraction of initial flow rate	-0.014 (0.96,<0.01)	0.05 (0.784, 0.56)
Average weekly filtration frequency	<0.000 (0.999, <0.01)	-0.057 (0.024, 0.56)
Average turbidity reduction	0.0018 (0.21, 0.02)	0.0005 (0.60, 0.56)

Based on the results presented in table 4.3, influent concentration of *E. coli* is the only significant predictor from the multivariate model of $\log_{10}$ *E. coli* reduction as the final (24-week) measurement of performance. When modeled individually, weekly filtration frequency also was a significant predictor of the final performance measurement of $\log_{10}$ *E. coli* reduction; however, it was not a significant predictor ($p< 0.05$) when other variables were included in the multivariate model.

When average performance in $\log_{10}$ *E. coli* reduction of each BSF was determined and modeled with the variables of interest, average influent *E. coli* was the most significant predictor of performance. When all of the variables were included in the multivariate model, average weekly filtration rate was also determined to be a significant predictor of filter performance based on $\log_{10}$ *E. coli* reduction. This model suggests that there is a 0.58 increase in $\log_{10}$ *E. coli* reductions for every one unit change in average influent *E. coli* concentrations. It also suggests that there is a 0.06 $\log_{10}$ decrease in average $\log_{10}$ *E. coli* reductions for each 1 unit increase in average weekly filtration frequency (filter doses per week). No other variables were found to be significant predictors of how each individual BSF performed on average to reduce *E. coli*. Overall, the univariate and multivariate linear regression analyses suggested that the most important predictor of performance of the biosand filter expressed as $\log_{10}$ *E. coli* reduction is the influent concentration of *E. coli*.

Average performance by month, flow rate and frequency

The relationship between BSF performance as $\log_{10}$ *E. coli* reductions and either flow rate reduction, time and frequency of use was further evaluated. In an effort to reduce some of the variability in the measurements from all of the individual filters, an average monthly $\log_{10}$ *E. coli* reduction was calculated for each month; essentially distilling the 671 individual observations into 6 observations. Linear regression was performed on the six average values. A graph of the monthly average $\log_{10}$ reduction of *E. coli* is plotted in figure 4.8. While the monthly *E. coli* $\log_{10}$ reduction varies, as shown by the errors bars representing standard deviations, there appears to be a positive linear relationship with $\log_{10}$ reduction values and months of operation of the BSFs. The linear regression suggests that for every month of BSF use, there is a 0.09 unit increase in *E. coli* $\log_{10}$ reductions and the r^2 value suggests relatively good fit of the regression to the average monthly $\log_{10}$ *E. coli* reductions. The results show a similar relationship between time after filter installation and reductions of *E. coli* as shown in figure 4.6. However, the fit is better when all of the values were averaged and modeled.

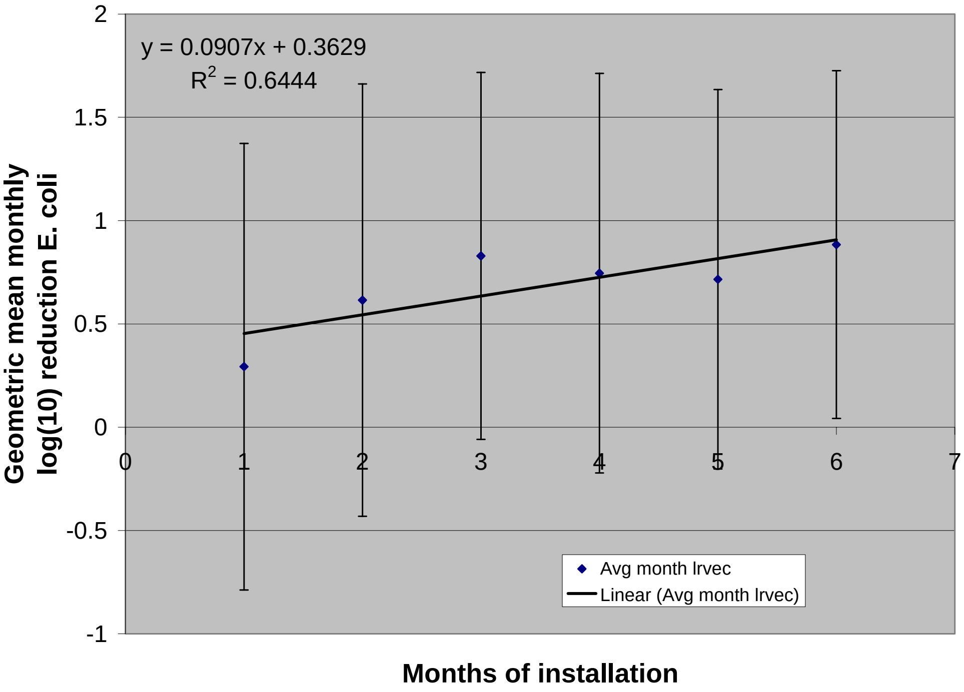


Figure 4.8 Graph of average monthly $\log_{10}$ E. coli reduction over time

The relationship between filter flow rate reduction and average $\log_{10}$ *E. coli* reductions was subjected to regression analysis for each month in which flow rate was measured: February, March, June and July. Average $\log_{10}$ reductions for each month were plotted against average filtration fraction Qt/Qi (flow rate at time t/ initial flow rate) (figure 4.9). The results suggest that as the fraction of initial flow rate decreased, the average $\log_{10}$ *E. coli* reduction increased. While the errors bars demonstrate considerable variability, the r^2 value of 0.66 suggests a relatively strong association with increased *E. coli* reductions as the fraction of flow rate decreased (or flow rate declined compared to initial flow rate).

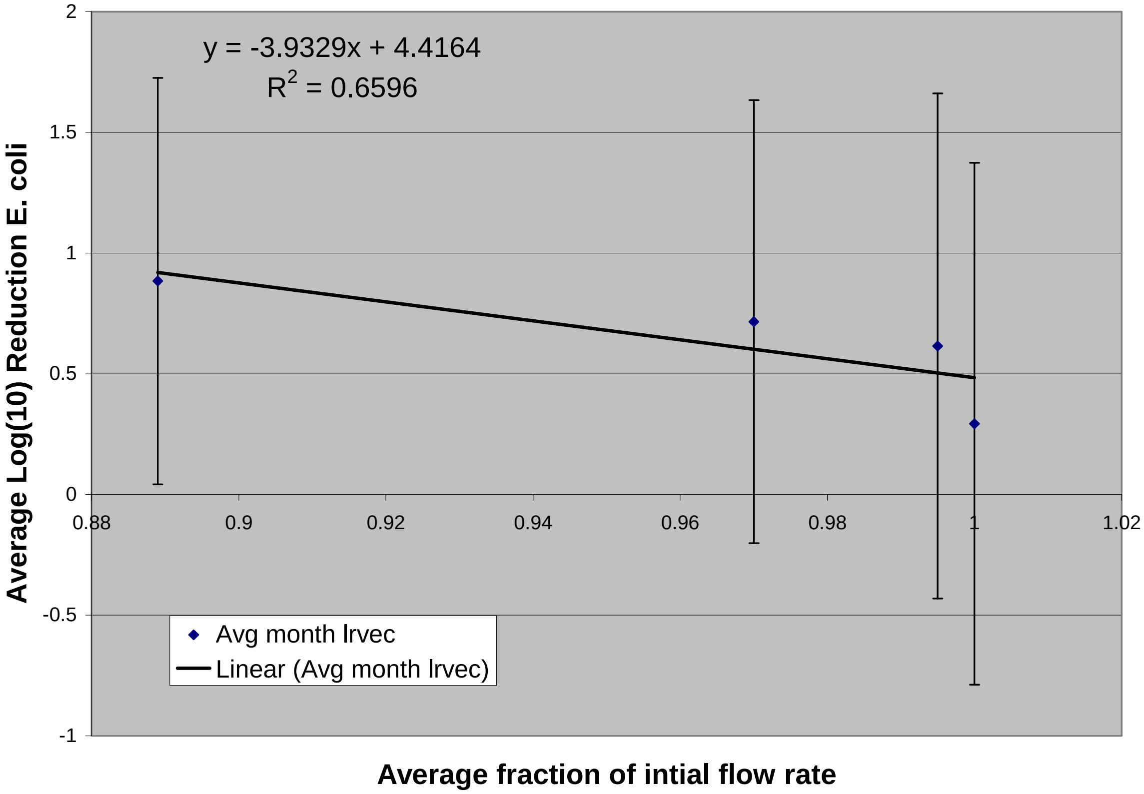


Figure 4.9 Effect of average reduction in flow rate on average log_{10} *E. coli reduction*

Average frequency of weekly filter use (as amount of times water was dosed) was made into a categorical variable where the average values were rounded to integers (0-7). Average $\log_{10}$ *E. coli* reductions were calculated for each unit frequency of weekly filtration and subjected to regression analysis, which is graphed in figure 4.10. There is a slight increase in $\log_{10}$ *E. coli* reduction as weekly filtration frequency increases. However, the r^2 value is low (0.29) suggesting only a weak positive relationship. This is weak association is also seen in the multivariate regression model of the relationship between average $\log_{10}$ *E. coli* reductions, average influent $\log_{10}$ *E. coli* concentration and water dosing frequency. However that model gave a negative relationship between these two variables instead of a positive one.

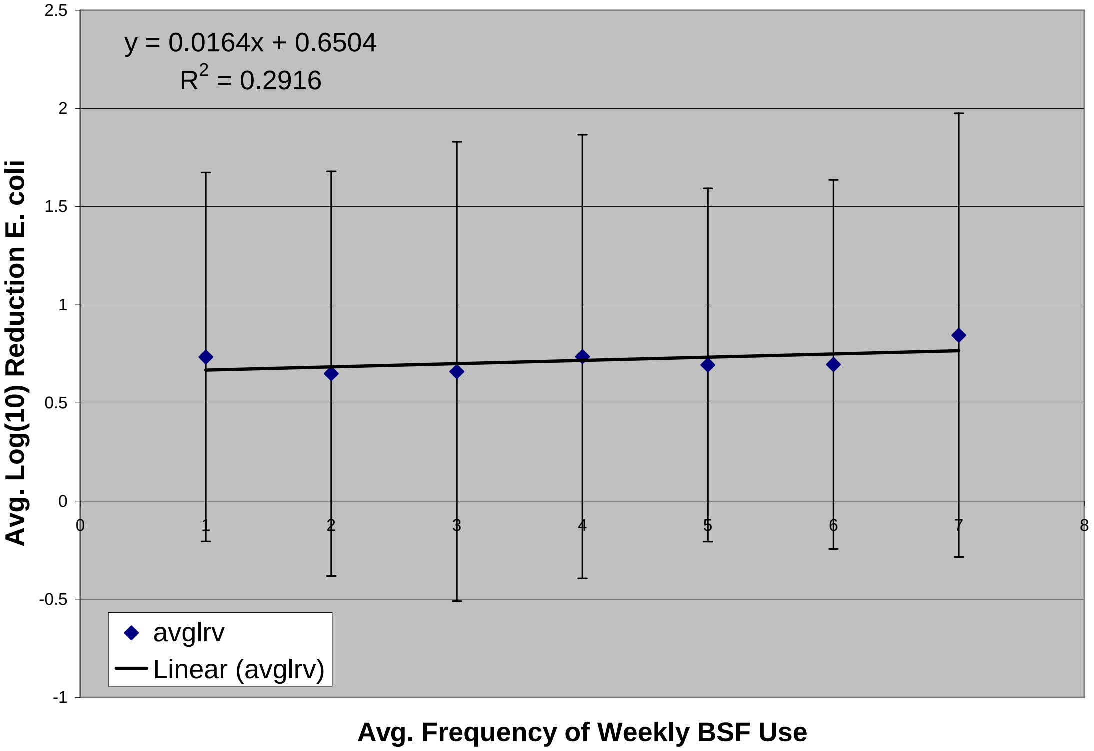


Figure 4.10 Graph of average frequency of use versus average log $_{10}$ reduction of E. coli for each frequency

Performance of 106 additional BSFs in the Dominican Republic

A total of 106 other households receiving BSF within the last year were visited to sample filter feed and filtered water. Filters had been installed for at least four months but no longer than 9 months. Turbidity reductions averaged 38% and ranged from <0 to 99%. Additional information on these 106 filters is presented in table 7.5. Average $\log_{10}$ *E. coli* reduction was 1.2 $\log_{10}$ and ranged from <0 to greater than 3.1 $\log_{10}$.

Table 4.5 Performance of BSF in 106 households (not involved in longitudinal study) located in the central region of Dominican Republic

Location	**# of samples**	**# of months installed**	**Average $\log_{10}$ Reduction *E. coli* (%)**	**Implementing organization**
Caño piedra/J.Central	20	9	0.9 (87)	Local health professional
Sonador	17	8	1.0 (90)	Peace Corps
Calle las Piedras	19	7	1.6 (97)	Peace Corps
Ingenio	20	4	1.2 (94)	Rotary Club Bonao
Aguacate	30	4	1.4 (96)	Arco la Cabirma
Total	**106**	**4-9**	**1.2 (94)**	**4 organizations**

4.5 Discussion

BSF Performance for Reduction of *E. coli,* Total Coliforms, Turbidity

Compared to BSF performance in field and lab

Microbial performance for the 75 BSFs during the 6-month longitudinal study was low, with an average 78-80% reduction of total coliforms and *E. coli.* Average influent *E. coli* levels in untreated drinking water also were relatively low, potentially influencing the ability to adequately quantify filter performance based on microbial reductions. The ability to quantify $\log_{10}$ *E. coli* reductions in the field can be limited by the lack of detection of *E. coli* (a non-detect) in a water sample. Such censored values prevent the calculation of a $\log_{10}$ reduction based on the difference in detectable levels of *E. coli* in filter feed and filtrate water samples. This can result in values for $\log_{10}$ reductions that are greater than what was documented. For example, if the influent concentration of *E. coli* was 10 MPN/100mL and the detection limit is 1/100mL, only a 1.0 $\log_{10}$ *E. coli* reduction can be measured. If zero *E. coli* were detected in the filtrate water, the $\log_{10}$ reduction in this example would be reported as >1 $\log_{10}$. Therefore, measurable but low concentrations of *E. coli* in unfiltered water and zero values in the filtered water, giving a lower detection limit value of <1/100 mL could contribute to an underestimation of the ability of the filter to reduce *E. coli* from the drinking water.

Average concentrations of total coliforms were ten fold higher than for *E. coli*. However, average reductions for total coliforms were even lower than for *E. coli*. This would indicate the ability to reduce total coliform concentrations was not likely underestimated by the detection limit of the assay or low concentrations of total coliforms in the unfiltered water. The observed low coliform reductions by the BSF are more likely the result of total coliform growth and survival inside the filter or on the treated water outlet tube. Average ambient temperatures ranged from 18-33℃, which are in the ideal temperature for total coliform growth. Total coliform survival and growth in drinking water distribution systems has been documented to be significantly influenced by various factors including temperature. In their study on coliform re-growth in distributions systems in a samples of systems in the US, the researchers found significant increases in both density and occurrence of coliforms when temperatures exceeded 15℃ (LeChevallier, Welch, & Smith, 1996).

Average turbidity reductions by the tested BSFs were also relatively low. This is probably influenced by the negative turbidity removals experienced in the early weeks after BSF installation. Households were instructed to filter 60 liters a day for three days following BSF installation as a step to flush out fine particles still present in the gravel and sand following initial installation. The negative removals in the initial weeks of BSF sampling are most likely due to inadequate flushing of these particles. However, after this initial period of likely fine particle flushing, turbidity reduction was still only approximately 20%, which is still low for a filtration process. The low turbidity reduction may be the result of relatively low average influent turbidities of <5 NTU. Average BSF treated water turbidity was under the US EPA standard of 1 NTU.

The relatively good quality of the unfiltered feed water and detection limit values for *E. coli* in filtered water probably contributed to modest $\log_{10}$ *E. coli* reductions possibly underestimating the ability of the BSF to reduce pathogens in water. However, average *E. coli* reductions of 80% suggest poor to moderate performance of the BSFs studied when compared to other BSF field studies. In a cross-sectional survey of households in Haiti, researchers found an average 98.5% reduction of *E. coli* in 107 BSFs (Duke et al., 2006). In the other 106 BSFs sampled in our study in the Dominican Republic, the performance for *E. coli* reduction was better compared to the 75 BSFs of the longitudinal study, with an average reduction of 94%. In both of these field studies, the higher average reductions of *E. coli* were documented for BSFs that had been installed well before sampling: at least four months for the DR BSFs and an average 2.5 years for the BSF in the Haiti study. In another study in Haiti, researchers installed and documented performance for BSFs in 80 households for a period of 3 months. They found average *E. coli* reductions by newly installed BSFs to be 76%; a value much lower than the average *E. coli* reduction for the filters that had been installed for an average 2.5 years (CAWST, 2006).

The limited bacterial reduction efficiency for newly installed BSFs is consistent with the expected ripening phase that filters go though to become biologically active. This ripening phase has been well documented in laboratory studies of the BSF. While laboratory studies report average *E. coli* reductions >95%, the filters demonstrate relatively low reductions initially when newly installed (Elliott et al., 2006; Stauber et al., 2006). Laboratory studies have shown *E. coli* reductions improving with time as the filters ripen and flow rates decline. It is also possible that the ripening process observed in the laboratory was accelerated by the use of a feed water of consistent poor quality as compared to the generally higher quality water in the communities in our study in Bonao. Additionally, while filter ripening in the laboratories studies was accompanied by an appreciable reduction in flow rate within 8 weeks, the filters in the 6-month field study did not experience such a decline in flow rate.

The relatively low average bacterial reductions by the newly installed BSFs in the DR and their limited decrease in flow rate over time suggests that these BSFs were still improving and continuing to ripen with use. The results suggest that the development of a biological layer did not occur as rapidly in the field study. The period of ripening of the filters for these waters is much longer than anticipated based on previous laboratory studies where filters can develop a biological layer in a period of six to eight weeks. The slow development of the biological layer suggests that microbial reduction efficiency will not improve and reach higher levels as would be indicated by controlled laboratory studies. Additional studies evaluating filter ripening and flow rate reduction can and should be completed on the filters in Bonao to more accurately understand how filter ripening happens in the field under these field conditions.

Fractional change in filter flow rate (Q_t/Q_i) was another important variable to consider when assessing performance of the filters in the field. When average fractional change was measured, there was found to be an improvement in filter performance as Q_t/Q_i decreased. For some filters, the Q_t/Q_i was as low as 0.1, but on average it remained at 0.8-0.9.

Factors Related to Filter Performance

Based on factors that appeared to influence microbial performance of BSFs in previous laboratory studies, a variety of water quality and BSF use parameters were measured and analyzed to determine the extent to which they were significant predictors of performance for reductions of *E. coli* in water in household use in the field. Perhaps not surprisingly, the most significant predictor of $\log_{10}$ reductions of *E. coli* was influent concentration of *E. coli*. There is a 0.60 – 0.70 increase in *E. coli* $\log_{10}$ reduction with each unit increase in $\log_{10}$ influent *E. coli* concentration. This observation suggests that BSFs will perform better when challenged with water having higher concentrations of *E. coli*. The results also suggest that perhaps performance based on $\log_{10}$ reductions may not be the most adequate measurement of filter performance in field studies; especially when water quality influent concentrations vary significantly. While the filters are not that effective in removing very low concentrations of *E. coli,* on average only low (5 MPN/100 mL) levels of *E. coli* remain in the filtered water. This suggests that perhaps filtered water quality may be a better indicator of filter performance for field sampling of the BSFs. The $\log_{10}$ reduction of *E. coli* and probably any other microbe in water is also influenced by the lower detection limit of the assay, as samples negative for the target microbe result in greater than (>) values for $\log_{10}$ reductions. However, when controlled for the lower detection limit, the influent concentration of *E. coli* remained the most significant predictor of BSF performance based on $\log_{10}$ reduction.

In addition to average *E. coli* concentrations in feed water, average frequency of weekly BSF use (a measure of volume of water dosed per unit of time) was an important factor influencing $\log_{10}$ reduction of *E. coli*. As filter use frequency increased, filters performance in $\log_{10}$ *E. coli* reduction decreased slightly. This phenomenon may due to the time the dosed volumes of water spend inside the BSF pore spaces, where parcels of water remaining in the filter pore spaces overnight have greater microbial reductions as filtrate water, apparently because they receive increased treatment. However, when the factor of filter dosing frequency was analyzed based on average filter dosing frequency and average $\log_{10}$ *E. coli* reductions, an opposite relationship was suggested. The BSFs provided slightly improved water based on $\log_{10}$ *E. coli* reductions when dosed more frequently (when averaged for each filter). These results seem to be conflicting and suggest that further investigation of the relationship in the laboratory and the field is needed. Filtration frequency is an important operational and management practice and more knowledge about it could be used to inform better use and practice in the field. In most of the laboratory studies, BSFs were dosed daily and frequency of use was not measured in terms of its ability to improve or reduce filter performance. The results from the field suggest that too many other variables were acting together to isolate the effect of filtration frequency and that this a topic best investigated in the laboratory at first.

No other factors were found to be significant predictors of filter performance on the basis of $\log_{10}$ *E. coli* reductions when all observations for the BSFs are analyzed for individual performance. However, two factors did seem to be potentially good indicators of average BSF performance when all of the data were averaged on the basis of $\log_{10}$ *E. coli* reductions for all filters studied: time in use and reduction in flow rate. Although wide variation in performance was observed, BSF performance improved over time in use and as filter flow rates decreased (compared to initial flow rates). This is the first study to document this effect in a longitudinal field study and the findings suggest that BSFs will likely improve in performance with time in use as they ripen or mature. Hence, unlike other filters, such as membranes that decline in performance over time and with increased use, BSFs are likely to maintain and even improve in performance over time and thereby be a sustainable technology. Some biosand filters installed in the field in the early 1990s in Latin America are still in use and apparently still performing effectively.

As demonstrated in laboratory studies, a reduction in BSF flow rate is an indication of filter ripening. The reduction in flow rate observed in these filters in household use in Bonao also suggests some form of filter ripening. However, this process did not seem to be as rapid as filter ripening observed in laboratory studies. The reasons for this difference are unknown and were not specifically investigated in the field, but they could be due to differences in water quality. Evidence from laboratory studies of BSFs dosed daily with constant amounts of water of about the same quality indicate that ripening occurred more rapidly and filter flow rate declined more rapidly for filters dosed with more contaminated feed water. The results from the analysis of average BSF performance suggest that the lower the fraction of initial flow rate, the better average performance of the BSF for $\log_{10}$ *E. coli* reduction. For example, in laboratory studies of the plastic BSF, flow rate decreased during the ripening process and reached fractions as low as 0.10 of initial flow rate in less than two months (Elliott et al., 2006). In this study, the average fraction of initial flow rate was 0.9 after six months of operation. The field results suggest that the conditions in these communities did not encourage the magnitude of ripening that was predicted by conditions used in the laboratory.

Overall, this is the first study that has attempted to determine what practices and performance characteristics can be used in the field to improve the *E. coli* reduction efficiency of BSFs. The results of this field study suggest that more research needs to be done to better understand the relationships between the results observed in the field to those observed in the laboratory and to use this information obtained to develop household management practices that improve field performance of the BSF.

Comparison of BSF to Other Household Water Treatment Technologies

There are a number of household water treatment technologies currently being used in many countries around the world. These technologies include boiling, solar disinfection, chemical disinfection, ceramic microfiltration and the biosand filter. In a simple cost-benefit analysis comparing a chemical coagulant and slow release disinfectant, ceramic filters and the biosand filter, it was found that based on laboratory evidence and the cost of the technologies, the biosand filter was the most cost effective treatment options (Casanova, Brown, Elliott, Stauber, & Sobsey, 2005). However, laboratory evidence alone would suggest that chemical disinfection and ceramic microfiltration can achieve higher microbial reductions for *E. coli* than does the BSF.

One of the major differences between the BSF and other treatment technologies is its ability to improve performance over time. Limited field studies suggest that months and years after implementation, the BSF continues to provide significant reduction of *E. coli* and perhaps even improved performance compared to initial field performance (CAWST, 2006). This is not necessarily the case with other treatment technologies. In a recent study of ceramic microfilters, researchers found that there was a linear decrease in filter use and filter function as time after implementation increased (Brown & Sobsey, 2006). Because the BSF is a relatively robust technology, it continues to function for years after implementation and because of the simplicity of its design; it requires little maintenance or replacement parts. Furthermore, in our field study and other studies of the BSF in the field, the findings suggest high potential for sustainability. The filters studied in Haiti had been used for 1-5 years and nearly all of the filters of this longitudinal study from the DR are still in use more than 1 year after installation ((Ortiz, Stauber, Aiken, & Sobsey, 2007) unpublished results). There are few studies of chemical or solar disinfection technologies that demonstrate continued use and sustainability. In a recent meta-analysis, the researchers found decreased compliance even over a six month period of study (Arnold & Colford, 2007). In contrast, the field evidence of this study suggests that >90% of households continued to use the BSF during the entire six-month study period and that they achieved high microbial reductions when treating poor quality water, both of which are evidence of effective performance, sustainability and robustness of the BSF technology.

Conclusions

Reductions of *E. coli* in 75 BSFs in the longitudinal portion of the field study were moderate, with 80% average reduction over the entire six month study. The 106 BSFs sampled in a cross-sectional analysis of filters installed by different implementers 4-9 month prior to sampling, demonstrated higher reductions of *E. coli* (94%). These differences suggest that the newly installed BSFs were still going through a period of initial maturation or ripening and therefore were performing less effectively than ripened filters. There was some evidence of increasing filter ripening over the six-month period of the field study on newly installed filters of the RCT, based on observation of a small decrease in filter flow rate and increased reductions of *E. coli*, total coliforms and turbidity over time. However, filter ripening in the field was not nearly as rapid as expected based on evidence from previous laboratory studies. This observed difference in ripening rate between laboratory controlled conditions and field conditions suggests that filter ripening in these communities was much slower, perhaps due to comparatively less contaminated water, based on relatively low *E. coli* concentrations and low turbidity compared to laboratory conditions.

The continued use of >90% of the installed filters at end of the 6-month observation period and the increase in performance in *E. coli* reduction over time suggests that the BSF has the potential to be a sustainable technology. The moderate performance in reduction of *E. coli* during the six-month field study does suggest that more information should be gathered to determine if this was specific to Bonao, DR and the water quality conditions presented in this study. Furthermore, the actual sustainability of these filters needs to be better documented for longer periods of time and compared to other treatment technologies. While other household water treatment technologies may have higher potential microbial reduction efficiency, their decrease in continued use over time and decreased effectiveness due to decreased use compliance over time may be a disadvantage of those technologies as compared to the BSF. The BSF may be less susceptible to such decreased use and declining performance over time, but further study will be needed to determine if this is the case over long period of time in user households and communities.

Chapter 5: Health Impact of the Biosand Filter

5.1 Introduction

More than one million people die annually as a result of diarrheal diseases. Although mortality from diarrheal disease is decreasing globally, morbidity is not. The average child in developing countries experiences three or more cases of diarrheal disease each year (Kosek et al., 2003) and this accounts for up to 4 billion cases of diarrheal disease annually. Diarrheal diseases make up 4% of the global burden of disease. A recent review suggests the environment plays an important role in the global burden of diseases. This review estimates that 94% of diarrheal diseases are attributed to environmental causes. This suggests that interventions can be made in water, sanitation and hygiene to attempt to decrease the burden of diarrheal disease (Pruss-Ustun & Corvalan, 2006).

Household drinking water quality interventions have been documented to reduce diarrheal disease by 40% and improve drinking water quality significantly (L. Fewtrell et al., 2005). A promising household water treatment technology is the biosand filter (BSF). An intermittently operated slow sand filter, the biosand filter has been implemented in more than 80,000 homes around the world. Laboratory evidence documents that it reduces fecal microbe contamination by about 90% for viruses, 90-99% for bacteria and >99.9% for protozoan parasites. However, the BSF lacks rigorous scientific evidence of its ability to reduce diarrheal disease of users.

The purpose of this study was to perform a randomized controlled trial of the biosand filter in Bonao, Dominican Republic (DR). The DR is a good location to evaluate the BSF because these filters have been implemented in the DR for the past seven years. Therefore, filter implementation capacity exists in the DR. The DR also continues to experience relatively high rates of diarrheal disease; with a two week point prevalence of diarrhea estimated at 14% in the 2002 Demographic and Health Survey (*Encuesta Demografica y de Salud: Republica Dominicana*, 2003).

5.2 Methods

The study was designed to evaluate the impact of newly installed biosand filter use on diarrheal disease rates. A randomized controlled trial (RCT) was performed. This trial did not involve the use of a placebo biosand filter. This is because such a placebo filter was considered technically and ethically unfeasible. The study area selected was in the capital of the province of Monseñor Nouel, Bonao. The RCT was performed in two communities of Bonao: a semi-rural community called Jayaco and an urban community called Brisas del Yuna. Field data collection began on September 19, 2005 and was completed on July 27, 2006.

Jayaco is a semi-rural community of approximately 700-800 homes surrounded by agricultural rice fields located eight miles north of the municipality of Bonao. The Jayaco community has access to health services through a rural health clinic run by the provincial government. Within the Jayaco community, five areas were selected for participation in the RCT of the BSF: Jayaco Arriba, Majaguay, KM 100, KM 101, and KM 103. Households in Jayaco have access to many different types of drinking water. The sources of drinking water in the community vary and typically depend on the geographic location of the household and season (wet or dry). They include piped water, wells, unprotected springs, river water, and collected rainwater. For example, households located in the region of the community that is closest to the entrance from the highway (Jayaco Arriba), have access to piped water supplies near or on their property. However, households located in the community the farthest from the entrance typically rely on rainwater collection, surface water or wells. Piped water, unless otherwise noted, is supplied via a system of aqueducts under the direction of the National Institute for Aqueducts and Potable Water (Instituto Nacional de Agua Potable y Aqueductos – INAPA).

In addition to Jayaco, a community inside the municipality of Bonao was also selected to participate in the RCT of the BSF. Brisas del Yuna is an urban community on the edge of the Yuna River comprised of 100-200 households. It represents an underserved community inside the city of Bonao, although it does have access to health services through a private clinic located in the center of the community. Households in Brisas del Yuna rely on piped water, well water, an unprotected spring or river water for drinking water. As in Jayaco, the drinking water source also typically depends on location of the household within the community. Both communities represent a diverse group of households having a range of access to services, levels of education, and wealth distribution, which was characterized in the study (see Appendix 1)

Household Selection and Sample Size

A cross-sectional study was performed in June – August 2005 in the two communities in Bonao. The purpose of the cross-sectional study was to collect data on diarrheal disease rates of community household members and potential risk factors for diarrheal disease such as socio-economic status, access to sanitation, etc. The households from this study were then asked to participate in the randomized controlled trial of the biosand filter. Requirements for inclusion in the study were: no BSF in the household, at least one child under five years of age and willingness to participate. At start of the longitudinal study, 187 households were enrolled. Households enrolled in September 2005 were visited for four months prior to randomization into BSF and control groups. During this period, households were visited and interviewed regarding water management practices, diarrheal disease and interviewers collected water samples periodically. After four months of data collection, households were selected into either BSF or control groups. However, households were not aware of whether or not they would be assigned to the BSF intervention group or the control (no BSF) group until one week prior to BSF installation. They were told that if they participated for the entire length of the study, they would receive a BSF; either at intervention time or at the end of the study period. Households were also allowed to leave the study at any point, but would not be allowed to keep the filter if they left prior to the end of the study. One week prior to randomization, all households were assigned a unique number and random numbers were generated to identify the ~50% of the households that were selected to receive BSF.

Sample Size Calculation and Household Recruitment

Prior to household recruitment, sample size calculations were performed to determine both the number of households needed and the length of the observation period to be able to detect a >15% reduction in diarrheal disease of users with 80% power and an α of 0.05. Calculations suggested that approximately 150 households would be needed and followed for a period of six months to detect a diarrheal incidence rate reduction of >15%. Based on these calculations, at the start of the longitudinal study 187 households were recruited. Although this number was higher than the number calculated, it took into account the attrition rate expected in a study that would last almost an entire year (somewhere between 10-20%).

Diarrheal Disease Surveillance

A system for diarrheal disease surveillance was established as part of weekly household interviews. During an initial cross-sectional interview, household primary respondents were identified. The primary respondent for the household was typically identified as the primary child care giver. At approximately 7 day intervals, the household's primary respondent was asked to verbally report cases of diarrheal disease for all participants in the household. If the primary respondent reported a case of diarrhea, they were asked: the date the case began, the frequency of the evacuations, duration and a description of stool consistency and the presence of blood in stools. If the case was on-going, it was followed up during the next household visit. Diarrheal disease surveillance began on September 19, 2005 and was completed on July 27, 2006. During this longitudinal study period, diarrheal disease surveillance was not performed during the weeks beginning on: December 26, 2005, January 2, 2006 and April 10, 2006, due to national holidays. In addition, for the week beginning October 24, 2005, surveillance was halted due to a local strike in the city of Bonao that made it too dangerous to travel to the communities. The study period before BSF installation for intervention phase consisted of 16 full weeks of household observation and the period after BSF installation for the intervention phase consisted of 23 weeks of household observation. All observations were included and intention to treat was used in the statistical analyses.

Biosand Filter Installation for Intervention Study

During the first week of February 2006, 81 concrete BSFs were installed in homes by a local filter technician. The technician explained use and operation of the BSF to a household participant and provided a brochure about the use of the filter for future reference. Households were instructed to add water to the BSF for five successive days before using it. No additional educational messages on sanitation or hygiene were provided. However, all households receiving the BSF were also given a 5-gallon narrow mouth container (or bottle) with a base that allowed water to filter directly into the container for safe storage of the BSF filtered water. A picture of the BSF with the water bottle and the base is shown in figure 5.1.

Figure 5.1 Biosand filter with base and water storage container in household in the KM 100 area of the Jayaco community

Drinking Water Quality Testing

In addition to weekly household surveys about diarrheal disease, households were asked at approximately two week intervals (but no longer than three weeks) to provide samples of stored drinking water. After initiating the BSF intervention, households that received filters during the longitudinal study period were asked to the provide the following household water samples: stored drinking source water prior to BSF, drinking water directly from the BSF outlet, stored BSF-treated water, and stored BSF-treated water that received any additional treatment. There were seven drinking water sampling periods prior to filter installation and initiating the BSF intervention, and 11 drinking water sampling periods after filter installation and during the BSF intervention period.

Water samples were poured directly out of household drinking water storage containers into 500mL sterile Whirlpak® bags and stored on ice until processing and analysis, which occurred within six hours. Water samples were collected in the field and transported to Dr. Mirna Peña's Clinical Laboratory where the samples were tested for total coliforms and *E. coli* via the IDEXX Colilert™ Quantitray system (IDEXX, Laboratories, Westbrook, ME). Water sample volumes of 100mL were combined with one packet of Colilert™ test reagent media in a 120mL capacity reagent bottled that contained sodium thiosulfate to neutralize chlorine. Samples were mixed briefly, poured into Quantitrays, sealed and incubated 20-24 hours at 35˚C (± 1). Wells which turned yellow were scored positive for total coliforms and wells that fluoresced blue under a long wavelength UV light were scored positive for *E. coli*. The values of positive wells counted from the Quantitrays were used to obtain most probable number (MPN) values according to an MPN table provided by IDEXX. Data from water quality analysis were $\log_{10}$ transformed and analyzed as both continuous and categorical values.

Data Analysis

Univariate and Stratified Analysis

The effect of the BSF on diarrheal disease rates of BSF users (intervention households) compared to BSF non-users (control households) was determined by comparing incident cases of diarrhea for each group. The definition for a case of diarrheal disease was based on the World Health Organization's definition: three or more watery evacuations in a 24-hour period or any evacuation with blood in it. If the participant reported symptoms in more than one week of the consecutive household visits; a new case of diarrhea was assigned only when the symptoms had been preceded by three or more successive days free of diarrheal disease. Diarrheal disease incidence rates were compared between the intervention and control groups for the study periods before and after the BSF intervention. To assess for effect measure modification, stratified analyses were performed to determine the effect of the following covariates: month (and rainfall), season, and gender.

Multivariate Analysis

Multivariate logistic regression was used to control for multiple covariates. The multivariate analysis was performed in Intercooled Stata 8.0 (Stata, StataCorp, College Station, TX). All observations were made as interviews performed at the household level during each week that a representative from the household was available for interview. The outcome variable was cases of diarrheal disease for an individual. All observations were included in the analyses. The main exposure variable was whether or not the participant was randomized to the BSF group or the control group and was classified according to intention to treat analysis. The variables and their coding schemes are listed in Appendix 2. Because the observation period was only seven days and the incidence rates of diarrheal disease were relatively low (less than 0.10), the odds ratio (OR) produced from the logistic regression models were used to approximate the incidence rate ratios (IRR). Therefore, all IRRs reported are based on the ORs from the logistic regression.

Covariates were assessed in a step-wise procedure in which covariates of interest were fitted in a full model and were deleted in a stepwise procedure using ordinary logistic regression. Selection criteria to keep covariates in the model were based on an a priori change in the coefficient of the exposure (BSF or control household) by 10% or more. After the simplified ordinary logistic regression model was determined, two additional models were used: generalized estimating equations extension of logistic regression and random-intercepts logistic regression. Generalized estimating equations were used to address correlation within the data set and these equations are appropriate for addressing correlation due to repeated sampling of individuals over time. However, they are not as adequate for addressing hierarchical structures with more than two levels. In three level models, individuals, who are repeatedly sampled, belong to groups and are nested in clusters. This type of hierarchical structure is illustrated in figure 5.2 (Rabe-Hesketh & Skrondal, 2005). Increasingly, mixed models are being used to account for three level hierarchical structures. Mixed models such as the random intercept logistic regression model incorporate both fixed and random effects. The data from this study lends itself well to the 3-level hierarchical model structure because individual participants are observed repeatedly, and they each belong to the household that was randomized into BSF or control household. The random intercepts logistic regression model can accommodate both between subject and between household variations and provides the most correct estimate of the standard error which is used to estimate the 95% confidence intervals. The results from all three models were reported.

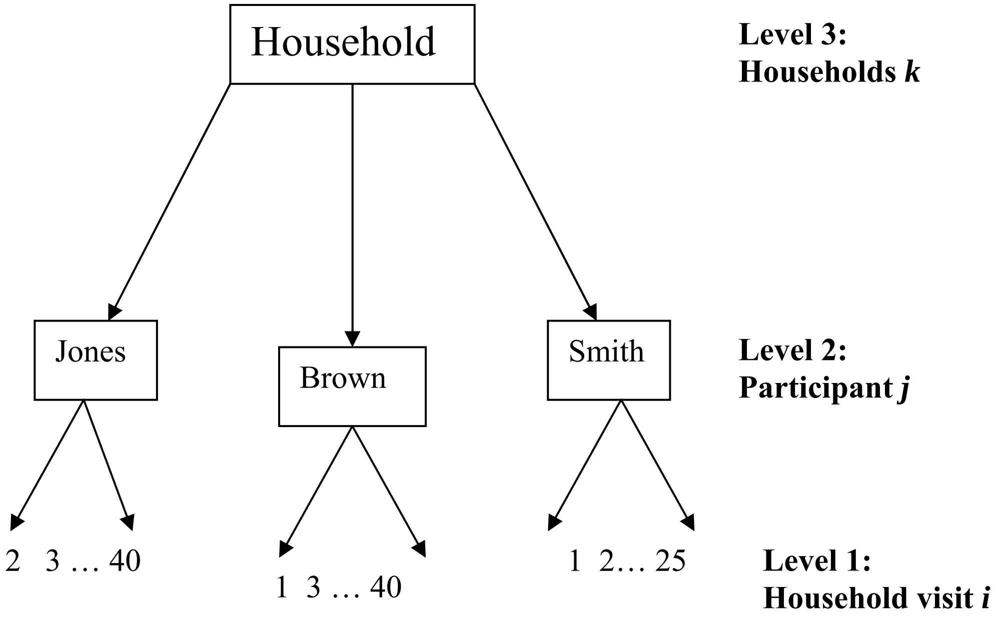


Figure 5.2 Illustration of three-level design

5.3 Results

Study Enrollment and Completion

After the cross-sectional study was completed in August 2005, all households with children under five years of age in Brisas del Yuna and Jayaco were asked to participate in the longitudinal phase of the study. A diagram of enrollment and participation is shown in figure 5.3. In September 2005, 187 households were enrolled and began the longitudinal portion of the study. From September 2005 to February 2006, 20 households left the study.

The primary reason for leaving the study was either: the household moved out of the area or the child that was living in the household left to live in a different household.

In February 2006, 81 households were randomly selected to receive the BSF. Of these, 75 (93%) households completed the study. Two households quit the study and returned the BSF; two households moved and returned the BSF. Two additional households reported still using the BSF; however, they were unavailable to participate in interviews because the principal respondent was working at the time of visit. Of the 86 households that did not receive the biosand filter in February 2006 (control households), 79 (92%) remained as participants in the study to receive the BSF in August 2006. Six households moved and one household could not participate in the interviews because the principal respondent was working at the time of visit.

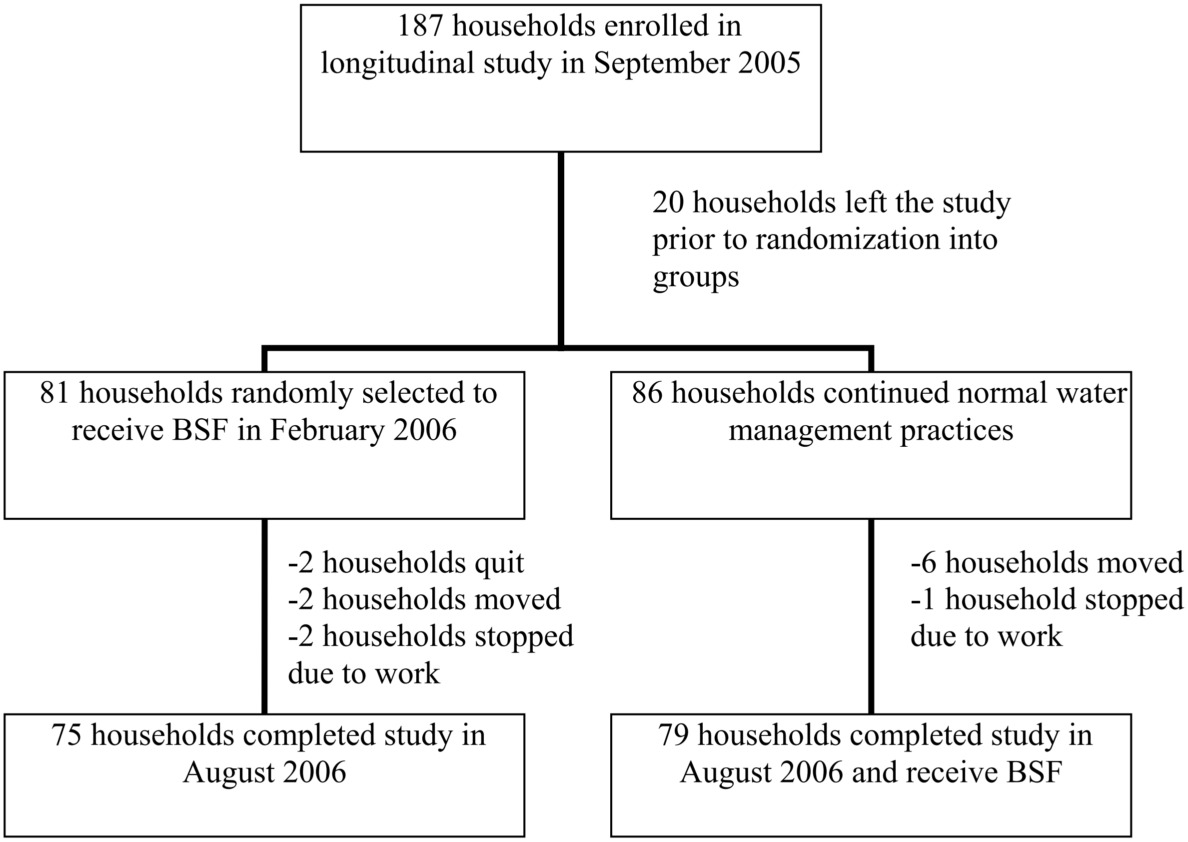


Figure 5.3 Diagram of household enrollment and participation in RCT for BSF

Baseline Characteristics and Group Comparability:

During June through August 2005, a cross-sectional study was performed to recruit households into the longitudinal study. During the cross-sectional study, data were collected both at the level of the individual participant and at the household level. The data collected at the level of the individual are summarized in table 5.1 for the following variables: location, age, and gender. At the time of filter installation, 907 people were participating in the study. Of those, 447 were randomized (at the household level) to the BSF intervention group and 460 were randomized to the control group. There were nearly equal numbers of participants in each community location with one exception. There are more participants in the control group from the community of Jayaco Arriba compared to the BSF group and this was found to be significantly different ($p < 0.05$). This is important because community location can serve as a proxy for environmental and socio-economic conditions. Other variables measured on the individual level suggest a relatively equal distribution of participants into both groups with one other exception: gender of participants under five years old. Control households have about 8% higher proportion of males under five than females as compared to the BSF households, although this was not found to be a significant difference. Almost all other individual and household characteristics were the same in both groups. Households average five people per household. Average age of children under five is two years old and average age of participants over five years old is 24 years old.

In addition to data collected about individual participants, data on water management and water practices in the BSF and control households are summarized in table 5.2. The results suggest a wide range of household water management practices. There are about equal numbers of households in BSF and control groups using river, rain, tap, well and bottled water (data not shown). In addition, household drinking water sources and use practices were assessed throughout the longitudinal study and were evaluated in the multivariate analyses. Approximately 50% of households report collecting drinking water at least once a day and 20% of participants reported collecting drinking water 1-2 times a week. In addition, 35-40% of both groups reported practicing some form of drinking water treatment, and 25% reported purchasing bottled water.

Also during the cross-sectional survey, households were asked to report the one-week point prevalence of diarrhea for members of the household, including children under the age of five years old. These data are shown in Table 5.2. Approximately 20% or 1/5th of all households reported having had a case of diarrhea in the seven days prior to the cross-sectional survey. Households in the BSF group reported a slightly higher (27% vs. 17% in control households) one-week point prevalence of household diarrhea but the difference was not found to be statistically significant. Approximately 80% of the households who reported diarrhea reported that diarrhea was in a child under five. Additional information on the two communities can be found in Appendix 1.

Table 5.1 Age (September 2005), gender and location for participants of the randomized controlled trial of the BSF in Bonao, DR in 2005-2006

	Groups		
variable	**Control (n=460) n (%)**	**Intervention (n=447) n (%)**	**Total (n=907) n (%)**
Location			
Jayaco Arriba*	99 (21)	62 (14)	161 (18)
Majaguay	44 (10)	53 (12)	97 (11)
KM 100	49 (11)	47 (10)	96 (10)
KM 101	59 (13)	65 (15)	124 (14)
KM 103	84 (18)	84 (19)	168 (18)
Brisas del Yuna	125(27)	136 (30)	261(29)
Age			
Participants ≥ 5 years old	332 (72)	332 (75)	664 (73)
Participants < 5	128 (28)	115 (25)	243 (27)
Mean Age (std. dev)†			
Participants (≥5)	24.2 (15.4)†	24.4 (15.6)	24.3 (15.5)
Participants < 5	2.0 (1.3)	2.2 (1.3)	2.1 (1.3)
Household Size			
Range (participants)	2 – 12 per house	3 – 15 per house	2 - 15
Average	5.3 per house	5.5 per house	5.5
Gender			
Male (<5)	69 (54)	52 (45)	122 (50)
Female (<5)	59 (46)	63 (55)	121 (50)
Male (≥5)	155 (47)	160 (48)	315 (47)
Female (≥5)	177 (53)	172 (52)	349 (53)

*** - p < 0.05 (t-test or chi-squared test), † -standard deviation of average age listed**

Table 5.2 Drinking water management practices and diarrheal disease from summer survey in BSF and control groups in RCT in Bonao, DR in 2005-2006

Variable	**Groups**		
	Control (n=86) n (%)	**Intervention (n=81) n (%)**	**Total (n=167) n, (%)**
# of times collect drinking water/7days			
1	8 (9)	1 (1)	8 (5)
2	10 (12)	17 (21)	27 (16)
3	12 (14)	16 (20)	28 (17)
4	3 (3)	3 (4)	6 (4)
5	0 (0)	1 (1)	1 (1)
7 or more	52 (60)	41 (51)	93 (56)
Miss	1 (1)	2 (2)	3 (2)
Report treating drinking water			
Yes	36 (42)	29 (36)	65 (39)
No	49 (57)	50 (62)	99 (59)
Missing	1 (1)	2 (2)	3 (2)
Report buying drinking water			
Yes	21 (24)	22 (27)	43 (26)
No	64 (75)	57 (70)	121 (72)
Missing	1 (1)	2 (3)	3 (2)
Soap at time of interview			
Yes	64 (75)	52 (65)	116 (69)
No	21 (24)	27 (33)	48 (29)
Missing	1 (1)	2 (3)	3 (2)
Diarrhea in last 7 days			
Yes	15 (17)	22 (27)	37 (22)
No	70 (82)	57 (70)	127 (76)
Missing	1 (1)	2 (3)	3 (2)
Diarrhea in last 7 days (< 5)			
Yes	14 (16)	17 (21)	31 (19)
No	71 (83)	62 (77)	133 (79)
Missing	1 (1)	2 (2)	3 (2)

Univariate Analysis for RCT of BSF:

Prior to BSF intervention, the BSF and control households had similar incidence rates of diarrheal disease. However, after BSF intervention, BSF households experienced a 53% lower incidence rate of diarrheal disease as compared to control households for all participants. As shown in table 5.3, the diarrhea incidence rate ratio (IRR) of BSF households to control households prior to BSF intervention is 1.03 (95% CI 0.83, 1.26), and after BSF intervention the IRR is 0.47 (95% CI 0.37, 0.59). Weekly incidence rates of diarrheal disease over the entire study period are shown in figure 5.4. The rates of diarrheal disease in BSF and control households overlapped frequently in the 16 weeks prior to BSF intervention. However after BSF intervention, the diarrheal disease rates only overlapped during five household visits during the 23 weeks of observation: visit numbers 29, 30, 31, 33, and 34. The period of time where rates overlap for BSF and control households, corresponds to time during the months of May and June 2006. These data suggest that the effect of the BSF intervention varies over the six months of the intervention period.

Table 5.3: Unadjusted incidence rates, incidence rate differences and incidence rate ratios for diarrheal disease in BSF and control groups prior to and after BSF intervention during the RCT in Bonao, Dominican Republic from September 2005 to July 2006

	Cases	**Person-weeks contributed**	**IR***	**IRD†**	**95% CI‡**	**IRR§**	**95% CI**
Prior to BSF							
Control	174	6,686	0.025				
Filter	172	6,932	0.026	0.001	-0.005, 0.006	1.03	0.83, 1.26
After BSF							
Control	234	9,687	0.024				
Filter	104	9,205	0.011	-0.013	-0.017, -0.009	0.47	0.37, 0.59

* - IR - incidence rate
† - IRD - incidence rate difference
‡ - 95% CI - 95% confidence interval
§ - IRR - incidence rate ratio with control as referent group

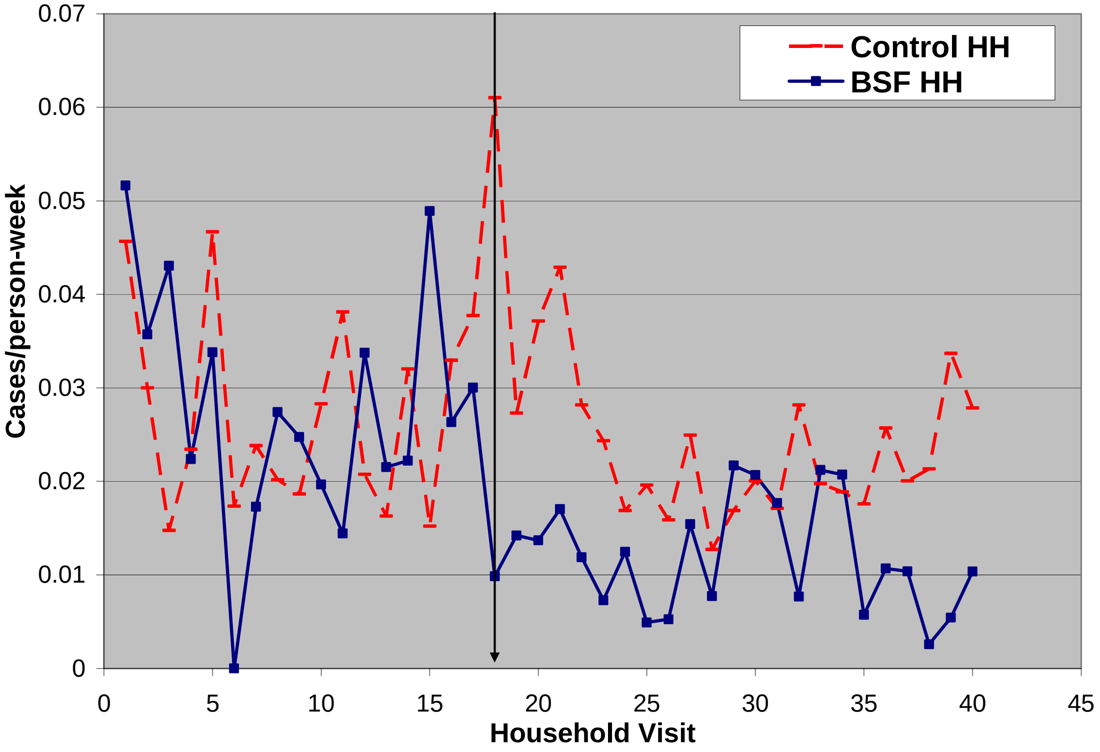


Figure 5.4 Incidence rates of diarrhea by household group over longitudinal study. Vertical line indicates installation of BSF in February 2006.

To examine the variation in occurrence of diarrheal disease in BSF and control households during the six months of the BSF intervention, IRRs were calculated for each month of observation, for February through July 2006. Incidence rate ratios stratified by month are shown in table 5.4. The monthly IRR ranged from 0.25 to 0.76, was highest in May and lowest in July. In the months of May and June, the reduction of diarrheal disease incidence rates by the BSF is 24 and 35 %, respectively, and the 95% confidence intervals of IRRs for these months crossed the null, demonstrating lack of statistical significance.

Rainfall varied throughout the study period. The highest monthly rainfall occurred in September 2005 with 497mm and the lowest monthly rainfall occurred in February 2006 with 58 mm. Rainfall for the six months of the intervention study period is also listed in table 5.4. Interestingly, rainfall during the intervention period was highest for the months of April and May (both > 440mm of rainfall) while all other months have less than 265 mm of rainfall. The high volume of rainfall in April and May, followed by lower incidence rates of diarrheal disease in control households in May and June, suggests a seasonal effect of diarrheal disease rates in control households due to rainfall. During the same period of time, rates of diarrheal disease also appear to increase in BSF households. This increase may be due to changes in water sources and potential changes in the types of organisms that are being transmitted as the cause of diarrheal diseases.

Table 5.4: Assessing effect measure modification of month of intervention on the relationship between group (BSF and control household) and diarrheal disease after BSF intervention in RCT in Bonao, Dominican Republic during intervention from February 2006 to July 2006

Rainfall per month in millimeters during intervention	**IRR†**	**95% CI‡**	**M-H Weight§**
February 2006 (58 mm)	0.29	0.15 – 0.56	19.31
March 2006 (251 mm)	0.49	0.31 – 0.78	26.57
April 2006 (445 mm)*	0.41	0.20 – 0.86	12.31
May 2006 (465 mm)*	0.76	0.45 – 1.26	17.01
June 2006 (160 mm)	0.65	0.39 – 1.08	18.17
July 2006 (261 mm)	0.25	0.12 – 0.49	20.86
Crude IRR	0.47	0.37 – 0.59	
Mantel Haenzel combined	0.47	0.37 – 0.59	
M-H test for homogeneity			*p = 0.063*

*- These two months have highest rainfall
† - IRR = Incidence rate ratio
‡ - 95% CI = 95% confidence interval
§ - Mantel Haenzel weight used for combined estimate

The data in figure 5.5 suggest a periodic effect of rainfall magnitude on diarrheal disease rates in both BSF and control groups prior to BSF intervention and in the control groups after BSF intervention. In this graph, increases in monthly rainfall occur one month prior to declines in monthly diarrheal disease rates. For example, rainfall amount for

September 2005 is plotted along with diarrheal disease rates for October 2005. Prior to BSF intervention, the two months that had high rainfall (> 440 mm per month), also had low incidence rates of diarrheal disease in both household groups (BSF and controls); less than 0.02 cases per person-week. Prior to BSF intervention, where rainfall monthly amounts do not exceed 167 mm per month, diarrheal disease incidence rates increased and were almost double those in the months of high rainfall. After BSF intervention, diarrheal disease rates were below 0.015 cases per person-week for all months in the BSF group. In the control group, monthly rates ranged from 0.04 - 0.017 cases per person-week. During the BSF intervention, incidence rates of diarrheal disease in the control group were lowest in May and June, the two months that corresponded to months experiencing high rainfall 1 month prior for each, i.e. April and May.

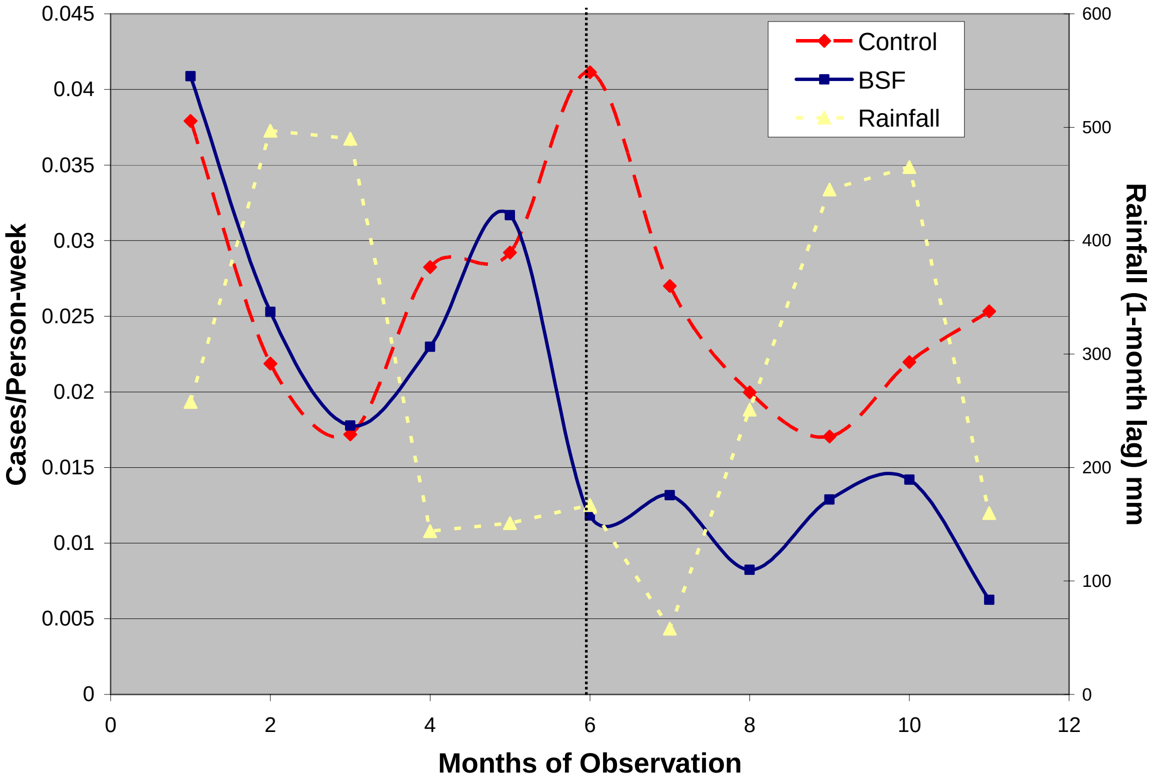


Figure 5.5 The effect of rainfall on average incidence rates of diarrheal disease in BSF and control groups (Rainfall is lagged one month behind diarrheal disease rates).

Based on the data from the stratified analysis in table 5.4 and the trends in figure 5.5, the months of the BSF intervention study period were classified into two categories: wet season and dry season. The effect of the BSF in these two different periods was calculated by classifying February, March, April and July as dry season months and May and June as wet season months. This classification takes into account a one month lag in rainfall amounts since the higher quantities of rainfall seem to correlate better with decreased incidence rates one month later. The unadjusted incidence rate ratio for diarrheal disease reduction by the BSF for the wet season is 0.70 (0.49-0.99) and for dry season is 0.34 (0.25-0.46), as shown in table 5.5 where wet season was defined as the period of time when rainfall exceeded 400mm per month and dry season was < 400mm per month. Effect measure modification was assessed using a Mantel-Haenzel test for homogeneity. The effect was found to be significantly different in the dry season versus the wet season. Based on these initial results, season was then also assessed in the multivariate model as an effect measure modifier.

Table 5.5: Assessing effect measure modification by season on the relationship between diarrheal disease and BSF intervention in RCT in Bonao, Dominican Republic, February 2006 – July 2006

Effect of season on intervention	**IRR†**	**95% CI‡**	**M-H Weight§**
Dry season 2006 *	0.37	0.27 – 0.49	79.00
Wet season 2006	0.70	0.48 – 1.00	35.29
Crude IRR	0.47	0.37 – 0.59	
Mantel Haenzel combined	0.47	0.37 – 0.59	
MH Test of homogeneity			*p = 0.007*

*- Dry season is defined as months Feb, March, April, July and wet season is defined as May and June.
† - IRR = Incidence rate ratio
‡ - 95% CI = 95% confidence interval
§ - Mantel Haenzel weight used for combined estimate

Water Quality Analysis

Household drinking water quality was compared over the entire study period for households with and without the biosand filter. Monthly water quality concentrations for *E. coli* per 100mL were averaged for each group, BSF intervention and control households, using arithmetic and geometric means. These data are presented in table 5.6. Arithmetic means can become skewed as a consequence of averaging in the higher concentrations. The data from the geometric means are more normally distributed (data not shown) as they are log transformed prior to averaging. Before BSF intervention, household groups had similar geometric mean MPN concentrations of *E. coli* per 100mL in household drinking waters: 23 for control households and 22 for BSF households. After filter intervention, households with the BSF had improved water quality compared to control households, based on *E. coli* concentrations. Control households had 23 *E. coli* per 100 mL compared to only 12 *E. coli* per 100 mL in BSF households. Also presented in table 5.6 are arithmetic averages. They suggest that prior to BSF intervention; BSF households had higher concentrations of *E. coli* with 181 MPN *E. coli* per 100mL as compared to control households with 159 MPN *E. coli* per 100 mL. After the BSF intervention, BSF households had improved water quality based on arithmetic averages, compared to control households. Control households had 192 MPN *E. coli* per 100mL compared a lower concentration of 116 MPN *E. coli* per 100mL in BSF households.

Table 5.6: Drinking water quality in filter and control groups prior to and after filter intervention (based on monthly averages)

	Geometric Mean MPN *E. coli* per 100 mL (SD)	Arithmetic Mean MPN *E. coli* per 100 mL (SD)
Prior to Intervention		
Control	23 (9)	159 (367)
Filter	22 (10)	181 (436)
After Intervention		
Control	23 (10)	192 (411)
Filter	12 (8)	116 (338)

Household drinking water quality variations were examined over time and compared to the monthly averages of rainfall. In table 5.7, the monthly geometric mean *E. coli* concentrations for each household group are compared. Household drinking waters between the two groups were similar in the five months preceding the BSF intervention, where the BSF household group had higher concentrations of *E. coli* three out of five months. After BSF intervention, BSF households had lower concentrations of *E. coli* for all six months. However, the difference in *E. coli* concentrations for the two groups is very small in

February. Water quality after BSF intervention varies from approximately 13 – 37 MPN *E. coli*/100 mL in control households and from 8 -25 MPN *E. coli/*100mL in BSF households.

There appears to be a pattern in the variation of *E. coli* concentration in drinking water by month with monthly rainfall quantity. Rainfall quantity and *E. coli* concentrations for both groups are presented in figure 5.6 and table 5.7. For the first month of high rainfall after the periods of low rainfall, there was an increase in concentration of *E. coli* in household drinking water for both BSF and control households. After BSF intervention, this increase in *E. coli* concentrations occurred during April 2006. Following four months of relatively low monthly quantities of rainfall, drinking water quality based on *E. coli* concentrations deteriorated for both groups in April. April *E. coli* concentrations doubled in control households and also increased in BSF households compared to the values for March. For months where rainfall quantities are low (about half of what they were in April) such as January, February or March, *E. coli* concentrations are all below 20 MPN per 100mL in both household groups. This pattern of association between low monthly rainfall and low monthly *E. coli* concentrations in rainwater did not occur in all months of the study and only explains a portion of the variation in drinking water quality. For example, there is one month where *E. coli* concentrations increased in both household groups but this did not correspond to a month with increased rainfall. The relationship between diarrheal disease and water quality is influenced by rainfall but the relationship is not entirely clear from these data.

Table 5.7: Geometric mean concentrations of *E. coli*/100mL by month for BSF and control households during the BSF RCT in Bonao, Dominican Republic 2005-2006

Month	**Control Household**	**Filter Household**	**Monthly Rainfall (mm)**
1 - September 2005	21	28	497
2 - October 2005	24	20	490
3 - November 2005	27	32	144
4 - December 2005	31	36	151
5 - January 2006	11	6	167
6 - February 2006	13	13	58
7 - March 2006	17	8	251
8 - April 2006	36	11	445
9 - May 2006	21	11	465
10 - June 2006	37	25	160
11 - July 2006	23	10	261

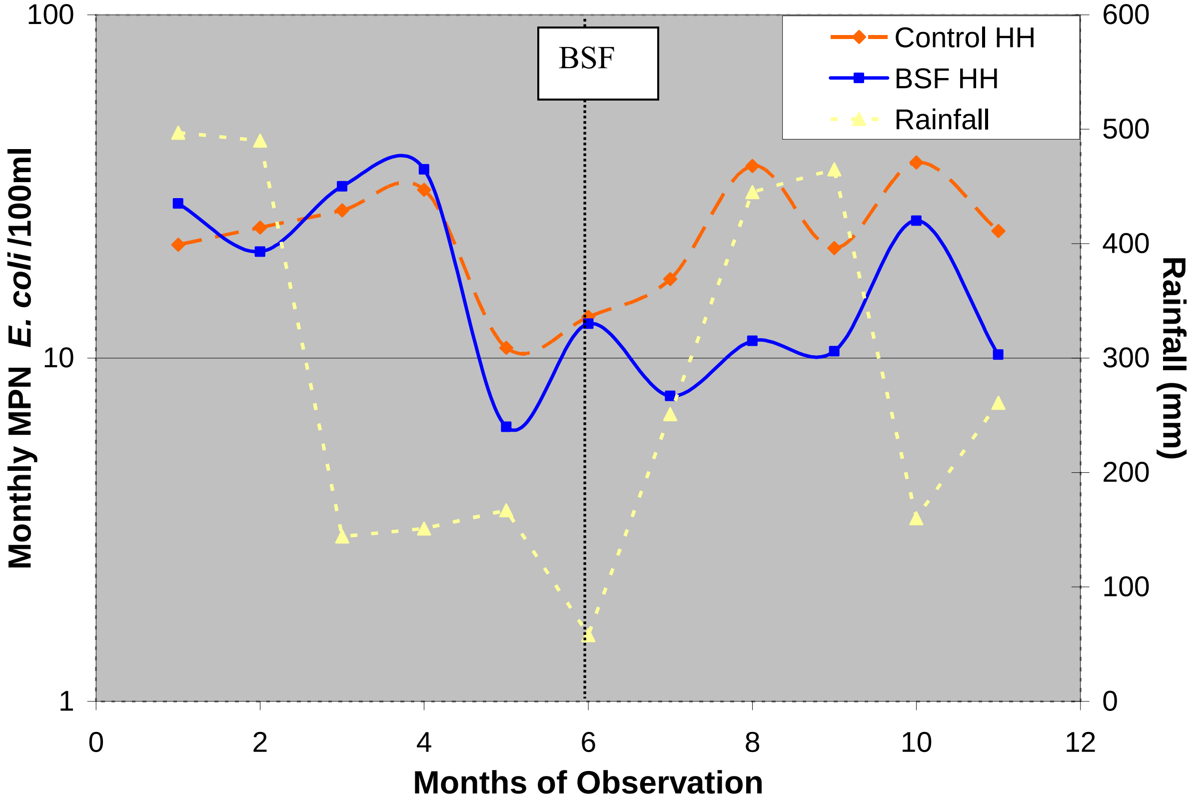


Figure 5.6 Water quality and rainfall in control and BSF households during RCT in Bonao, DR in 2005-2006

To further examine the effect of periodic changes in *E. coli* concentrations in household water and its potential effect on diarrheal disease rates, the data for these two variables were compared over the entire study period. Figure 5.7 is a graph of water quality and diarrheal disease rates. Prior to BSF intervention, water quality based on *E. coli*/100 mL in control and BSF households as well as the diarrheal disease rates in control and BSF households essentially overlapped for both of these variables. After BSF intervention, households with the BSF experienced lower rates of diarrheal disease and improved water quality based on *E. coli* concentration as compared to control households. Fluctuations occurred in both monthly diarrheal disease rates and monthly water quality based on *E. coli* concentrations for both groups. Both monthly *E. coli* concentrations and monthly diarrheal disease rates are lower in BSF households compared to those in control households. However, there are other unaccounted exposure and disease outcome factors that make these results difficult to interpret, including the transmission of diarrheal diseases through other potential exposure routes, such as person-to-person contact and contaminated foods.

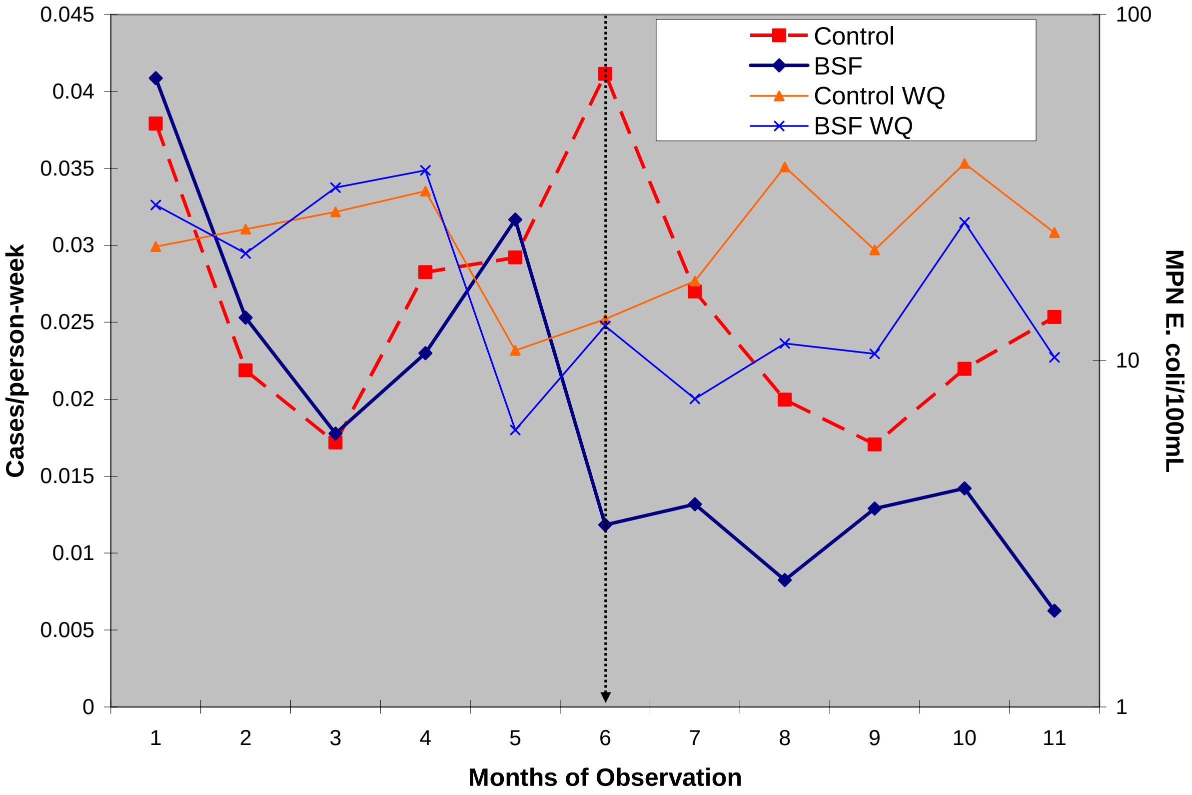


Figure 5.7 Diarrheal disease rates and water quality in BSF and control households during a randomized controlled trial of the BSF in Bonao, Dominican Republic 2005-2006.

Multivariate Analysis

The incidence rate ratio of diarrheal disease for the BSF household group versus the control household group was also estimated using multivariate logistic regression models with and without an interaction term for season. A full model was fitted to the data using ordinary logistic regression, GEE extensions and random-intercept logistic regression. Results from the three full models without the interaction term are listed in table 5.8. Even though the households were randomly selected to either receive the BSF intervention or be in the control group, potential covariates of interest were assessed for confounding during model formulation. The list of all covariates assessed appears in Appendix 2. Based on these analyses, a categorical age variable was included in the model. The incidence rate ratio of the diarrheal disease of biosand filter users as compared to the control households, adjusted for age of participant, was between 0.52 and 0.53 for all of the models fitted: ordinary logistic, GEE extensions and the random-intercept logistic regression. Additional covariates considered in the model included an indicator of wealth as listed in Appendix 1 and Appendix 2 and the most frequently used source of drinking water such as tap water, well water, etc. None of the variables remained in the model during stepwise elimination based on the 10% change in effect. However, when source of drinking water was treated as a group of variables, it had a 12% change in coefficient of the main exposure variable and should be more thoroughly investigated to determine the relevance of the variable.

The results from the models suggest that the BSF households experienced a 47% reduction in incidence rates of diarrheal disease during the 6-month period from February to August 2006. The 95% confidence intervals do not cross the null value for any of the three model estimates of IRRs, and therefore the IRR estimates are considered significantly different from the null value (of 1).

As expected, the ordinary logistic regression model produced the smallest standard error and most precise confidence intervals. However, the ordinary logistic model does not take into account the effect of clustered data and therefore does not correctly estimate the standard error of the effect measure. The standard error of the GEE extension of logistic regression was greater than the ordinary logistic regression but not as large as the standard error estimated by the random intercepts logistic regression. The random-intercepts logistic regression model accounts for variation between participants as well as between households and therefore results in the largest standard error of all three models. Regardless, all three analyses provided data documenting considerably lower diarrheal disease incidence rate ratios for BSF households compared to control households and therefore a protective effect against diarrhea risk.

Table 5.8 Incidence rate ratios for diarrheal disease in BSF compared to control households from multivariate model without interaction adjusted for categorical age of participant in randomized controlled trial of BSF in Bonao, DR 2005-2006

	Ordinary Logistic IRR (95%CI)	GEE extension of Logistic IRR (95%CI)	Random Intercepts Logistic Regression IRR (95%CI)
Intervention group (Control was referent)	0.52 (0.41, 0.66)	0.53 (0.40, 0.71)	0.53 (0.36, 0.79)

In addition to the full models without the effect measure modifier, the full model was assessed with season as an effect measure modifier. The covariate of categorical age was also included in these models. The beta-coefficients from the three multivariate models for ordinary logistic regression, GEE extensions of logistic regression and random-intercepts logistic regression are listed in Tables 5.9, 5.10 and 5.11 respectively.

Table 5.9 Results of multivariate model using ordinary logistic regression with season interaction term

Variable	**beta**	**SE**	**p-value**	**95% CI of beta**
Intervention group - (1 if filter, 0 if control)	-0.223	0.190	0.242	-0.596, 0.150
Season - (1 if dry, 0 if wet)	0.345	0.144	0.017	0.062, 0.628
Group* Season - (1 if dry and filter, 0 otherwise)	-0.692	0.246	0.005	-1.175, -0.209
Categorical age - (0 if <2, 1 if 2 – 4, 2 if > 4)	-1.344	0.076	0.000	-1.493, -1.195

Table 5.10 Results of multivariate model using generalized estimating equations extensions of logistic regression with season interaction term

Variable	beta	SE	p-value	95% CI of beta
Intervention group - (1 if filter, 0 if control)	-0.187	0.204	0.360	-0.588, 0.213
Season - (1 if dry, 0 if wet)	0.346	0.141	0.014	0.070, 0.622
Group* Season - (1 if dry and filter, 0 otherwise)	-0.710	0.249	0.004	-1.120, -0.222
Categorical age - (0 if <2, 1 if 2 – 4, 2 if > 4)	-1.344	0.076	0.000	-1.497, -1.119

Table 5.11 Results of multivariate model using random effects logistic regression with season interaction term

Variable	beta	SE	p-value	95% CI of beta
Intervention group - (1 if filter, 0 if control)	-0.152	0.273	0.577	-0.688, 0.383
Season - (1 if dry, 0 if wet)	0.357	0.194	0.066	-0.024, 0.738
Group* Season - (1 if dry and filter, 0 otherwise)	-0.769	0.301	0.011	-1.359, -0.178
Categorical age - (0 if <2, 1 if 2 – 4, 2 if > 4)	-1.387	0.103	0.000	-1.589, -1.186

The coefficients from these models along with the variance-covariance matrix were used to calculate the incidence rate ratios and 95% confidence intervals for BSF households versus control households in the two seasons. The IRR was calculated for both the dry season and the wet season for all three models. The IRRs from all the models are presented in table 5.12. The IRR and 95% CI for BSF households vs. control households was 0.40 (0.29, 0.55) and 0.80 (0.57, 1.20) during the dry and wet season, respectively, for the ordinary logistic regression model. GEE extensions of logistic regression produced a slightly higher estimate for IRR: 0.41 (0.29, 0.58) and 0.83 (0.59, 1.27) for the dry and wet season, respectively. The random intercepts logistic regression model IRR for BSF vs. control households in was 0.40 (0.25, 0.62) and 0.86 (0.50, 1.48) for dry and wet season, respectively.

Table 5.12 Incidence rate ratio of diarrheal disease in BSF households vs. control households, adjusted for categorical age of participants by season in the BSF RCT in Bonao, DR 2005-2006

Season	Ordinary Logistic Regression IRR (95% CI)	Logistic Regression with GEE IRR (95%CI)	Random Intercepts Logistic Regression IRR (95% CI)
Dry Season	0.40 (0.29, 0.55)	0.41 (0.29, 0.58)	0.40 (0.25, 0.62)
Wet Season	0.80 (0.57, 1.20)	0.83 (0.53, 1.28)	0.86 (0.50, 1.48)

The introduction of the interaction term decreases the precision of the IRR for both wet and dry season because it limits observation to only months classified as dry season months or wet season months. However, the results from all three models suggest the effect of the biosand filter intervention is greater during the dry season, with 60% reduction in incidence rates of diarrheal disease in BSF households as compared to control households. The effect in the wet season ranges from 20-14% reduction in incidence rates of BSF households compared to control households and the 95% confidence intervals cross the null value, which indicates that the difference between the incidence rates of diarrheal disease in the BSF and control groups during the wet season is not statistically different. Hence, the biosand filter has a significant protective effect against diarrheal disease during the dry season but this protective effect is diminished and appears to not be significant during the wet season because diarrheal disease rates in the control households decreased during the wet season period. The decrease in diarrheal disease rates is likely due to decreased risk from drinking water sources, drinking water quality and perhaps diarrheal disease transmission rates differ from those in the dry season. If more observations had been performed during the wet season, it is possible that a significant difference among the groups would have been found.

5.4 Discussion

Effect of BSF on Diarrheal Disease

This is the first known study to have performed a prospective, randomized controlled trial to determine the ability of the BSF to improve water quality and reduce diarrheal disease. The main finding from this study is that the presence of the biosand filter in households in Bonao, DR is associated with a decrease in *E. coli* in household drinking water and a decrease in incidence rates of diarrheal disease in BSF households as compared to control households.

It is important to note that due to the lack of a placebo biosand filter, the ability to determine whether or not the reduction of diarrheal disease was the result of underreporting of diarrheal disease by BSF households is not possible. This effect sometimes referred to as the placebo effect or the Hawthorne effect results when study participants underreport illness. While this is a weakness of the current study design, this similar study design has been employed in all but two or three studies of point-of-use household water treatment technologies. Additional research can and should be undertaken to determine whether or not the effect of the BSF was influenced by such a placebo effect.

The multivariate analysis found a 47% reduction in diarrheal disease rates in BSF households as compared to control households, when adjusted for participant's age and clustering. This finding of considerably less diarrheal disease in households with POU water treatment compared to households without such treatment is consistent with studies of other household water treatment technologies such as solar disinfection, chlorine disinfection and ceramic microfiltration, all of which have been found to reduce diarrheal disease from 30-70% in various field trials like this one (Arnold & Colford, 2007; T. Clasen et al., 2005; L. Fewtrell & Colford, 2005).

When diarrheal disease rates were higher in the control group during the dry season, the BSF was found to reduce diarrheal disease by 60%. The ability to measure an effect of the BSF on diarrheal disease risk was not as great during the wet season. In this season the ability of the BSF to reduce diarrheal disease was less than 20%, due to decreased rates of diarrheal disease in the control group.

Transmission of diarrheal diseases seasonally and fluctuations in diarrheal disease rates with season are not unique to this study. Often, an increase in diarrheal diseases is seen during an increase in rainfall or during wet weather events. This phenomenon was documented in Gambia, where researchers found an increase in diarrheal disease during summer rains (Rowland, 1986). However, other diarrheal disease transmission patterns are associated with the dry season. For example, rotavirus transmission was more effective during the hot dry months in one study in Kenya (Mutanda, Kinoti, Gemert, & Lichenga, 1984). Another study found a decrease in diarrheal disease after rainfall began in Thailand. Researchers found that while there was not a direct correlation with rainfall, diarrheal disease rates decreased after the summer rains began much like the effect in this study (Pinfold, Horan, & Mara, 1991).

In this current study, the effect of rainfall also appeared to result in reduced diarrheal disease after the rain began. This happened twice in the ten month study period; once prior to BSF intervention and once after BSF intervention. The fluctuation in diarrheal disease rates and the return of the diarrheal disease rates to higher levels in the dry season after a decline in the wet season suggests that the decreased diarrheal disease rates are not likely to be only an artifact of study fatigue, where households report fewer cases of diarrheal disease as study time increases. There is a possible study fatigue effect present; however this effect seems to be limited in magnitude and scope and rainfall or season seems to have a greater effect on rates of diarrheal disease.

There are many possibilities as to why increased rainfall resulted in decreased diarrheal disease rates. First, households may utilize rainwater for drinking water instead of other more contaminated sources. This results in improved water quality and therefore possibly decreased exposure to pathogens during periods of heavier rainfall. The utilization of rainwater may also result in larger quantities of relatively clean water available for use for other household needs such as hand-washing, cleaning or bathing; all of which may reduce exposure to diarrhea-causing pathogens. The opposite effect, namely increased risk of diarrhea, may occur during periods of decreased rainfall or dry seasons. Households that rely on rainwater for drinking may have to rely on other, more contaminated sources or they may have to store rainwater for extended periods. The increased storage time can result in degradation of rainwater quality (Wright et al., 2004). In addition, there may be overall less water available for use in households that rely on rainwater for a portion of their household water.

Effect of the BSF on Household Drinking Water Quality

Drinking water quality based on *E. coli* concentrations was better for BSF households as compared to control households, typically manifested as a lower *E. coli* concentration by

50% or even more. However, this reduction in *E. coli* concentration in water of BSF households is well below typical bacterial reductions documented by the BSF (Duke et al., 2006; Stauber et al., 2006). Unlike the water quality data of many other household water studies, the water quality measurements for BSF households included all water designated for consumption in BSF households and also all water designated for consumption in control households. In BSF households, this included water directly from the BSF outlet, stored BSF-treated water and other untreated sources, if households indicated it was being consumed without treatment. Likewise, drinking water from control households included both untreated water designated for consumption as well as treated water (including stored boiled and stored chlorinated water as well as purchased bottled water). Therefore, the estimates of reduction in *E. coli* concentrations by BSF treatment are likely to be underestimates of the actual bacterial reductions in water coming directly from the BSF treatment process. However, the measured *E. coli* concentrations of the various waters consumed in the households more accurately estimates the actual quality of the drinking water being consumed in both groups of households, BSF and non-BSF.

Over the six month BSF intervention period, BSF households had 50% fewer *E. coli* per 100mL in all months except for February and June 2006. During the water sampling period in February, BSFs had only been in use for an average of one week. Laboratory studies suggest that the BSF performance improves over time and therefore the microbial quality of water samples from the initial week after installation represent the performance of un-ripened BSFs. In June 2006, drinking water quality is poorer in both groups, suggesting some event that caused elevated *E. coli* concentrations in both groups. Unlike in April, where drinking water quality appeared to decline in control households as a result of high rain volumes, June had only 161 mm of rain. Perhaps the drinking water quality became deteriorated as a result of increased storage time for those who collect and store rainwater. Another possibility could be changes or problems in the water distribution system for the households that have access to some form of piped water. Overall the patterns of *E. coli* concentration and rainfall are difficult to explain, and it is likely that other factors may be influencing these relationships such as seasonal fluctuations in temperature and water quality that may promote growth or enhance survival of *E. coli* in drinking water.

Relationship between Drinking Water Quality and Diarrheal Disease in the Study

The relationships between drinking water quality based on fecal indicator microbes and diarrheal disease risk is not consistent among published studies and therefore is not easily interpreted. A recent meta-analysis on the relationship between point of use drinking water microbial quality and diarrheal disease risk did not find a direct correlation between increased diarrhea with increased levels of *E. coli* or thermotolerant coliforms in drinking water when tested at the point of use (Gundry, Wright, & Conroy, 2004; Wright et al., 2004). The use of indicator bacteria such as *E. coli* or thermotolerant coliforms does not always predict the presence of either bacterial pathogens or other, non-bacterial pathogens in drinking water. For example, in our study, the reduction of *E. coli* by the BSF likely underestimates the reduction of protozoan pathogens. This is because previous research has shown the BSF to be extremely effective at reducing *Giardia* and *Cryptosporidium* (by >99.9%) in laboratory studies (Palmateer et al., 1999). For viral pathogens, bacterial indicators like *E. coli* are unlikely to reliably predict the reductions of enteric viruses. This is because studies by our laboratory have shown that viruses are reduced less extensively (by about 90%) than are enteric bacteria like *E. coli* (typically by about 90-99%).

Another phenomenon observed for fecal indicator bacteria as a measure of drinking water quality is the potential for a “threshold effect;” where only when there are high concentrations (>1000/100 mL) in drinking is there a concomitant increase in diarrheal disease. This effect was documented in an observational study of water quality and diarrheal disease in the Philippines (Moe, Sobsey, Samsa, & Mesolo, 1991). In this research, higher rates of diarrheal diseases were associated with drinking waters with >1000 *E. coli* per 100mL but not with lower concentrations of *E. coli* in water. While the geometric means in this study are much lower than those found by Moe et al., 1991, households with the BSF rarely have geometric mean *E. coli* concentrations above 10MPN per 100mL, whereas in contrast, control household waters consistently have geometric means above 10 MPN *E. coli* per 100mL. Furthermore, it is possible that other non-measured confounders complicate the relationships between diarrheal disease risk and microbial water quality.

A major limitation of this study is the lack of a placebo BSF and the lack of blinding of interview staff. While the interview staff was trained to standardize the interview process and had visited households repeatedly prior to BSF intervention, there is a possibility that the interview process was different in BSF and control households after introducing the BSF intervention. In an attempt to limit interviewer bias in the diarrheal disease surveillance, the interview staff was supervised (accompanied by the author) at random times throughout the entire field study. While the lack of a placebo BSF is a limitation to the study, nearly all of the more than 20 other epidemiological field trials of household water treatment technologies in developing countries have also omitted the use of placebos, including those for solar disinfection, chlorine disinfection, coagulation-flocculation-disinfection product and ceramic microfiltration. Therefore, the results of this study can at least be compared in terms of observed findings to these other studies on household water treatment technologies.

Finally, it should be noted that it was difficult to measure filter use and compliance in BSF households. The reduced concentration of *E. coli* in drinking waters in BSF households suggests that households were using improved water and this was likely the result of the use of the BSF. However, there is no treatment-related indicator agent to measure in the treated water, as there is for example in chlorine intervention studies, where one can measure the free chlorine concentration in the water. Furthermore, the ability to generalize these results may be limited, perhaps just to this particular location and setting. For the BSF to be documented as robust and consistently effective as the other technologies, this type of field trial should be repeated in other locations and under other circumstances. Showing that the BSF improves water quality and reduces household diarrheal disease in other regions and countries and for other water sources and environmental conditions would further document that the results observed here are repeatable and generalizable.

Chapter 6: Impact of Biosand Filters on Household Drinking Water Quality and Diarrheal Disease

6.1 Introduction

The recent estimates of the global burden of disease suggest that 4% of the disease burden can be attributed to diarrhea (Pruss et al., 2002). The portion of diarrheal diseases that are attributed to environmental factors has been estimated at 94% (Pruss-Ustun & Corvalan, 2006). Many who suffer from this diarrheal disease burden in the developing world also lack access to clean water and proper sanitation. The high proportion of diarrheal disease attributable to environmental factors suggests that interventions in water, sanitation and hygiene have the potential to greatly reduce this burden by reducing the contribution from the environment. In an attempt to reduce diarrheal diseases transmitted by water, a number of household water treatment (HWT) technologies have been promoted to both improve microbiological quality of drinking water and reduce diarrheal disease (L. Fewtrell et al., 2005).

New HWT technologies are being developed for applications in developing countries. They range from chemical disinfection to filtration to combinations of both filtration and chemical disinfection. Recently, a laboratory study investigated a water purifier that combines carbon block filtration with disinfection. The unit was reported to remove 99.9999% of bacteria, 99.99999% of viruses and >99.9% of protozoan parasite surrogates. However, its performance has not yet been tested in the field (T. Clasen, Nadakatti et al., 2006).

Other HWT technologies have been field tested including: chlorine, solar disinfection combined coagulation-flocculation, disinfection and porous ceramic filters. In a randomized controlled field trial, researchers recently evaluated the efficacy of sodium dichloroisocyanurate tablets that deliver about 2 mg/l free chlorine to reduce thermotolerant coliforms in water (T. Clasen, Saeed, Boisson, Edmondson, & Shipin, 2007). While new technologies continue to be developed and evaluated, important gaps in knowledge still need to be filled for existing technologies that are already being used in the field.

The biosand filter (BSF) is a relatively recent technology that has not been the subject of rigorous research to document performance. Developed in the late 1980's and early 1990's, the BSF is a household scale intermittently operated slow sand filter. The BSF is similar to a conventional slow sand filter (SSF) in that there is a biologically active surface layer thought to provide much of its functionality in reducing contaminants in water. It typically has no pretreatment or backwashing and operation is simple in that water is added and gravity-driven through the medium rather than delivered by pump pressure filtration. Cleaning is by periodic scouring of the upper centimeters of sand by manually stirring to release impurities of the disturbed sand into the centimeters of water above the sand that is then removed by decanting. Like conventional SSFs, the sand bed remains wetted throughout operation and a ripening process occurs, during which a biolayer (or schmutzdecke) forms, head loss increases and performance in contaminant reduction improves.

Unlike a conventional SSF, however, the BSF does not operate continuously but instead, intermittently by delivering a charge of feed water (typically up to 20 L), although multiple daily charges are possible each day. Other differences of the BSF from that of the

SSF are that its filtration rate is up to 100 times greater than that for the SSF (1m/h in contrast to a recommended 0.08-0.4 m/h) (Fox et al, 1994). The depth of the BSF sand layer is also about 50% less than that for the SSF (0.4 m compared to a recommended starting depth of >0.8 m for the SSF with a minimum of 0.5 – 0.7 m). The particle size range of the BSF sand is typically broader than in SSF. For example, the uniformity coefficient of BSF sand may typically exceed 4.0, compared to a recommended value of <3 for SSF sand. In addition, BSF sand quality often differs because it is locally available and sized material, whereas most SSF sands are from a commercial source.

Since the BSF introduction into homes in the developing world, there have been only three peer-reviewed, published studies on its microbial reductions from water. In the earliest laboratory study, protozoan parasite reduction efficiency from seeded water was >99% for both *Giardia* and *Cryptosporidium* (Palmateer et al., 1999). In the other laboratory, the reduction efficiency of *E. coli* from seeded surface water improved over time with ripening and reached a maximum of 99% (Stauber et al., 2006). In the published paper on a field study, there was an average 98.5% reduction of *E. coli* by the BSF installed in homes in Haiti for an average of two years (Duke et al., 2006).

6.2 Objective

The objective of this study was to document the ability of the BSF to improve drinking water quality in households in Bonao, Dominican Republic. A total of 75 households that received a BSF for randomized controlled trial and a similar number of control households that did not receive a filter were monitored for household drinking water quality. Variations in drinking water quality were examined and an attempt was made to determine if there was a discernible relationship between diarrheal disease risk and drinking water quality over the entire study period of 39 weeks; both prior to and after randomization into BSF and control groups.

Longitudinal field water quality sampling began immediately following an initial cross-sectional survey in the two selected communities in Bonao, Dominican Republic described previously in this document. The field trial took place from September 19, 2005 to July 27, 2006, included 7 water quality sampling times prior to randomization and installation of the biosand filter and another 11 water quality sampling times after the BSF intervention was implemented.

6.3 Methods

Drinking Water Sampling and Analysis

At approximately two week intervals, households were asked to provide samples of drinking water being stored in the home. After completion of the household interview, interview staff collected approximately 500mL of water in sterile Whirl-Pak® bags (Nasco, Inc. Modesto, CA). If households reported using more than one source water, all sources being used were collected. After the BSF installation, BSF households were asked to provide a sample of the water prior to filtration, water directly from the BSF outlet, stored BSF-treated water, and if present, water that received treatment in addition to BSF treatment. All water samples were stored on ice to keep them cool and processed within 8 hours of collection. Water samples were analyzed for total coliforms and *E. coli* using the Colilert™ Quantitray system (IDEXX, Westbrook, Maine). Sample water pH, turbidity, and free and total chlorine were also analyzed using the following methods: pH with a Sension1 meter, turbidity with a turbidimeter 2100p and free and total chlorine by the colorimetric method using a pocket colorimeter II (all meters for pH, turbidity and chlorine analysis were provided by Hach, Loveland, CO).

Data Collection and Analysis

Comparison of Water Quality of the BSF and Control Households

All data were entered into Excel or EpiInfo and imported into Stata and GraphPad for additional analyses that could not be performed in Excel or EpiInfo. Household drinking water quality was compared for BSF and control households both prior to and after filter installation using parametric and non-parametric statistical tests. In addition, household drinking water qualities were compared for each sampling period to determine if there were variations in water quality with time. Control households and BSF households were compared for differences in the quality of drinking water collected from control households and drinking water directly from the BSF. For continuous variables, t-tests were used, for other tests Chi-squared test statistics were used.

Analysis of Stored BSF-Treated Water and Evaluation of its Recontamination

Stored BSF-treated water samples were collected from filter households during 8 of the 11 sampling periods after BSF intervention. The quality of these waters was compared to filtered waters taken directly from the filter outlet in all BSF households. In addition, the quality of stored BSF-treated drinking water of BSF households was also compared to the quality of water from control households.

Methods for Diarrheal Disease Surveillance

A system for diarrheal disease surveillance was established as part of weekly household interviews. At approximately 7 day intervals, the household's primary respondent was asked to verbally report cases of diarrheal disease for all participants in the household. Diarrheal disease surveillance began on September 19, 2005 and was completed on July 27, 2006. The study period before BSF installation for intervention phase consisted of 16 full weeks of household observation and the period after BSF installation for the intervention phase consisted of 23 weeks of household observation. All observations were included and intention to treat was used in the statistical analyses.

Multivariate Data Analysis

Multivariate logistic regression was used to control for multiple covariates. The multivariate analysis was performed in Intercooled Stata 8.0 (Stata, StataCorp, College Station, TX). All observations were made as interviews performed at the household level during each week that a representative from the household was available for interview. The outcome variable was cases of diarrheal disease for an individual. The main exposure variable was water quality: an ordinal variable where 0 = <10 *E. coli*, 1 = 10-99 *E. coli* and 2 = ≥ 100 *E. coli* per 100mL. The variables and their coding schemes are listed in Appendix 2. Because the observation period was only seven days and the incidence rates of diarrheal disease were relatively low (less than 0.10), the odds ratio (OR) produced from the logistic regression models were used to approximate the incidence rate ratios (IRR). Therefore, all IRRs reported are based on the ORs from the logistic regression.

Since household drinking water quality was not sampled at each household visit, drinking water quality values for each month of observation were averaged and that average was used as the measure of drinking water quality. Water quality was analyzed as both a continuous variable and a categorical variable (although not all analyses were reported). Initial analyses examined water quality as a continuous variable using log10 of E. coli/100mL as the continuous variable. In addition, ordinal variables were created based on 2-5 distinct categories of E. coli concentrations. Ultimately, a three level ordinal water quality variable was selected for the main exposure variable. A list of all variables considered in the model building procedure is listed in Appendix 2.

The final model for the multivariate analysis was built using a three level ordinal water quality variable as the exposure and the outcome was cases of diarrheal disease. The model was developed using a backward elimination strategy where covariates were fitted into the model and eliminated based a 10% a priori change in the coefficient of exposure. Clustering was taken into account by using generalized estimating equations extensions of logistic regression and random intercepts logistic regression.

6.4 Results

A total of 4673 water samples were processed and analyzed during the longitudinal study; 1567 samples prior to the BSF intervention and 3106 samples after BSF intervention. A total of 1807 water samples were from control households, of which 1044 were collected after BSF intervention. A total of 2781 samples were collected from households given a biosand filter (intervention) in February 2006, of which 2060 were collected after the BSF installation. Data on types and numbers of drinking water samples are summarized in table 6.1. Controls households reported approximately 30% of all water samples provided were exposed to some form of treatment. Prior to BSF intervention, BSF households reported approximately 30% of all households exposed to some form of treatment; however after BSF intervention that proportion increased to 65%.

Table 6.1 Types and numbers of drinking water samples collected during the RCT of the BSF in two communities of Bonao, Dominican Republic

Type of Sample	Prior to Intervention N (%)	After Intervention N (%)
Control households - all	**N = 763**	**N = 1044**
- untreated	536 (70)	735 (70)
- stored treated	227 (30)	309 (30)
- treated by boiling	171 (22)	240 (23)
- treated by chlorination	50 (6.5)	48 (4.6)
- treated by other	6 (0.9)	21 (2.0)
Filter household – all	**N = 721**	**N = 2060**
- untreated	490 (68)	718 (35)
- treated	231 (32)	1342 (65)
- treated by boiling (only)	169 (23)	35 (1.7)
- treated by chlorination (only)	60 (8.3)	1 (< 0.1)
- treated by other	2 (0.2)	0 (0.0)
- treated by BSF directly		796 (39)
- treated by BSF and stored		506 (25)
- treated by BSF, boiled, stored		114 (6)

Drinking Water Sources and Water Quality

Of the total water samples collected in households, some were not treated prior to consumption. In control households, 70% were not treated prior to consumption, while in BSF households the percentage not treated before consumption is much lower (but not reported). There were six different sources of drinking water used in the two communities: piped, well, rain, spring, bottled and river water. Presented in table 6.1 is the geometric mean monthly *E. coli* MPN/100mL for each untreated water source as a function of time, along with data on monthly rainfall over the study period (10 months).

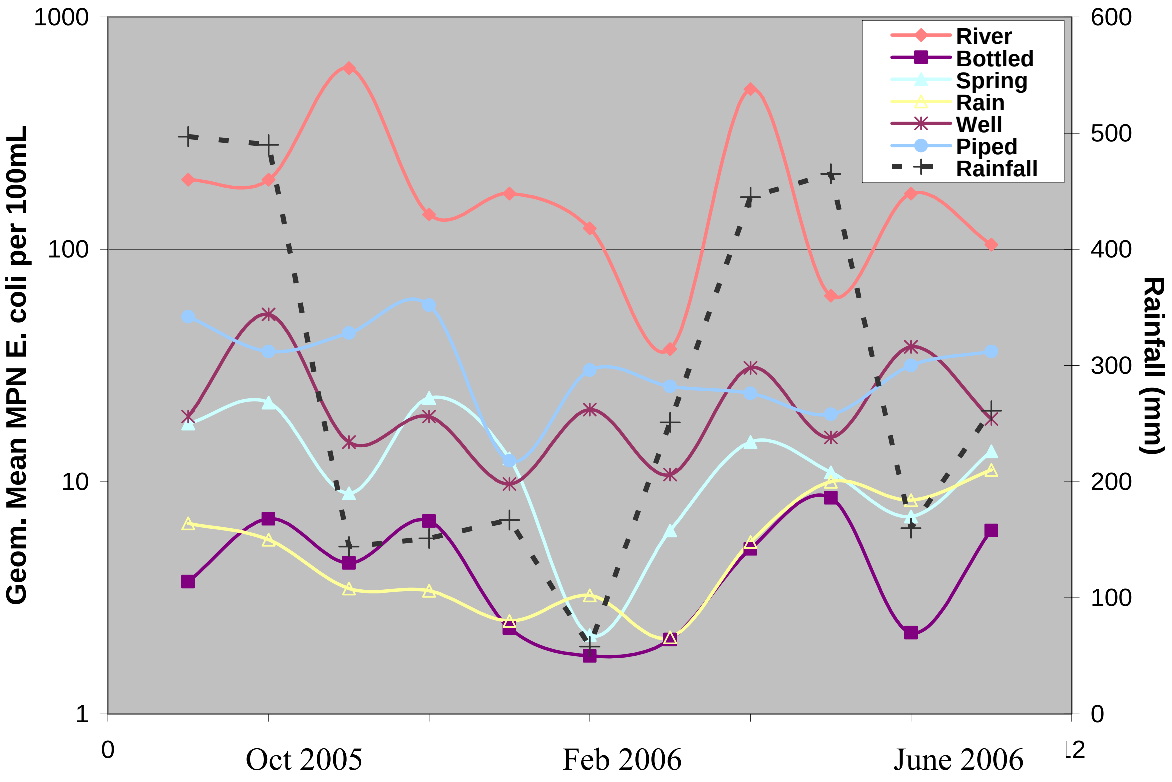


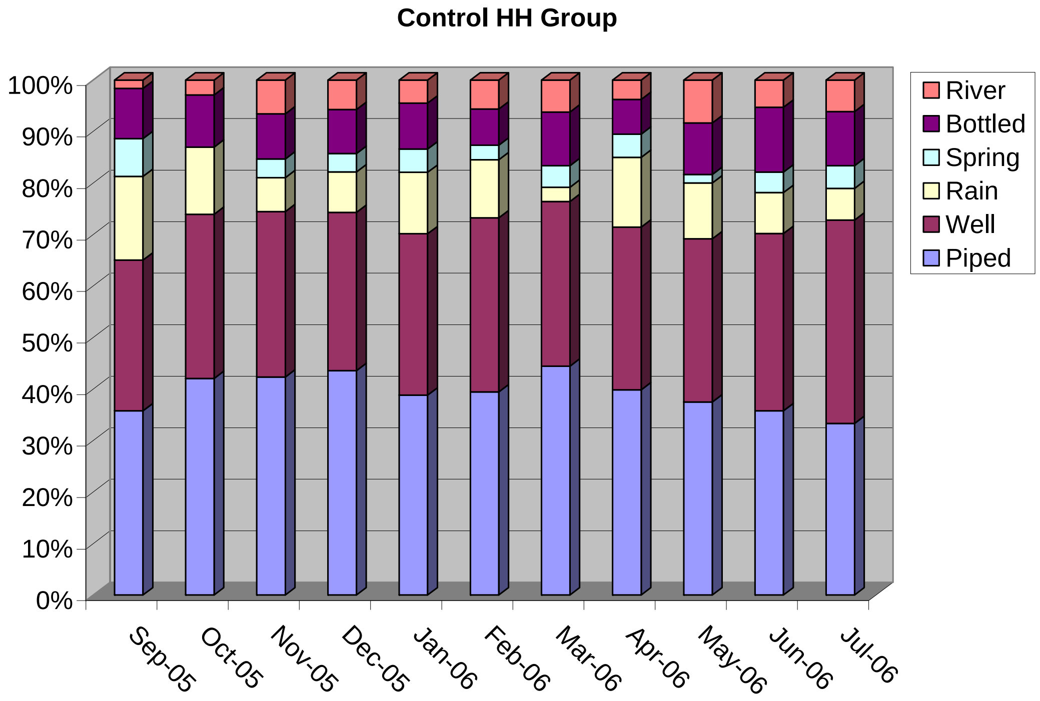
Figure 6.1 Average E. coli/100mL in six sources of household water during the longitudinal study period in Bonao, Dominican Republic.

As shown in figure 6.1, the two least contaminated water sources on the basis of *E. coli* concentrations per 100 mL were bottled water and collected rain water. These waters had <10 *E. coli*MPN/100mL for almost all ten months of the study period. The most contaminated drinking water source was river water, with monthly averages >100 MPN *E. coli*MPN/100mL for eight of the ten months. Piped water, well water and natural spring water were intermediate in quality, with 10-100 MPN *E. coli/*100mL for the entire study in piped and well water and for 7 of 10 months for natural spring water. The order of *E. coli* contamination of the different sources from lowest to highest contamination was: bottled, rain, spring, well, piped, river.

Temporal variations in water quality based on *E. coli* concentrations were compared with average monthly rainfall amounts. During BSF intervention period, April 2006 had a high amount of rainfall after a 4-month period with relatively little rainfall. The increased rainfall period corresponded to increased *E. coli* concentrations in both of the surface water sources (river and spring) of drinking water. The *E. coli* concentration in both the well water and piped water varied between 10 and 100 MPN/100mL. However, the temporal pattern of *E. coli* fluctuation in these waters did not appear to be the same pattern as changes in rainfall amounts.

Because *E. coli* concentrations in drinking water varied among the different water sources, water source was an important factor influencing the average *E. coli* concentrations in household drinking water. At each month, the total proportion of water samples from each of the six water sources was determined for both control and BSF households. These data are presented in figure 6.2. BSF and control households relied on some form of piped water for approximately 40% of their untreated source waters prior to BSF intervention. After BSF intervention, the proportion of piped water usage increased in BSF households to 50% upon introduction of the BSF and to >50% for the month of June 2006. Prior to BSF intervention, well water accounted for 20 and 30% of their water samples respectively in BSF and control households. After BSF intervention, reliance on well water increased slightly from 20% to 30% in BSF households. Spring and river water together represented 10% of all untreated drinking water samples for both BSF and control households. Rain water usage accounted for 5-20% of untreated drinking water samples. The proportion of rain water samples increased in September and October 2005 as well as in April and May 2006 for both BSF and control households. Monthly rainfall amounts were >400mm for each of those four months.

The largest difference in water types between BSF and control households prior to BSF intervention was for bottled water. Prior to BSF intervention, 15-20% of BSF household untreated drinking water samples were from a bottled water source, while for control households it constituted 10% of untreated water samples. After BSF intervention, the proportion of samples from bottled sources decreased from about 20 to 4%, while in control households it remained at approximately 10%. In summary, the main differences in sources of untreated waters in BSF households after the BSF intervention, were fewer bottled water samples and somewhat more samples from piped supplies and wells, compared to the pre-intervention period in BSF households.



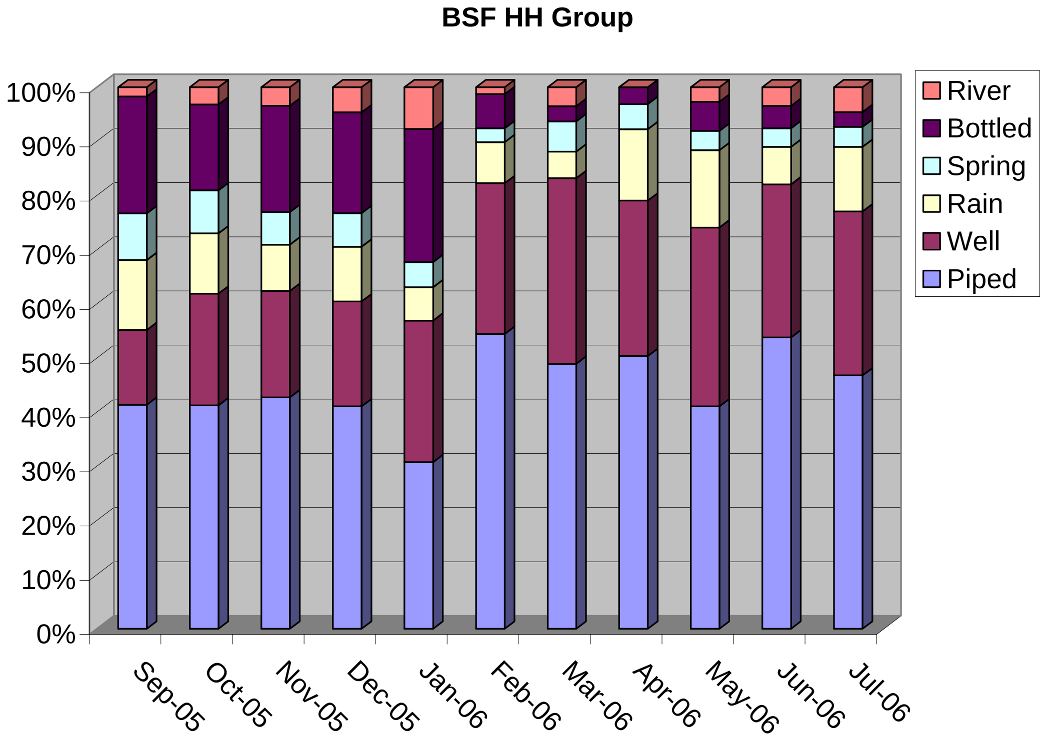


Figure 6.2 Percentage of drinking water sources used by BSF and control households

Table 6.2 Distribution of drinking water sources before and after BSF intervention in randomized controlled trial in Bonao, Dominican Republic

variable	**Before BSF**		**After BSF**	
	Control HH %	**BSF HH %**	**Control HH %**	**BSF HH %**
Piped	41	40	39	49
Well	31	20	33	30
Rain	11	10	8	10
Spring	4	7	4	4
Bottled	9	20	10	4
River	4	3	6	3

Drinking Water Quality in BSF and Control Households for *E. coli*

Geometric and arithmetic mean *E. coli*MPN/100mL values for BSF and control households were compared before and after the BSF intervention. All household drinking water samples designated for consumption were averaged for each phase of the study (prior to and after BSF) and compared by two sample t-tests. Neither geometric nor arithmetic mean *E. coli* concentrations were statistically different between BSF and control households before the BSF intervention. However, there was a statistically significant difference after BSF intervention, where the BSF households had decreased geometric and arithmetic mean concentrations of *E. coli* and total coliforms as compared to control households. The data are summarized in table 6.3. After the BSF intervention, average drinking water quality is improved in BSF households compared to control households.

Table 6.3 Drinking water quality prior to and after installation of the biosand filter in Bonao, Dominican Republic

Variable	**Groups**		
	Control HH	**Intervention HH**	**Total**
Arithmetic mean *E. coli* MPN/ 100mL prior to BSF	N = 763 160	N = 721 155	N = 1484 157
Geometric mean *E. coli* MPN/ 100mL prior to BSF	N = 763 16	N = 721 14	N = 1484 15
Arithmetic mean *E. coli* MPN/ 100mL after BSF§	N = 1044 *182	N = 1644 113	N = 2682 140
Geometric mean *E. coli* MPN/ 100mL after BSF	N = 1044 *12	N = 1644 7.2	N = 2682 8.7
Arithmetic mean coliform MPN/100mL prior to BSF	N = 763 1333	N = 721 1262	N = 1484 1298
Geometric mean coliform MPN/100mL prior to BSF	N = 763 *589	N = 721 437	N = 1484 513
Arithmetic mean coliform MPN/100mL after BSF	N = 1038 *1804	N = 1644 1352	N = 2682 1527
Geometric mean coliform MPN/100mL after BSF	N = 1038 *891	N = 1644 490	N = 2682 617

* - indicates significant difference between the BSF and control groups by two sample t-test

§ - Averages included all water designated for consumption in BSF and control households. For control households this includes treated and untreated; in BSF households this includes untreated but consumed, treated and stored, and water directly from the filter

Average drinking water *E. coli* concentrations in control and BSF households, while statistically significantly different; suggest only modest *E. coli* reductions by the BSF. In order to better understand *E. coli* reductions by the BSF and potential exposure levels to contaminated drinking water, household drinking water quality was further examined during the BSF intervention period. All samples were stratified into the following categories: untreated, treated and stored, direct from BSF outlet, and BSF-treated followed by boiling and storage. The source-stratified *E. coli* data for BSF and control households are presented in tables 6.4 and 6.5 respectively. In table 6.6 the results from two sample mean t-tests are presented for all of the possible comparisons for the various drinking water concentrations of *E. coli*. Values indicate whether or not there was found to be a statistically significant difference between the mean values. If $p < 0.05$, the difference between the two means was considered significantly different, when $p > 0.05$, there was no statistically significant difference.

Of the 21 individual comparisons made, only six results found no statistically significant difference among the comparison groups. Those six results were for the following comparisons: BSF treated direct from outlet and BSF-treated boiled stored; BSF-treated direct from outlet and control boiled stored; BSF-treated direct from outlet and control chlorinated, stored; BSF-treated boiled stored and control boiled stored; BSF-treated boiled stored and control chlorinated stored; and control boiled stored and chlorinated boiled stored. Of the comparisons that were found to be statistically significant, important to note was that geometric mean untreated drinking water *E. coli* MPN/100m was 17 and 21 for control and BSF households, respectively. BSF households had a higher initial concentration of *E. coli* in untreated drinking water. *E. coli* concentrations in water directly from the BSF were lower by 79%, compared to untreated drinking water in BSF households (also a statistically significant difference). When stored BSF treated waters were compared with stored treated control waters and BSF treated waters directly from the outlet, stored treated (boiled or chlorinated) control waters and BSF treated waters direct from the outlet were found to have significantly lower *E. coli* concentrations as compared to those in stored, BSF-treated drinking water (4.9, 4.6 and 10 MPN *E. coli*/100mL respectively). Finally, stored, BSF-treated, boiled drinking water had a significantly lower *E. coli* concentration of 5.2 MPN/100 mL as compared to stored BSF-treated water (unboiled) at 10 MPN/100 mL. Despite statistically significant differences among some of these BSF treated waters, all were in the WHO low risk category of 10 or fewer *E. coli* per 100 mL.

Table 6.4 Water quality in BSF households after BSF intervention

Type of Sample	**Number (%) of Samples Total n = 2060**	**Geometric Mean *E. coli* MPN/100mL (SD)**	**% Reduction (from untreated water)**
Untreated	718 (35)	21 (11.7)	-
Treated – direct from BSF outlet	682 (33)	4.6 (6.5)	79
Treated – stored BSF treated	506 (25)	10 (8.3)	53
Treated – stored, BSF treated boiled	114 (5)	5.2 (9.5)	76
Treated – boiled, chlorinated or other but not BSF treated	40 (2)	2.5 (4.9)	88

Table 6.5 Water quality in control households after BSF intervention

Type of Sample	**Number (%) Total n = 1044**	**Geometric Mean *E. coli* MPN/100mL (SD)**	**% Reduction (from untreated water)**
Untreated	735 (70)	17 (10.7)	-
Treated (boil, chlorine, other)	309 (30)	4.9 (8.5)	72
-boiled	240 (25)	5.3 (8.7)	70
-chlorinated	48 (5)	3.8 (7.1)	78
-other	21 (2)	5.1 (9.7)	71

Table 6.6 Statistical comparison of geometric means for BSF and control household drinking waters

Type of water	BSF-treated direct	BSF-treated stored	BSF-treated, boiled, stored	Control untreated	Control boiled, stored	Control chlorinated stored
BSF untreated	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$
BSF-treated direct	-	$p < 0.05$	$p = 0.40$	$p < 0.05$	$p = 0.19$	$p = 0.41$
BSF-treated stored	-	-	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$
BSF-treated, boiled, stored	-	-	-	$p < 0.05$	$p = 0.92$	$p = 0.36$
Control untreated	-	-	-	-	$p < 0.05$	$p < 0.05$
Control boiled, stored	-	-	-	-	-	$p = 0.30$
Control chlorinated stored	-	-	-	-	-	-

E. coli concentrations in BSF and control household drinking waters were examined and compared at month intervals for the six-month intervention study period and they were also compared to rainfall patterns (Figure 6.3). Geometric mean monthly *E. coli* MPN values of stored, BSF-treated drinking water were >10 MPN/100mL for June 2006 but were <10MPN/100mL for the other three months sampled. *E. coli* concentrations were highest during June 2006 for all categories of sampled water. Levels of monthly average *E. coli* concentrations did not correspond well with high or low monthly rainfall but did tend to fluctuate in periods of high rainfall. Overall, BSF households had water of higher quality for *E. coli* compared to control households, even though untreated drinking water quality suggested higher concentrations of *E. coli* in BSF households compared to control households.

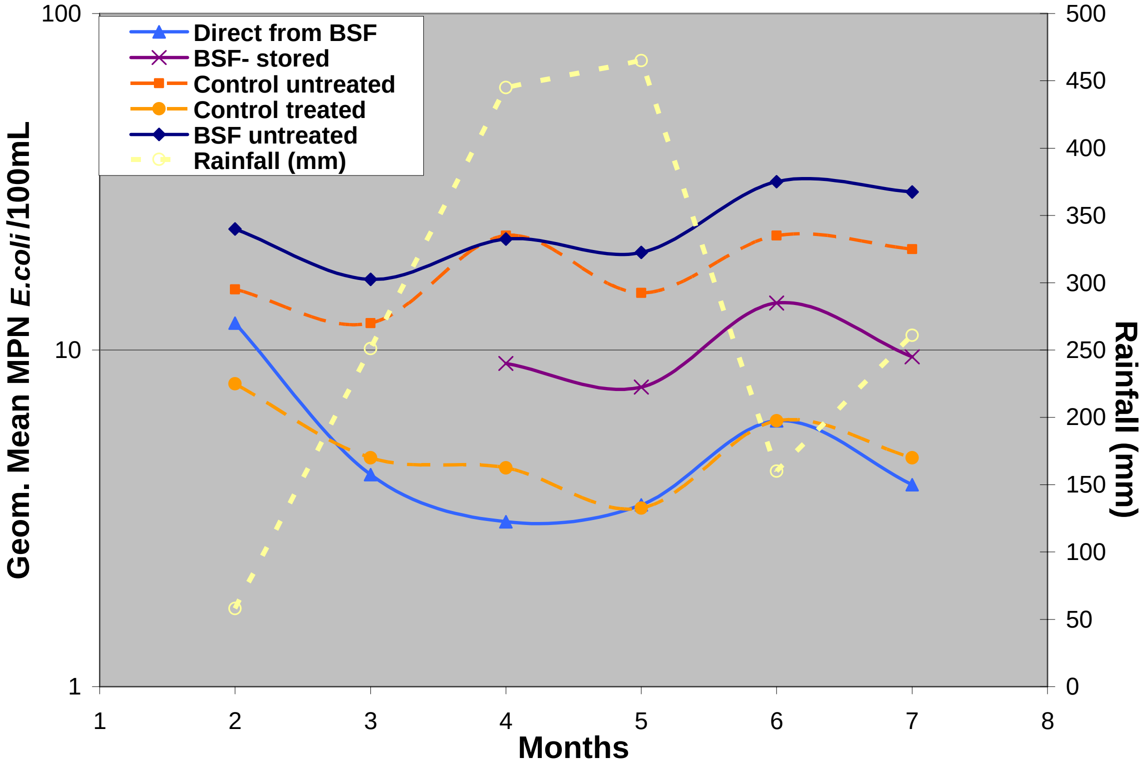


Figure 6.3 Comparison of drinking water quality from control and BSF households during BSF intervention

To determine whether or not a higher proportion of BSF households compared to control households had water that met World Health Organization guideline of 0 *E. coli*/100mL, household drinking water was divided into order of magnitude categories of *E. coli* concentration for both household groups. These data are presented in figure 6.4 and compared both prior to and after BSF intervention for both groups. BSF households had a slightly higher proportion of samples that had <1 E. coli MPN/100mL (28%) as compared to control households (25%) after intervention. BSF households had a significantly higher proportion of samples with <10MPN *E. coli*/100mL (65%) compared to control households (49%). Overall, BSF households had statistically significantly different proportion of samples ($p < 0.05$) at every decimal category of *E. coli* concentrations than did control households, except for samples with <1 E. coli MPN/100mL.

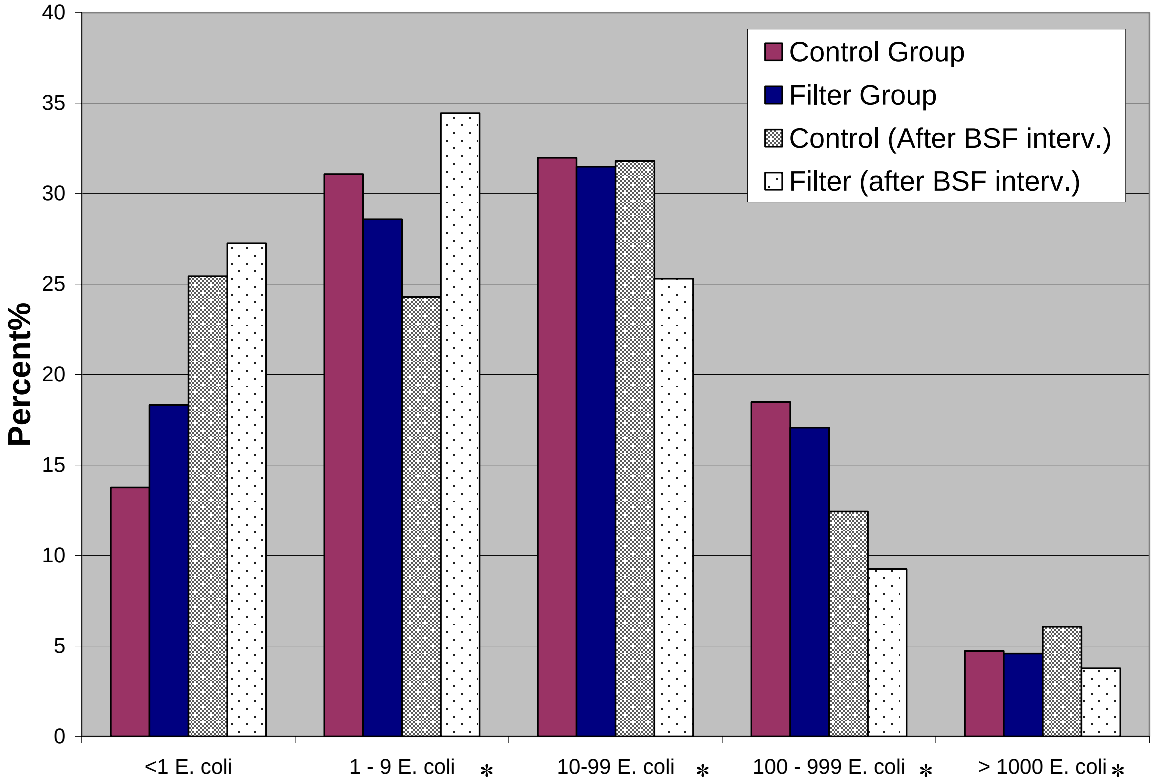


Figure 6.4 Percent of drinking water contamination for BSF and control households for before and after BSF intervention by concentration of E. coli
* indicates a statistically significant difference among BSF and control groups after BSF intervention

Relationship between Diarrheal Disease and Drinking Water Quality

In order to determine whether or not there was a relationship between improved drinking water quality for *E. coli* and decreased diarrheal disease risk, household diarrheal disease rates were compared to *E. coli* concentrations as a measure of drinking water microbial quality for all households and for the entire study period. Drinking water quality was categorized into three groups based on geometric mean monthly *E. coli* MPN per 100mL: <10, 10-99, ≥100. An ordinary logistic regression model was used to examine the relationship between the ordinal indicator of water quality based on categorical *E. coli* concentration and diarrheal disease cases over the entire study period. Based on this model, the unadjusted odds ratio for drinking water quality was 1.07 (95% CI 0.98, 1.17). This suggests that there were slightly increased risks of diarrheal disease associated with 10-fold increases in *E. coli* concentrations but that this was a weak association and not statistically significant because the 95% confidence interval crossed the null value.

The model was then adjusted for clustering using either GEE or random intercepts logistic regression and the other covariates that were identified to be confounders of the relationship between *E. coli* water quality and diarrheal disease: age, community location and season. Results from the model are shown in Table 6.7. Based on the results from the random intercepts logistic regression model, the odds ratio was 1.08 (95% CI 0.97, 1.20). The model again suggests a small (8%) but non-significant increased risk of diarrheal disease associated with each 1-unit change in the exposure variable (10 fold increase in *E. coli* concentrations). When the relationship was examined for the BSF intervention period only, the odds ratio was 1.11 (0.98, 1.28), again suggesting an increased but not significant effect of a positive association of increased *E. coli* with increased diarrhea risk during the intervention period.

Table 6.7 Odds ratios and 95% confidence intervals from results of multivariate model using ordinary logistic regression, GEE extensions of logistic regression and random intercepts logistic regression

Variable	**Ordinary**	**GEE**	**Random Intercepts**
Ordinal water quality	1.08 (0.99,1.18)	1.08 (0.98,1.20)	1.08 (0.97,1.20)
Season	1.19 (1.02,1.38)	1.19 (1.02,1.39)	1.21 (1.03,1.42)
Community	1.86 (1.59,2.16)	1.86 (1.49,2.32)	1.89 (1.39,2.56)
Categorical age	0.26 (0.24,0.29)	0.28 (0.25,0.32)	0.24 (0.21,0.28)

6.4 Discussion

Source Water Contamination and Distribution of Source Water in Households

Households of the two communities of this study used several different untreated sources waters, ranging from piped water to local river water. Notably, piped water had high levels of *E. coli* contamination and was second only to river water in *E. coli* contamination levels. The piped water has an important impact on household drinking water contamination because almost 40% of all household water samples were stored piped water. Source water quality exhibited frequent fluctuations in quality based on *E. coli* concentrations throughout the study. Increased *E. coli* concentrations correlated with rainfall for the two surface water sources of rivers and springs. This is possibly due to a “first flush” effect where high rainfall after prior dry periods increases runoff and introduces pollutants into surface waters. However rainfall data were available only as monthly values, and therefore, we can only speculate on the potential for this first flush effect associated with individual precipitation events. Furthermore, while piped water and well water quality fluctuated over the study, this did not correlate with rainfall amounts. This lack of correlation suggests other effects could have been important determinants of water quality for these sources.

The utilization of the various source waters by households were affected by two main events: rainfall amounts and the biosand filter intervention. In months of higher rainfall, a higher proportion of households used rain water which was of relatively good microbial quality throughout the study. This likely decreased waterborne exposure to higher concentrations of *E. coli* and related fecal pathogens, which thereby could have affected diarrheal disease risks in the households during or following months of higher rainfall. The introduction of the BSF into households changed the proportion of households relying on piped and bottled water, resulting in a significant decrease in bottled water and a significant increase in piped water. It is likely that households with the BSF now chose to treat their water with the filter and stopped or reduced purchase of bottled water. The BSF-treated water direct from the filter had levels of *E. coli* similar to those levels found in untreated bottled water with <10MPN E. coli/100mL. This low level of *E. coli* suggests that these households were not at risk for high level pathogen exposures, as a result of having changed drinking water sources. This change in water source also likely resulted in decreased expenses for BSF households, because bottled water was approximately $1US per five gallon bottle.

Average Drinking Water Quality and BSF Intervention

Overall, average *E. coli* concentrations in drinking waters were relatively low with no monthly average >1000 *E. coli* MPN/100mL. The relatively low concentrations of *E. coli* in untreated drinking waters may have hindered the ability to quantify *E. coli* reductions by the biosand filter, due to low *E. coli* levels detectable in both feed and product water. However, even with relatively low *E. coli* concentration in untreated waters, the BSF improved microbial quality over the entire six month study period. Drinking water taken directly from the BSF had a mean of 5 *E. coli* MPN/100mL for the six month study period and a mean of <10 *E. coli* MPN/100mL for all drinking water samples throughout the entire intervention phase of the study. There was a statistically significant improvement in overall water quality for *E. coli* for BSF households, with a significantly lower percentage of samples having >10 *E. coli* MPN/100 mL, compared to control households.

The results of this study document the potential for stored BSF-treated drinking water to become re-contaminated after filtration. Comparison to stored treated water from control households, stored BSF-treated water had increased concentrations of *E. coli*. The lack of the presence of a residual disinfectant makes the BSF-treated water vulnerable to *E. coli* re-growth and post-filtration contamination in the storage container. Despite increased concentrations of *E. coli* in stored water after filtration, stored BSF-treated water remained relatively high quality with average *E. coli* concentrations of <15 MPN/100mL during a 4-month observation period. Because *E. coli* recontamination was at a relatively low level, with concentrations well below 100 MPN/100 mL, the BSF appears to be an important technology to improve drinking water microbial quality, at less cost than boiling, which is also subject to recontamination after treatment and has the additional expense for fuel.

Drink Water Quality and Household Diarrheal Disease

In an effort to understand the relationship between diarrheal disease and household drinking water microbial quality, generalized estimating equations extensions and random intercepts logistic regression were used to model the relationship between an ordinal variable of water quality in relation to diarrheal disease rates. The multivariate modeling suggested a relationship between *E. coli* concentrations in drinking water and diarrheal disease rates, but it was a relatively weak relationship, with 95% confidence interval crossing the null value and $p = 0.10$ for the coefficient of exposure. When adjusted for age, season and community, risk of diarrheal disease when drinking water had 10-99 *E. coli*/100 mL was 1.08 (95% CI 0.98 – 1.27) times the risk from water with <10 *E. coli*/100mL.

The relatively low concentrations of *E. coli* in drinking water, low rates of diarrheal disease and lack of association between *E. coli* and reductions in diarrheal disease deserve attention. While the relationship between *E. coli* and diarrheal disease is weak at best, the lack of the association in this study suggests that further research should be performed. Existing data from the current study and additional research can and should be undertaken to determine whether or not the lack of a relationship between *E. coli* and diarrheal disease can be the result of underreporting of diarrheal disease by participants that received the BSF during the intervention period. Since there was not a placebo filter employed in the study, the possibility of a placebo effect can not be ruled out and needs to be further investigated.

The relatively week relationship between *E. coli* concentration in water and diarrheal disease has been documented in previous studies. In a year-long prospective cohort in Pakistan, researchers were not able to find a statistically significant association between *E. coli* concentrations in household drinking water containers and diarrheal disease (Jensen, Jayasinghe, van der Hoek, Cairncross, & Dalsgaard, 2004). Furthermore, in a meta-analysis on household drinking water quality and diarrheal disease, there was also no significant association between indicator bacteria concentration and diarrheal disease (Wright et al., 2004).

There are many plausible explanations as to why concentration of *E. coli* is not always an accurate indicator or predictor of risk of diarrheal disease. As mentioned previously, recent estimates suggests that 94% of diarrheal disease can be attributed to environmental factors. These environmental risk factors for exposure are not limited to water alone and can also be related to pathogen exposures through poor sanitation, hygiene and person-to person contact. The limited focus of this study did not include taking these various factors into account through rigorous interventions and therefore, non-water related environmental exposure factors may confound the analysis.

It is noteworthy that *E. coli* is perhaps not a reliable predictor for the reduction of other potential pathogens of diarrheal disease in the household waters of this study. For example, the active mechanisms of microbial reduction in the BSF may differ among pathogen types or classes, such that indicator bacteria such as *E. coli* may not be reliable indicator of the risks of all types of microorganisms. The reductions of protozoan parasites in laboratory studies are a good example. Researchers previously found >99% reduction of *Giardia* and *Cryptosporidium* but relatively low reductions (83%) of heterotrophic bacteria in the same study. Hence, it is likely that *E. coli* do not accurately estimate reductions of protozoan parasites or viruses in this field study and nor the impacts of their reductions on waterborne diarrheal disease risks.

A major limitation of the study is the limited ability to reliably measure compliance with the BSF intervention at the household level. Unlike field trials of chemical disinfectants or even solar disinfection, there are currently no techniques to detect whether or not households actually treated the water with the BSF or just reported such treatment to the interview staff without really doing it. It is possible that *E. coli* reductions by the BSF are underestimated because households reported treating water with the BSF when the water actually remained untreated. Such misclassifications of household treatment have been observed in previous studies on chlorine and the combined coagulant-flocculent-disinfectant, for example. We suspect that there is probably a small portion of households that provided stored filtered water samples that were actually not BSF-treated. However, this effect is not important in the overall ability to relate microbial water quality or health outcomes in this study because additional water samples collected directly from the BSF faucet made it possible to compare filtered water quality and unfiltered water quality based on *E. coli* levels to diarrheal disease risks

Overall, the BSF technology improved household water quality, there was only relatively low level post-filtration contamination, and there was weak but positive evidence of reduced diarrheal disease risks from improved water quality such as the improvements in BSF-filtered household water compared to unfiltered household water.

Chapter 7: Summary and Discussion of Research

7.1 Summary of Significant Results

The research presented in the preceding chapters represents a significant contribution to the knowledge of the microbiological effectiveness and the human health impact of the biosand filter for drinking water. The most important findings from the research can be grouped into three categories: laboratory evidence for improving drinking water quality, field performance in improving drinking water quality and human health impact in the field.

Laboratory Evidence

The most significant results from the laboratory research were:

1) This is the first study to document the reduction of *E. coli* and two bacteriophages, MS-2 and PRD-1, from seeded feed water at the same time in the same filter. The results suggest that the BSF is more effective at reducing bacteria than bacteriophages, although reductions of both classes of microbes were observed. The average reductions during the experiment were 90, 65 and 61% for *E. coli*, MS-2 and PRD-1 respectively (averaging seven time points during the 43-day experiment). However, the range of reductions was large for both bacteria and bacteriophages. *E. coli* reductions ranged from as low as 63% to a maximum reduction of 99%. The range of reductions for viruses was also large ranging as low as 10% to as high as 87%. Results also indicate that initial reductions of bacteria are

moderate but they improve over time due to some form of biological ripening of the filter. Average reductions in an unripened filter were 67% and 97% in a ripened filter. While the laboratory experiments suggested a ripening behavior similar to that for slow sand filtration, microbial reductions do not reach levels reported for conventional slow sand filtration.

2) This was the first study to document the effect of water volume filtered and residence time of water in the filter bed on reductions of bacteria and viruses. The results suggest that water volume dosed per unit time plays an important role in BSF reductions and may be an important management or operation tool to enhance microbial reductions in field BSFs. More specifically, there appears to be an increased reduction of microbes from water that is in contact with the BSF filter bed for the longest time. Water that spent time overnight in the filter had 0.3-0.5 higher $\log_{10}$ reductions as compared to water that passed through the filter the same day for bacteria and bacteriophages. The microbial quality of water that has remained in the filter overnight and is collected as filtrate separately has far better microbial quality than water that has not remained in the filter overnight and is discharged as filtrate after the overnight water.

Field Performance of BSFs for Improving Water Quality

The most significant results from the field research on filter performance were:

3) This was one of the first studies to document BSF performance in the field for newly installed filters that were monitored longitudinally for 6 months.

The results suggest that BSFs are effective at reducing at reducing *E. coli* and coliforms in water and that the extent of reduction can be better documented when there are high rather than low concentrations of *E. coli* in the feed water. Initial reductions after filter installation were as low as 49% and reached a maximum of 92% but averaged only 80% for the entire six month study. The apparent greater *E. coli* reduction when feed water *E. coli* concentrations are high is probably an artifact created by the opposite result, which is a lower *E. coli* reduction caused by the lack of *E. coli* detection in filtrate water when feed water *E. coli* concentrations are low. However these results suggest that field filter performance is perhaps best judged by BSF-treated water quality and not necessarily microbial reductions. The BSF produces filtrate water with <10 E. coli per 100 mL most of the time, which according to WHO guidelines, is considered of low health risk. Households with the BSF filter had > 65% of samples with less than 10 *E. coli*/100mL for water directly from the filter and stored filtered water during the intervention period.

4) There was some degree of BSF ripening of newly installed filters in the field after 6 months of operation, as evidenced by slightly decreased flow rates and improved *E. coli* reductions with increasing time since installation. However, the rate of ripening in these BSFs in Bonao, DR was not nearly as rapid as in laboratory studies of the plastic BSF.

5) The BSF treatment resulted in significantly improved drinking water in households compared to the untreated water applied to the filters, but there

was high potential for recontamination during storage of BSF treated water. On average stored BSF-treated waters had 10 MPN *E. coli*/100mL as compared to 5 MPN *E. coli*/100mL in water directly from the BSF.

Health Impact

The most significant results from the health impact research were:

6) This was the first rigorous prospective randomized controlled trial to document reduction in diarrheal disease in BSF households compared to households without BSF filters and using prevailing household water management practices in the study communities. There was a 47% reduction in diarrheal disease in BSF households compared to control households over the entire six month period of BSF use.
7) Over six months, user compliance remained high with >90% of households still using the BSF by the end of the 6-month intervention period.

Research Limitations and Recommendations for Future Research

This research has begun to fill significant gaps in the knowledge about biosand filter performance for water quality improvement and reduction of waterborne disease risk; both under controlled laboratory conditions and in the field. However, there are also some limitations to the research. The laboratory studies were done with a plastic version of the BSF yet at the time of this research the majority of BSFs in field use were made of concrete. More research on the concrete BSFs in the laboratory could help to determine if they perform the same as concrete filters in the field and to better understand and interpret the results from field studies of the BSF. It is also possible that the laboratory research created artificial conditions not representative of BSF use in the field. Perhaps laboratory and field studies comparing more realistic operating conditions in the field, such as varying dosing frequency, fluctuation in volume applied, and changes in water quality, would further enhance the knowledge on how these operational practices influence microbial reduction by the BSF.

The field research on both the water quality performance of the BSF and the health impact, suffer from lack of generalizability. The BSFs and the participants who were part of the randomized controlled trial represent a specific set of conditions and factors that are unique to the communities in Bonao, Dominican Republic and do not encompass the range of conditions under which people collect, treat and use household water. For example, the relatively low levels of *E. coli* in many of the drinking source waters may have resulted in an underestimation of microbial effectiveness of the BSF. It is also likely that there is an underestimation of health impact, because pathogen health risks and the magnitude of their reductions are dependent on pathogen levels in water and waterborne exposure risk to pathogens from both untreated and BSF-treated water, as well as the types of pathogens present in untreated and BSF-treated water. However, this limitation is not restricted to this study alone but is a limitation of all studies that take place in only one location at one point in time. Because the study results are not generalizable, there is a need to perform these studies in various settings to determine the robustness of the technology for a range of populations and their infectious disease burden, sanitation, hygiene, water quality and quantity, and environmental conditions.

Another important weakness of the health impact study is the lack of a placebo BSF and the ability to compare the placebo group to the actual BSF group in a masked (blinded), randomized controlled trial. There is no simple way to eliminate the possibility of a placebo effect. Previously performed studies on POU treatment devices that have been done with a placebo have yielded results indicating that there is a placebo effect in these interventions studies. In these studies, there was not a significant difference in diarrheal disease in households or other comparison groups with and without the household water intervention.

However, the majority of randomized controlled trials of household drinking water treatment technologies were performed without a placebo. Therefore, lack of a placebo and masking is not only a limitation of this study but of most others on household water treatment health effects epidemiology studied as RCTs or by other prospective cohort designs.. Therefore, lack of a placebo or masking does not limit the ability to compare the results of this BSF intervention study to those of other similar POU treatment technology intervention studies.

7.2 Results and Existing Evidence about the Performance of the BSF

Comparison of Laboratory Results

Previous laboratory research on the BSF has focused primarily on removal or reductions of naturally occurring indicator bacteria. Of the five known laboratory studies existing on the BSF, none of the five attempted to standardize the concentration of bacteria dosed onto the filter (Buzunis, 1995; Donison, 2004; Lee, 2001; Palmateer et al., 1999; Sattar, 1998). All five measured only the naturally occurring bacteria that were found in the surface waters and/or mixtures of surface water and in sewage that was used as the source of microbes for feed water dosed in the studies. This laboratory study is unique among this body of existing evidence because the concentration of *E. coli* was carefully controlled for all dosing of feed water during each of the longitudinal dosing experiments. Laboratory results varied significantly for the bacteria reductions found in the five laboratory studies but they ranged from a modest initial removal of 60% to reductions of 90-99% in three of the studies. The laboratory results of the present study found similar bacteria reductions averaging 90-99% overall, and a similar pattern of improvement in bacteria reductions over time (initially, as low as 65% and improving to >99% reduction).

There is only one other study of virus removal by the BSF. The researchers examined reduction of hepatitis A virus in filters that had been in use for a period of weeks. In their study, hepatitis A virus removal was 66% (an average of 3 sampling points) but there are limited details on how the study was performed (Sattar, 1998). Our laboratory study is the first study to examine reduction of viruses in a longitudinal dosing experiment where the reduction is examined over a sustained period of weeks of constant daily dosing of virus-seeded water. Average reductions of the bacteriophage MS-2 and PRD-1 were about 60% and improved over time to reach maximum values of 78 and 87% respectively. These results are similar to those of Sattar, but they also suggest that virus reductions can improve over time and deserve further determination of performance over sustained periods of filter use, including periods before and after filter sand cleaning. This initial study of virus and bacteria reductions from water in controlled dosing experiments of the BSF shows similar results as to those of limited previous studies on these microorganisms. However the current experiments provide more performance information than previous studies because they systematically investigated two key parameters: effect of ripening and feed water dosing volume.

The initial experiments reported here suggested that dosing volume is an important factor influencing microbial reductions. The data from these initial experiments suggests that water retained in the filter bed over night demonstrated greater reductions of both *E. coli* and the two bacteriophages as compared to water that passed through the filter on the same day of dosing. While the initial two experiments described here only provided limited evidence of the magnitude of this phenomenon, this evidence was the basis for three subsequent full-scale laboratory studies that further examined this effect. The subsequent work of Mark Elliott and others has further elucidated the effect of water volume applied to the BSF as an important operational or use variable influencing microbial reduction efficiency (Elliott et al., 2006).

Comparison of Field Performance Results

The largest body of previous research on the BSF has focused on the performance in the field. This research consists of more than 15 studies that have documented reductions of bacteria in BSF-treated waters in many countries around the world. The results from these studies suggest that average reduction of bacteria in the field is 90-99%; however the results also show significant variation in bacteria reductions. Microbial reduction results in the field study of this research project also document considerable variation in filter performance. However, average bacteria reduction found in this study was typically lower than the average reductions documented in previous field studies (80% compared to 90-99%). This difference is likely due to the fact that the current study included measurements of microbial reductions from water starting immediately after filter installation in the field, followed by a longitudinal observation period of six months.

Only two of the previous field studies documented the performance of the BSF longitudinally. One study examined reductions of bacteria during eight weeks of initial operation of one BSF (Snider, 1998). This study found that the initial reductions by the BSF were low for both coliforms and *E. coli* (less than 89%) but they improved over the 8 weeks to 98%. The only other study that attempted to document *E. coli* reduction from initial installation and over time was performed in Haiti (Baker, 2006). In this study researchers found that average reduction of *E. coli* was initially low (76%) but improved over three months of the study. However, the filters in the longitudinal study did not achieve *E. coli* reductions demonstrated by other filters in the same locations that had been installed for >12 months.

Our study of ~ 75 BSFs in Bonao adds new information about BSF performance compared to other field research. We were able to document a ripening effect in the field, as demonstrated by improved *E. coli* reductions and decreased filter flow rates over the six month study period. No previous study attempted to characterize the ripening process in the field. We also attempted to identify parameters that were likely related to the observed magnitude of microbial reductions in the field. In particular, the role of initial concentrations of *E. coli* in dosed feed water was identified as a significant predictor of the microbial reduction efficiency of the BSF. While this may not be surprising, it suggests that reporting bacterial reduction efficiency alone is not an adequate parameter to quantify field performance of the BSF. The *E. coli* concentrations of both the feed water dosed to the filter and the filtered water need to be reported. Furthermore, the *E coli* levels in the filtrate need to be considered in relation to levels of risk defined by the WHO Guidelines for Drinking Water Quality. If filtrate water is consistently <10 *E. coli* per 100 mL, the water is likely to pose a low risk of waterborne disease, based on this guidance. Further studies are needed to determine if this is the case, but the results of this study support this guidance.

Other revealing results from the field study suggest that dosing frequency is an important parameter to consider in further laboratory studies. This parameter has yet to be adequately evaluated for the extent to which it affects BSF performance in improving water quality. This is important because depending upon the relationship between dosing frequency and improved water quality, modifications in dosing frequencies and volumes can possibly be made at the user level to enhance microbial reduction efficiency. Overall, this is the first field study that has attempted to understand operational parameters in the lab and the field and relate them to performance based on microbial reductions as a measure of improved water quality. The results suggest that there are many factors influencing bacterial reductions and water quality and that these need to be further investigated in field settings.

Health Impact Study and other Evaluations of Health Impact of the BSF

Numerous implementing organizations have attempted to assess improvements in user health as a result of the introduction of the BSF. The majority of these studies were not designed to rigorously assess any specific health improvement outcome measure or specific disease burden reduction. Instead, they have focused on whether or not the user of the BSF judges their own health to be improved as a result of using the BSF. For example, in multi-country on the BSF, 98% of participants interviewed stated that the BSF has improved the health of their household (Kaiser et al., 2002). The lack of rigorous scientific evidence about the ability of the BSF to reduce diarrheal disease resulted in more focused attempts to determine the impact of the BSF on health of users. Two additional studies attempted to document improvements in health as a result of BSF presence and use. The first of these studies, by Samaritan's purse in Ethiopia, assessed the two-week point prevalence of diarrheal diseases in a cross-sectional study of villages with BSFs and control villages (Maertens & Buller, 2006). The researchers found significantly lower prevalence of diarrheal disease in BSF villages than in villages without BSFs. In the second study, researchers attempted to document the reductions in diarrheal disease as a result of installation of the biosand filter in Haiti using a prospective cohort study design (CAWST, 2006). The researchers documented reductions in diarrheal disease in the communities with the newly installed BSFs compared to pre-intervention rates of diarrheal disease. While these studies deserve consideration, neither was sufficiently rigorous. Both had the limitation of the lack of a control group as an adequate basis for comparison of health impact.

This study is the first using a prospective cohort design based on a randomized controlled trial of the biosand filter. This is the first study rigorously attempting to document a decrease in diarrheal disease as a result of the introduction of the BSF into a randomly selected portion of sample households. There was a 47% reduction in diarrheal disease attributable to the BSF in households of two communities of Bonao, DR, when followed longitudinally for six months (from February 2006 to August 2006). These results are consistent with previously reported evidence that the BSF can result in improved health of the user by reducing diarrheal disease. While the results of this study should not be generalized to all other communities, this study is the first step in building an evidence base consisting of rigorous evidence for health impacts on users of the BSF. As a result of this research, a new initiative has begun to increase production and implementation of a plastic version of the biosand filter.

7.3 Discussion in Context of Other Technologies

Laboratory and Field performance of the BSF compared to Other Technologies

Laboratory evidence suggests that while the BSF can provide reductions of bacteria, it is less effective in reducing bacteria than some other available household water treatment technologies. Currently available household water treatment technologies such as solar disinfection, chlorine disinfection and ceramic microfiltration have been documented to reduce bacteria by >99% (M.D. Sobsey, 2002). Therefore, in comparison to these technologies, the BSF achieves only moderate reductions of bacteria in typical filed use. However, with modifications in use practices, based on water doing frequencies and volumes, the BSF may have to potential to reach bacteria reduction efficiencies of the other technologies, as was documented by the increased removals of bacteria the BSF from dosed water that remained in the filter bed overnight before being collected as filtrate.

Viral reductions by the BSF are only moderate (about 90%) compared to the much greater reductions achieved by solar disinfection and chlorine disinfection. These treatment technologies have been demonstrated to reduce viruses in the laboratory by >99% while average virus reductions by the BSF in our studies are only 80-90%. However, the BSF achieves virus reductions similar to those of ceramic microfilters, typically no more than ~90%.

Field studies of the various household water treatment technologies have primarily focused on the reduction of bacteria. Based on results from our field study, the BSF provides moderate (<90%) reductions of *E. coli* upon initial installation and the performance improves somewhat over time (to 90-99%). Few technologies document improved and sustained performance over time. For example, chlorine disinfection use compliance was found to decrease over time (Arnold & Colford, 2007). In a study on the sustainability of ceramic microfilters in Cambodia, researchers found continued effective performance of the filters in bacteria reductions over time in use, but due to breakage and lack of replacement parts, household use of filters decreased significantly over time (Brown, 2006). The BSF is thought to improve over time and with few or no replacement parts necessary, the BSF may be a technology that is less prone to technical failure and therefore easier to sustain regular and continued use.

In a limited cost-benefit analysis of the three household treatment technologies, the BSF was found to be relatively cost-effective when compared to ceramic filters and a chemical coagulant/disinfectant (Casanova et al., 2005). One of the reasons for the increased benefits derived from the BSF despite only moderate reductions of microorganisms was the potential for the BSF to have sustained use for a period of at least 5-10 years without failure or replacement. While the research of this current study is limited to only six months of longitudinal observation, other evidence from the field suggests that the BSFs can be used for many years without operational problems or growing disuse.

Health Impact Study Results for the BSF and Other Technologies

There is a growing body of epidemiological evidence documenting that interventions to improve drinking water quality at the household level are effective at reducing diarrheal disease burdens. Currently there are three review papers that document both improved drinking water quality and reduced diarrheal disease by the use of chlorine disinfection, combined chemical-coagulation and chlorine disinfection, solar disinfection and ceramic filtration. The average reduction in diarrheal disease measured from these studies ranges from 30-40% (Arnold & Colford, 2007; T. Clasen, Roberts et al., 2006; L. Fewtrell & Colford, 2004). The results from this randomized controlled trial of the BSF suggest a 47% reduction in diarrheal disease rates attributable to the BSF intervention. These results are consistent with those from the existing literature on the health impacts of household water treatment technology interventions and suggest the BSF is a promising candidate household water treatment technology in the effort to reduce the burden of diarrheal diseases globally.

7.4 Further Research on BSF

As many research projects do, the results from this research have led to the development of more research questions that can and should be addressed to increase the knowledge base on the BSF.

1) Laboratory research should continue to investigate the effects on performance of use conditions that are like those in the field for frequency of use, water volume dosed, and variations or changes in water quality. This will provide better information on how such variable conditions influence filter performance in reducing microbes in water.

2) Field evidence is needed to determine which potential factors of filter use practices can contribute to improved filter performance to reduce microbes in water.

3) The sustainability of BSF filter use in the field needs to be better assessed. A sustainability assessment of BSF is underway in Cambodia and another one will also be performed on the implemented filters of this study in the Dominican Republic. These studies are important next steps in trying to determine whether or not the BSF has a continued positive impact on the health of users and the quality of their water.

4) Additional analysis on the field data from this study can and should be performed to more clearly identify and better understand the relationship between water quality and diarrheal disease. This should involve re-classifying water quality exposure levels by more specifically assigning water quality to each participant and not generating average monthly values for their observations. Furthermore, an attempt should also be made to conduct further health impact analysis of the data in the form of case-control analyses.

5) Additional research on health impact of the BSF should be performed in different locations to determine and quantify the generalizability of the impact on BSF use on household diarrheal disease. Such health impact projects are planned by for a plastic version of the BSF in three countries, each on a different continent. They should provide a better evidence base to determine the effectiveness and robustness of the BSF in improving water quality and reducing diarrheal disease as a positive health impact.

Appendix 1: Study Area and Cross-sectional Survey of Study Communities

Study Area and Communities

The study area selected for the field studies in the Dominican Republic is the capital of the province of Monseñor Nouel, Bonao. Recent population estimates for Bonao suggest 73,000 people. The Demographic and Health Survey of the Dominican Republic cites four provinces in the country with diarrheal disease prevalence greater than 20% of which Monsenor Nouel is one (*Encuesta Demografica y de Salud: Republica Dominicana*, 2003). Within Bonao, the community of Jayaco, eight miles north of the city, was selected for an initial cross-sectional survey.

Our research has focused on the Jayaco community near Bonao since June 2003. The Centre for Affordable Water and Sanitation Technology (CAWST), an NGO based in Calgary, Canada, organized a meeting at that time to coordinate efforts to study the biosand filter in the Dominican Republic. The University of North Carolina participated in that meeting at CAWST's request. During the meeting a potential study community, Jayaco, was identified and visited, and contact was made with the local health clinic and a recent medical graduate serving as the clinic physician, Dr. Gloria Ortiz. In a subsequent visit in November 2003, a map of the community was copied and records for the more than 700 families were collected. Analysis of the data suggested that approximately half of the households did not have a reliable source of piped water with less then 1/5th reporting piped water in the home. Another important factor in choosing this as the study community was the relatively limited distribution of the biosand filter there. At the time of this initial visit, no BSFs had been installed in any household in Jayaco. However, in 2005 when the cross-sectional study was initiated, biosand filters had been installed in two sections of the community approximately six to nine months earlier.

Jayaco is a community of approximately 700-800 homes surrounded by agricultural rice fields with a rural health clinic run by the provincial government. Because it encompasses a large geographic area, households have access to many different sources of drinking water, depending on location. These sources vary from piped water to wells to unprotected springs and river water to collected rainwater. For example, households located in the region of the community closest to the entrance from the highway (Jayaco Arriba), have access to piped water supplies near or on their property. However, households located in the community most distant from this entrance typically rely on rainwater collection, surface water or wells. Piped water, unless otherwise noted, is supplied via a system of aqueducts under the direction of the National Institute for Potable Water (Instituto Nacional de Agua Potable y Aqueductos – INAPA).

In addition to Jayaco, a community inside the municipality of Bonao and on the edge of the Yuna River, Brisas del Yuna, was also selected for study. Brisas del Yuna represents an urban, underserved and rather transient community. It is comprised of 100-200 households, and has access to health services through a private clinic located in the center of the community. The clinic is run by a Spanish priest and is not part of the provincial health system. Households in Brisas del Yuna rely on piped water, well water and unprotected spring or river water for drinking water. As in Jayaco, drinking water source also typically depends on the geographic location of the household within the community. Both communities represent a diverse group of relatively poor, under served households with a wide range of access to services, levels of education, and wealth distribution.

Survey Methods

Surveys were developed based on prior surveys provided by Dr. Rob Quick of the US Centers for Disease Control and Prevention (CDC) and Prof. Dale Whittington, Department of Environmental Sciences and Engineering, UNC. The structure and content of the surveys was similar to that developed and used by USAID; yet the survey in our study was condensed and focused only on the core household questions used in the surveys from Project Measure

Demographic and Health Surveys (USAID survey website).

(http://www.measuredhs.com/aboutsurveys/dhs/questionnaire_archive.cfm)

The survey for the cross-sectional portion of the study is located at the end of this appendix. It was developed to collect information on household characteristics regarding levels of education, household assets, typical drinking water sources, access to levels of sanitation, as well as knowledge about diarrheal disease and hygiene behavior. In addition, households were asked to report whether or not any member of the household had experienced or were experiencing a case of diarrhea. The classification for diarrhea used was based on the World Health Organization's definition of diarrhea of three or more loose or watery stools, or any stool with blood in it, in a 24 hour period. In addition to interviews, households were asked to provide a sample of drinking water currently being used in the home. If households had more than one type of drinking water in the home, they were asked to provide all types of drinking water being used.

These surveys were developed in English, translated into Spanish and tested in a section of the community not selected to participate in the cross-sectional study because of prior presence of biosand filters. After initial testing, the surveys were modified to address questions that were difficult to interpret. Seven women from the community of Jayaco were selected to serve as interviewers, they were trained in administering the questionnaire and they were observed and given feedback while administering the questionnaire in order to enhance their proficiency. After approximately two months of questionnaire development and training, the cross-sectional study was started.

Household interviews during the cross-sectional period of the study began on June 14th, 2005 and were concluded on August 30th, 2005. The surveys were administered to every household that had at least one child reported to be less than five years of age in five selected areas of Jayaco and in Brisas del Yuna.

Due to the presence of the biosand filters, the participation of two areas of Jayaco was excluded: Jayaco Central and San Isidro. In addition, El Llano and Peñalo sections of Jayaco were also excluded because they have access to a privately supplied piped drinking water. At the time of initial household visit and interview, the purpose of the interview was explained to the primary caretaker of the household and its children and she or he was asked to participate in the interview. If she or he agreed to participate, the interviewers administered the cross-sectional interview and collected a sample of stored household drinking water if available.

Household interviews were conducted in the mornings and lasted 20-30 minutes. The results from the cross-sectional study are listed both for the individual participants and the households that enrolled in the longitudinal prospective cohort study that were present until filter distribution and therefore reached randomization (September 2005 - February 2006).

The results were analyzed and compared for households and individuals that were randomized into the control group of households and the BSF group of households. The results for age, gender and community location are presented at the individual level. The rest of the data were analyzed and compared for differences between BSF and control groups at the household level. One of the main purposes of the cross-sectional survey was to determine whether or not the randomization of households for filter selection worked by resulting in a similar and not statistically significant distribution of potential risk factors for diarrheal disease and other study variables between what would eventually become intervention (biosand filter) and control (no biosand filter) household groups for the intervention study (randomized controlled trial). If the results of the cross-sectional study showed unequal and statistically significant differences in the distributions of variables between the two household groups, these variables would then be controlled for, if necessary, in the data analysis. All survey data were compared among the BSF and control groups using a t-test for continuous variables and a chi-squared test for binary variables. Survey results are presented in the Results section, with results for variables that were found to be statistically significantly different highlighted and discussed.

After the cross-sectional interviews were completed, they were entered into a database in Microsoft Access. The data tables from Access were then analyzed in Stata. Univariate distributions of variables for all of the households (or participants) and for the control and BSF group were examined and compared. The variables ranged from characteristics at the participant level such as age, gender as well as variables collected at the household level including levels of primary education for main survey respondent, assets, household construction materials etc.

Water Quality Analysis

All household drinking water samples were collected in the field in sterile 500 mL Whirlpak bags, stored on ice, transported to the Dr. Mirna Clinical Laboratory in Bonao, and processed within 8 hours of collection. The samples were tested for total coliforms and *E. coli* via the IDEXX Colilert Quantitray system (IDEXX, Laboratories, Westbrook, ME). The samples were also tested for turbidity using a portable turbidimeter (2100p), pH using a portable pH meter (Sension1).and free and total chlorine using a colorimetric test system (pocket colorimeter II). All laboratory supplies were generously donated by IDEXX for bacteriological analysis and by Hach (Loveland, Colorado) for analysis of physical and chemical parameters.

For bacteriological analysis of total coliforms and *E. coli*, a 100-mL water sample was combined with one packet of Colilert test reagent media in a 120mL reagent bottled that contained sodium thiosulfate. Samples were mixed, poured into IDEXX Quantitrays, sealed and incubated 20-24 hours at 35˚C (± 1). Wells which turned yellow were counted positive for total coliforms and wells that fluoresced blue under a long wavelength UV light were scored positive for *E. coli*. The numbers of positive wells of each size from the Quantitrays were used to look up MPN values from an MPN table provided by IDEXX. Data from water quality analysis of bacteria were $\log_{10}$ transformed and analyzed as both continuous and categorical values.

Survey and Water Quality Results

Data Collected for Individuals

The data collected at the level of the individual are summarized in table A1.1 for the following variables: community location, age, and gender. At time of filter distribution, which was the beginning of the BSF intervention period for the randomized controlled trial, 907 people were participating in the study. Of those, 447 were randomized (at the household level) to the filter intervention group and 460 were randomized to the control group. There were nearly equal numbers of participants in each community location, with one exception. There were more participants in the control group from the community of Jayaco Arriba than there were in the BSF group and this was found to be significantly different ($p < 0.05$). This difference in participant location in intervention and control groups is important because community location can serve as a proxy for both environmental as well as socio-economic conditions. Other variables measured on the individual level suggest a nearly equal and not statistically significant distribution of participants into both intervention groups with one other exception: gender in the participants under five years old. Control households have about 8% higher proportion of males under five than females as compared to the BSF households although this was not found to be a statistically significantly different ($p = 0.05$) However, for the rest of the variables, there were very few differences in their representation in the two groups. Households had on average five people in the home. Average age of children under five was approximately two years old and average age of participants over five years old was 24 years old. Also, gender was about equally distributed in both children under five years old and participants over five years old (with close to the ideal of 50% female/50% male).

Housing Structure Characteristics

The majority of the information from the cross-sectional study was collected at the household level, including: characteristics of the housing structure, education level, household assets, household water sources, management and practices in the home, as well as the outcome variable of 1-week recall of diarrheal disease prevalence. Household construction materials can be an important variable because they can limit or otherwise modify the effect of exposure to environmental risk factors for infectious disease. They can also serve as a proxy for or an indicator of economic status. Data on household structures and construction materials as well as access to sanitation facilities are presented in table A1.2. The households in the study were typically constructed with wood, and/or concrete and blocks. For both household groups, household construction materials were similar. However, the control group had a statistically significantly higher proportion of households that did not have concrete floors; 17% as compared to only 6% in the BSF group. Also important to note is that households reported high levels of access to improved sanitation such as latrines or even flush toilets. However, about 1/5th of the households in each group did not have the latrine/toilet directly on the property and they shared access to it.

Table A1.1 Age, gender and location for participants of the randomized controlled trial of the BSF in Bonao in 2005

variable	**Household Groups**		*** $p < 0.05$ by t-test or chi-squared test**
	Control (total n=460) n (%)	**BSF (total n=447) n (%)**	**Total (total n=907) n, (%)**
Location			
Jayaco Arriba*	99 (21)	62 (14)	161 (18)
Majaguay	44 (10)	53 (12)	97 (11)
KM 100	49 (11)	47 (10)	96 (10)
KM 101	59 (13)	65 (15)	124 (14)
KM 103	84 (18)	84 (19)	168 (18)
Brisas del Yuna	125(27)	136 (30)	261(29)
Age			
Participants ≥ 5 years old	332 (72)	332 (75)	664 (73)
Participants < 5	128 (28)	115 (25)	243 (27)
Mean Age (std. dev)			
Participants (≥ 5)	24 (15)	24 (16)	24 (15)
Participants < 5	2.0(1.3)	2.2 (1.3)	2.1 (1.3)
Household Size			
Range (participants)	2 – 12 per house	3 – 15 per house	2 - 15
Average	5.3 per house	5.5 per house	5.5
Gender			
Male (<5)	69 (54)	52 (45)	122 (50)
Female (<5)	59 (46)	63 (55)	121 (50)
Male (≥ 5)	155 (47)	160 (48)	315 (47)
Female (≥ 5)	177 (53)	172 (52)	349 (53)

* - deemed significantly different by t-test or chi-squared test ($p < 0.05$)

Table A1.2 Characteristics of housing structures in the Jayaco and Brisas del Yuna communities of Bonao

variable	**Groups**		
	Control (total n=86) n (%)	**BSF (total n=81) n (%)**	**Total (total n=167) n, (%)**
Rooms in house			
1	21 (24)	15 (19)	36 (22)
2	42 (49)	35 (43)	77 (46)
3	20 (23)	18 (22)	38 (23)
4	0 (0)	10 (12)	10 (6)
5	1 (1)	1 (1)	2 (1)
missing	2 (3.)	2 (3)	4 (2)
Household walls wood			
Yes	52 (60)	44 (54)	96 (57)
No	33 (39)	35 (43)	68 (41)
Miss	1 (1)	2 (3)	3 (2)
Household walls concrete/block			
Yes	45 (52)	40 (49)	85 (51)
No	40 (47)	39 (48)	79 (47)
Miss	1 (1)	2 (3)	3 (2)
Household floor concrete*			
Yes	70 (81)	74 (91)	144 (86)
No	15 (18)	5 (6)	20 (12)
Miss	1 (1)	2 (3)	3 (2)
Access to latrine/toilet at house			
Yes	70 (81)	60 (74)	130 (78)
No	15 (18)	19 (24)	34 (20)
Miss	1 (1)	2 (3)	3 (2)
Sanitation Facilities			
Shared latrine	17 (20)	21 (26)	38 (23)
Private latrine	64 (74)	52 (64)	116 (70)
Shared/private toilet	4 (5)	6 (7)	10 (6)
missing	1 (1)	2 (3)	3 (2)

* - deemed significantly different by t-test or chi-squared test ($p < 0.05$)

Education Levels of Primary Respondent and Spouse

In an attempt to determine levels of education for certain members of the household, the primary respondent was asked to provide information on his or her own level of education as well as the education levels of his or her spouse. These data are summarized in table A1.3. There were very few differences among the two household groups when comparing them on levels of education. Approximately 11% of households in both groups had primary respondents reporting no formal education and approximately 40% had completed primary education (eight years). The data on levels of secondary education are also very similar between the groups with only 13% completing secondary education in both groups. For the spouse of the primary respondent, control and BSF households also have very similar distributions of education levels; however, these reported spousal educations levels are much lower than the primary respondent's education level. Approximately, 20% of spouses were reported to have completed primary education and 30-36% of the spouses had received no formal education at all. These levels indicate that percent of spouses of primary respondents without any formal education is almost three times as high as the primary respondent.

Household Assets

Information was collected on 35 household assets ranging from electric generators to motorcycles to animals. A subset of the data is presented in Table A1.4. Differences between households from control and BSF groups were found by comparing these results. A larger proportion of BSF households with had refrigerators, televisions, fans, and washing machines than did households without filters (p values < 0.05 by chi-square test). A higher proportion of BSF households also had motorcycles, gas stoves, radios and cell phones; although these differences were not found to be statistically significant.

Six assets were selected to develop an arbitrary asset index for the numbers of assets each household possessed. The assets chosen were: motorcycle, washing machine, fan, television, cell phone and refrigerator. These were selected based on information from the most recent Demographic and Health Survey and the univariate distribution of the asset as well as its purpose. The most recent DHS 2002 suggested that the largest increase in asset ownership in between the DHS surveys was found for washing machines (25 to 61%). Other assets were selected as proxies for information about household wealth, food management practice and access to information. For example, televisions represent modes of receiving information; motorcycles are the most popular form of transportation especially in smaller cities and communities; refrigerators and stoves are an indicator of the ability to conserve and prepare food. In addition, the univariate distributions of the assets were considered prior to inclusion in the simple asset index. Televisions and gas stoves represented assets that are found in almost 80% of all households whereas fans and washing machines are only found in about 50% of all households. The six assets were then used to develop an ordinal variable for numbers of assets. The households were classified into seven categories depending on the number of assets owned with zero being none of the six assets and six being all six of the assets. Any number in between 1 and 6 was a combination of the six assets listed. Control households had three assets on average as compared to BSF households that had four assets on average. In addition, seven (almost 10%) of the control households reported having none of the six assets while none of the BSF households were classified into that asset category. Hence, there appeared to be a difference between the number of household assets in BSF and control groups, with the control group having fewer assets than the BSF group. This suggests that for these variables, randomization was not as effective. This difference was considered during subsequent analysis of the health impact data.

Table A1.3 Levels of education for households in Bonao

variable	**Household Groups**		
	Control (total n=86) n (%)	**Intervention (total n=81) n (%)**	**Total (total n=167) n, (%)**
Primary Education Level (of primary respondent)±			
0 years	10 (12)	10 (12)	20 (12)
1	3 (3)	1 (1)	4 (2)
2	7 (8)	3 (4)	10 (6)
3	4 (5)	7 (9)	11 (7)
4	9 (10)	7 (9)	16 (10)
5	5 (6)	6 (7)	11 (7)
6	6 (7)	9 (11)	15 (9)
7	6 (7)	4 (5)	10 (6)
8	34 (40)	32 (39)	66 (39)
missing	2 (2)	2 (3)	4 (2)
Secondary Education Level±			
0	59 (69)	53 (66)	112 (68)
1	8 (9)	4 (5)	12 (7)
2	3 (3)	6 (7)	9 (5)
3	1 (2)	6 (7)	7 (4)
4	13 (15)	10 (12)	21 (14)
missing	2 (2)	2 (3)	4 (2)
Primary Education Level (of spouse of primary respondent)§			
0 years	31 (36)	25 (31)	56 (33)
1	2 (2)	0 (0)	2 (1)
2	1 (1)	0 (0)	1 (1)
3	4 (5)	7 (8)	11 (6)
4	8 (10)	8 (10)	16 (10)
5	6 (7)	10 (12)	16 (10)
6	6 (7)	5 (6)	12 (7)
7	4 (5)	8 (10)	12 (7)
8	22 (26)	16 (20)	38 (23)
missing	1 (1)	2 (3)	4 (2)
Secondary Education Level§			
0	70 (81)	69 (85)	139 (83)
1	5 (6)	1 (1)	6 (4)
2	2 (2)	2 (2)	4 (2)
3	3 (3)	3 (4)	6 (4)
4	5 (6)	4 (5)	9 (5)
missing	1 (1)	2 (3)	4 (2)

± - Refers to primary respondent’s education level

§ - Refers to spouse of primary respondent’s education level

Table A1.4 List of specific assets for households

	Groups		
	Control (total n=86) n (%)	Intervention (total n=81) n (%)	Total (total n=167) n, (%)
Household Possessions			
Motorcycle (moped)			
0	42 (49)	31 (38)	73 (44)
≥ 1	43 (50)	48 (59)	91 (55)
missing	1 (1)	2 (3)	3 (2)
Refrigerator*			
0	47 (55)	28 (34)	75 (45)
≥ 1	38 (44)	51 (63)	89 (53)
missing	1(1)	2 (3)	3 (2)
Television*			
0	18 (21)	7 (8)	25 (15)
≥ 1	67 (78)	72 (89)	139 (83)
missing	1 (1)	2 (3)	3 (2)
Beds			
1	9 (11)	8 (10)	17 (10)
2	35 (41)	34 (42)	69 (41)
3	27 (31)	21 (26)	48 (29)
4	10 (12)	12 (15)	22 (13)
5	2 (2)	3 (4)	5 (3)
6	2 (2)	1 (1)	3 (2)
missing	1 (1)	2 (3)	3 (2)
Fan *			
0	40 (47)	20 (24)	60 (36)
≥ 1	45 (52)	59 (73)	104 (63)
missing	1 (1)	2 (3)	3 (2)
Washer*			
0	43 (50.00)	22 (27)	65 (39)
≥ 1	42 (49)	57 (70)	99 (59)
missing	1 (1)	2 (3)	3 (2)
Radio			
0	34 (40)	32 (39)	66 (39)
≥ 1	51 (59)	47 (58)	98 (59)
missing	1 (1)	2 (3	3 (2)
Electric Stove			
0	78 (91)	70 (86)	148 (88)
≥ 1	7 (8)	9 (11)	16 (10)
missing	1 (1)	2 (3)	3 (2)

Gas Stove			
0	11 (13)	4 (5)	15 (9)
≥ 1	74 (86)	75 (93)	149 (89)
missing	1 (1)	2 (3)	3 (2)
Cellular			
0	44 (51)	34 (42)	78 (47)
≥ 1	41 (48)	45 (55)	86 (51)
missing	1 (1)	2 (3)	3 (2)
Asset index distribution *			
0	7 (8)	0 (0)	7 (4)
1	7 (8)	3 (4)	10 (6)
2	15 (17)	7 (9)	22 (13)
3	18 (21)	16 (20)	34 (20)
4	14 (17)	16 (20)	30 (18)
5	9 (10)	19 (22)	28 (17)
6	13 (16)	17 (21)	30 (18)
missing	3 (3)	3 (4)	6 (4)

* - The variable here in an ordinal variable that indicates the presence of one (or more if asset > 1) of the following: motorcycle, refrigerator, washer, fan, television and cell phone.

Principle Components Analysis of Household Assets to Construct a Wealth Index

In addition to the asset index, principle components analysis (PCA) was used to evaluate and generate a household score using the following assets: motorcycle, refrigerator, television, fan, washer, cellular phone, primary and secondary education, floor construction materials, access to latrine, use of gas for cooking and car (Vyas & Kumaranayake, 2006). The results from PCA were used to generate an asset index score and a wealth index. The asset score index generated from PCA was generated for each household and then the households were divided into quintiles of wealth. The results from the PCA and the classification of households into quintiles of wealth are presented in Table A.15.

Table A1.5 Results of PCA and classification into wealth quintles

	Groups		
	Control (total n=86) n (%)	**Intervention (total n=81) n (%)**	**Total (total n=167) n, (%)**
Quintiles of Wealth			
Lowest 20%	24 (29)	7 (9.0)	31 (19)
2nd lowest 20%	18 (22)	15 (19)	33 (21)
Middle 20%	13 (16)	18 (23)	31 (19)
2nd highest 20%	14 (17)	20 (26)	34 (21)
Top 20%	14 (17)	18 (23)	32 (20)
missing	3 (3)	4 (5)	7 (4)

Based on the results of PCA and the classification into wealth quintiles, there are a higher proportion of control households that were classified into the lower 40th percent of all households (50% of control households compared to 28% of filter households). The results suggest that households with filters may have improved socioeconomic conditions and this may have an effect on household rates of diarrheal disease. The results from PCA will be incorporated into the statistical analysis of diarrheal disease and filter intervention.

Household Drinking Water Management Practices

The previous sections of this chapter described data collected initially and only once during the cross-sectional phase of the study, with the exception of age. This was done because it was believed these variables were unlikely to change drastically over the relatively short duration of the study period (approximately one year). However, household drinking water management practices can change quite frequently including: frequency of collection, drinking water source, drinking water quality, and household drinking water treatment. These variables were measured during the cross-sectional survey and they were also measured at every household visit for the entire longitudinal portion of the study. Two additional variables, one for a covariate of exposure and one for outcome, were also measured in the cross-sectional survey and measured at each household visit: presence of soap and diarrheal disease.

Data collected on water management and water practices in the home are summarized in table A1.6. Approximately 50-60% of households report collecting drinking water at least once a day and about 20% reported collecting drinking water only once or twice a week. Household water treatment practice was also very similar between control and BSF households during the initial cross-sectional survey. For example, in both groups, households reported that 35-40% practiced some form of drinking water treatment. About 1/4th of the households in both BSF and control groups reported purchasing bottled water; and similarly high percentages (65-75%) had hand soap at the time of interview.

Household drinking water sources and drinking water quality are summarized in table A1.7 and in figureA1.1. During the cross-sectional interview, households were asked to list all of the drinking water sources they used. Many households reported using more than one source for drinking water. In table A1.7, the percentages reported reflect multiple sources per household and therefore do not add up to 100%. Both control and BSF households reported using a variety of drinking water sources ranging from surface water to purchasing bottled water. Approximately 50% of all households reported using some form of piped water outside of the home for drinking water.

Households had on average 10 (1.0 $\log_{10}$) *E. coli* MPN per 100mL. This value is an average of all samples; including samples that received some form of drinking water treatment in the home or drinking water that was purchased. If only untreated drinking water samples were considered, the average *E. coli* concentration was 23 (1.4 $\log_{10}$) *E. coli* MPN per 100mL. The better *E. coli* quality of drinking water samples exposed to some form of treatment suggests improved quality compared to that of untreated water samples. When the BSF and control household groups were compared, mean *E. coli* MPN concentrations per 100 mL were 7.4 (log10 0.87) in control households and 15. 5 (1.2 log10) in BSF households, which was a statistically significant difference. The distribution of drinking water quality between the control and BSF household groups is illustrated in figure 4.1 on the basis of decimal categories of *E. coli* concentrations. Percentages of water samples free of *E. coli* per 100 mL, were 35% for control households and 25% for BSF households. Percentages of water samples with >100 E. coli MPN per 100 mL were more than 25% for BSF households and slightly more than 15% for control households. While the distribution of water quality in control households and BSF households differed, the proportions of water samples at each decimal category of *E. coli* were not found to be statistically significant between BSF and control groups. In untreated water samples, control households had an average 15.5 *E. coli* per 100 mL whereas BSF households had an average 35.4 *E. coli* per 100 mL. These results suggest higher levels of *E. coli* contamination of drinking water in BSF households than in control households during the cross-sectional study.

Household Diarrheal Disease

Households were asked to report the one-week point prevalence of diarrhea for all members of the household as well as for children under the age of five years old. Approximately 20% or 1/5th of all households reported having had a case of diarrhea in the last week. One-week point prevalence of diarrhea was higher in BSF households (27%) than in control households (17%) but the difference was not statistically significant. It is important to note that a large proportion of the diarrhea was reported in children under the age of five years old. For approximately 80% of the households reporting diarrhea, that case was in a child less then five years old.

Table A1.6 Drinking water quality and management practices and diarrheal disease

variable	**Household Groups**		
	Control (total n=86) n (%)	**Intervention (total n=81) n (%)**	**Total (total n=167) n, (%)**
Frequency of obtaining drinking water			
1	8 (9)	1 (1.)	8 (5)
2	10 (12)	17 (21)	27 (16)
3	12 (14)	16 (120)	28 (17)
4	3 (3)	3 (4)	6 (4)
5	0 (0)	1 (1)	1 (1)
7 or more	52 (60)	41 (51)	93 (56)
miss	1 (1)	2 (3)	3 (2)
Report treating drinking water			
Yes	36 (42)	29 (36)	65 (39)
No	49 (57)	50 (61)	99 (59)
Missing	1 (1)	2 (3)	3 (2)
Report buying drinking water			
Yes	21 (24)	22 (27)	43 (26)
No	64 (75)	57 (70)	121 (72)
Missing	1 (1	2 (3)	3 (2)
Soap at time of interview			
Yes	64 (75)	52 (64)	116 (69)
No	21 (24)	27 (33)	48 (29)
Missing	1 (1)	2 (3)	3 (2)
Diarrhea in last 7 days			
Yes	15 (17)	22 (27)	37 (22)
No	70 (82)	57 (70)	127 (76)
Missing	1 (1)	2 (3)	3 (2)
Diarrhea in last 7 days (< 5)			
Yes	14 (16)	17 (21)	31 (19)
No	71 (82)	62 (76)	133 (80)
Missing	1 (1)	2 (3)	3 (20)

Table A1.7 Distribution of source waters for household drinking water

variable	**Groups**		
	Control (n=86) Total n (%)	**Intervention (n=81) Total n (%)**	**Total (n=167) Total n, (%)**
River*			
Yes	13 (15)	9 (11)	22 (13)
No	70 (81)	69 (85)	139 (83)
Missing	3 (4)	3 (4)	6 (4)
Well			
Yes	35 (41)	31 (38)	66 (39)
No	48 (56)	47 (58)	95 (57)
missing	3 (3)	3 (4)	6 (4)
Unprotected spring			
Yes	9 (11)	5 (6)	14 (8)
No	74 (86)	73 (90)	147 (88)
Missing	3 (3)	3 (4)	6 (4)
Rainwater			
Yes	5 (6)	8 (10)	13 (8)
No	78 (91)	70 (86)	148 (88)
Missing	3 (3)	3 (4)	6 (4)
Piped water inside the home			
Yes	6 (7)	5 (6)	11 (6)
No	77 (90)	73 (90)	150 (90)
Missing	3 (3)	3 (4)	6 (4)
Piped water outside of the home			
Yes	38 (44)	41 (51)	79 (47)
No	45 (52)	37 (45)	82 (49)
Missing	3 (3)	3 (4)	6 (4)
Bottled water			
Yes	17 (20)	18 (22)	35 (21)
No	66 (77)	60 (74)	126(75)
Missing	3 (3)	3 (4)	6 (4)
Drinking water quality			
Avg. Log_{10} MPN *E. coli* /100mL§	0.9 (n = 98)	1.2 (n = 91)	1.0 (n = 189)
Avg. Log_{10} MPN coliforms /100mL	2.2 (n = 98)	2.2 (n = 91)	2.2 (n = 189)

* - Categories of drinking water sources are not mutually exclusive. Households may have reported using more than one drinking water source.

§ - indicates a statistically significant difference ($p < 0.05$)

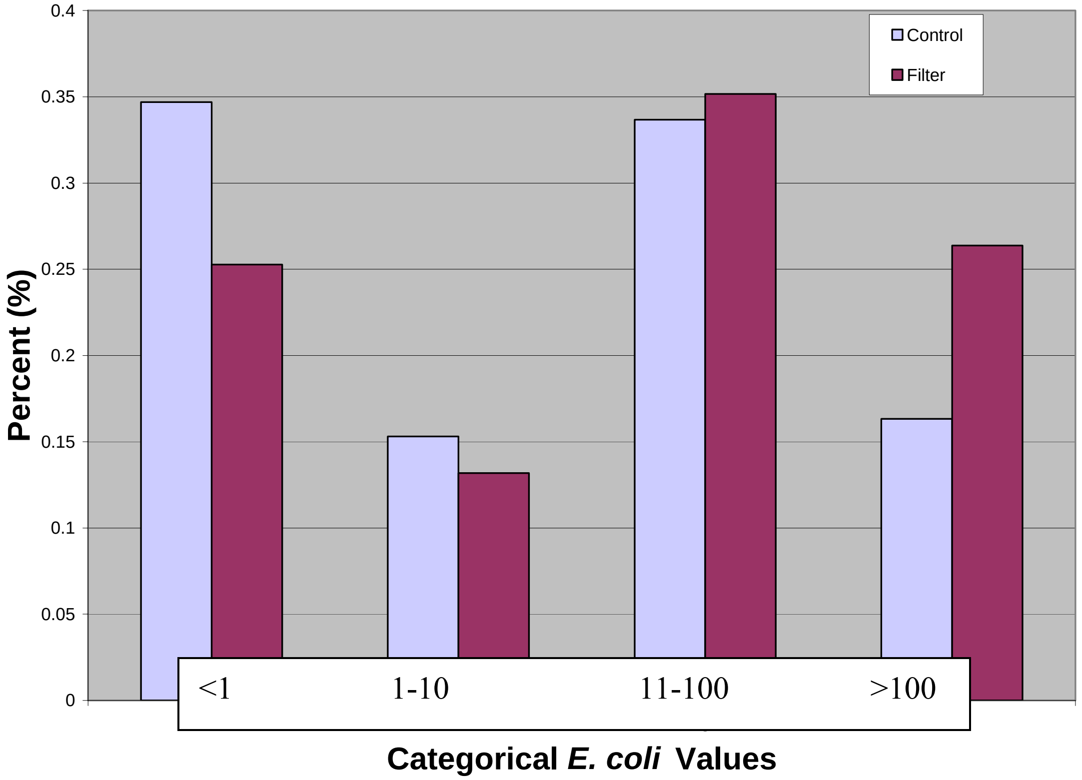


Figure A1.1 Distribution of drinking water quality during cross-sectional survey

Missing Data:

There were six households that did not provide data for the information to be collected during the cross-sectional survey. These households were not present during the initial cross-sectional survey of the study, and therefore they were not administered the complete questionnaire and represent missing data from this dataset. Overall, it is unlikely that these missing data from the initially recruited households would significantly change the univariate distributions described because they comprise less than 4% of the total data and there are approximately equal numbers of missing observations in each household group (control and BSF groups).

Summary

Data from the cross-sectional study can be summarized as follows:

- Households randomized into the control and BSF groups had about the same distribution of gender, age, household size, household construction materials, drinking water sources, drinking water management practices and access to sanitation.
- BSF households were found to have more assets than control households based on analysis on a subset of household assets. A larger percentage of control households were classified into lower 40 % wealth quintiles based on PCA.
- BSF households were found to have higher levels of drinking water contamination with *E. coli*; even when controlled for household drinking water treatment.
- BSF households reported a higher prevalence of diarrhea than control households but this difference was not found to be statistically significant ($p > 0.05$).

Appendix 2: List of Variables and Coding used in Logistic Regression

Variable	**Description**	**Coding**
Case of diarrhea	*Outcome variable.* Describes where or not participant is experiencing a case of diarrhea during each week of observation	0 = no diarrhea 1 = case diarrhea Missing – if still experiencing a continuing case of diarrhea
Intervention group	*Main Exposure variable in Chapter 5 analysis.* It is generated at the household level and describes whether or not the household was selected into the filter group or control group.	0 = control group 1 = filter group
Phase of study*-	Whether or not the measurements were taken prior to or after filter installation	0 = prior to BSF intervention 1 = after BSF intervention
Location	Describes the community location	0 = Jayaco community 1 = Brisas del Yuna community
Gender	Participant's gender	0 = female 1 = male
Latrine	Whether or not the household had access to a latrine on the property	0 = no access on property 1 = access on property
Treatment	Describes household water treatment practice for each week of observation. This is irrespective of whether or not the household had a filter. It includes reported boiling, chlorination or other treatments such as non-BSF filtration.	0 = if no treatment reported 1 = if treatment(s) reported for week of observation
Season	Describes period of time when lack of rainfall seems to have strongest effect on diarrheal disease rates and modifies the effect of the filter. Although April and May were high rainfall months, this variable identifies May and June as the months in which the effect of the rainfall is seen on filter and on diarrheal disease (considered a one month lag of the rainfall effect). This variable is only defined during the intervention period.	0 = May, June (wet season) 1 = February, March, April, July (dry season)

Group*Season	*Interaction variable.* This variable is an interaction of intervention group and the season (wet or dry). Season was deemed to be a strong effect measure modifier and therefore put into the model as an interaction variable.	0 = if control group and any season 0 = if filter group and wet season (May or June months) 1 = if filter group and dry season (February, March, April, July)
Household Assets	Summary of number of six household assets (an ordinal variable). The assets included are: motorcycle (moped), refrigerator, television, washer, fan, cell phone.	0 = if none of the six assets 1 = if any one of the six 2 = any two of the six assets 3 = any three of the six assets 4 = any four of the six assets 5 = any five of the six assets 6 = all six assets
Education	Describes whether or not the primary respondent and primary respondent's spouse	0 = if no primary education 1 = if any primary education
Drinking Water Source	Describes the source of drinking water reported during each week of observation. These were not mutually exclusive and households could have reported more than one source in each week. Indicator variables used to code for the following sources: river, well, rain, spring, tap inside home, tap outside home (on property), tap outside home and off of property, bottled water	Indicator variables for all 8 possible sources.
Age	Ordinal variable that classifies participants into one of three age groups: <2, 2-4 and 5 years of age and older. Age was age at time of observation.	0 = if < 2 years of age 1 = if 2 to 4 years of age 2 = 5 years of age and older
Drinking Water Quality	Log_{10} most probable number of *E. coli* in 100 mL as a monthly average for households. Each 1-unit change represents a 10-fold increase in E. coli concentrations.	Continuous variable (Log_{10} *E. coli* per 100 mL) Range -0.045 – 3.38, Lower detection limit -0.045 Upper detection limit 3.38
Ordinal Water Quality	*Main Exposure variable in Chapter 6 analysis.* This variable is generated at each month for each household and may be age specific if households are providing different quality water for adults and children. Monthly geometric mean.	0 = < 10 *E. coli* per 100mL 1 = 10-99 *E. coli* per 100mL 2 = >99 *E. coli* per 100mL
Class	This variable was generated by grouping households into two classes based on principle components analysis of assets.	0 = if lower 40% of SES index 1 = if upper 60% of SES index

Literature Cited

Arnold, B. F., & Colford, J. M., Jr. (2007). Treating water with chlorine at point-of-use to improve water quality and reduce child diarrhea in developing countries: a systematic review and meta-analysis. *Am J Trop Med Hyg, 76*(2), 354-364.

Baker, D. (2006). *Project BRAVO, Executive Summary*: Centre for Affordable Water and Sanitation Technology.

Brown, J. (2006). Personal communication.

Brown, J., & Sobsey, M. (2006).

Buzunis, B. J. (1995). *Intermittently Operated Slow Sand Filtration: A New Water Treatment Process.* Unpublished Master's of Engineering, University of Calgary, Calgary.

Casanova, L., Brown, J., Elliott, M., Stauber, C., & Sobsey, M. (2005). *P3 Report: Comparative Analysis of Three Sustainable Point of Use Drinking Water Treatment Technologies for Developing Nations*: Environmental Protection Agency

People Prosperity and the Planet.

CAWST.*Summary of all lab and field tests*, from http://www.cawst.org/technology/watertreatment/summaryoflabandfield.php

CAWST. (2006). *Project BRAVO: A field study in Haiti (summary of results)*: Centre for Affordable Water and Sanitation Technology.

Clasen, T., Garcia Parra, G., Boisson, S., & Collin, S. (2005). Household-based ceramic water filters for the prevention of diarrhea: a randomized, controlled trial of a pilot program in Colombia. *Am J Trop Med Hyg, 73*(4), 790-795.

Clasen, T., Nadakatti, S., & Menon, S. (2006). Microbiological performance of a water treatment unit designed for household use in developing countries. *Trop Med Int Health, 11*(9), 1399-1405.

Clasen, T., Roberts, I., Rabie, T., Schmidt, W., & Cairncross, S. (2006). Interventions to improve water quality for preventing diarrhoea. *Cochrane Database Syst Rev, 3*, CD004794.

Clasen, T., Saeed, T. F., Boisson, S., Edmondson, P., & Shipin, O. (2007). Household water treatment using sodium dichloroisocyanurate (NaDCC) tablets: a randomized, controlled trial to assess microbiological effectiveness in Bangladesh. *Am J Trop Med Hyg, 76*(1), 187-192.

Clasen, T. F., Brown, J., Collin, S., Suntura, O., & Cairncross, S. (2004). Reducing diarrhea through the use of household-based ceramic water filters: a randomized, controlled trial in rural Bolivia. *Am J Trop Med Hyg, 70*(6), 651-657.

Clasen, T. F., & Cairncross, S. (2004). Household water management: refining the dominant paradigm. *Trop Med Int Health, 9*(2), 187-191.

Craun, M. F., Craun, G. F., Calderon, R. L., & Beach, M. J. (2006). Waterborne outbreaks reported in the United States. *J Water Health, 4 Suppl 2*, 19-30.

Cutler, D., & Miller, G. (2005). The role of public health improvements in health advances: the twentieth-century United States. *Demography, 42*(1), 1-22.

Dennehy, P. H. (2005). Acute diarrheal disease in children: epidemiology, prevention, and treatment. *Infect Dis Clin North Am, 19*(3), 585-602.

Donison, K. S. (2004). *Household Scale Slow Sand Filtration in the Dominican Republic.* Unpublished Master of Engineering, Massachusetts Institute of Technology, Boston.

Duke, W. F., Nordin, R. N., Baker, D., & Mazumder, A. (2006). The use and performance of BioSand filters in the Artibonite Valley of Haiti: a field study of 107 households. *Rural Remote Health, 6*(3), 570.

Earwaker, P. (2006). *Evaluation of Household Biosand Filters in Ethiopia.* Unpublished MSc in Water Management (Community Water Supply), Cranfield University, Silsoe.

Eisenberg, J. N., Cevallos, W., Ponce, K., Levy, K., Bates, S. J., Scott, J. C., et al. (2006). Environmental change and infectious disease: how new roads affect the transmission of diarrheal pathogens in rural Ecuador. *Proc Natl Acad Sci U S A, 103*(51), 19460-19465.

Elliott, M. A., Stauber, C. E., Koksal, F., Liang, K. R., Huslage, D. K., DiGiano, F. A., et al. (2006). *The operation, flow conditions and microbial reductions of an intermittently operated, household scale slow sand filter.* Paper presented at the Recent Progress in Slow Sand and Alternative Biofiltration Processes.

Encuesta Demografica y de Salud: Republica Dominicana. (2003). United States Agency of International Development.

Esrey, S. A. (1996). Water, waste, and well-being: a multicountry study. *Am J Epidemiol, 143*(6), 608-623.

Esrey, S. A., Feachem, R. G., & Hughes, J. M. (1985). Interventions for the control of diarrhoeal diseases among young children: improving water supplies and excreta disposal facilities. *Bull World Health Organ, 63*(4), 757-772.

Esrey, S. A., Habicht, J. P., & Casella, G. (1992). The complementary effect of latrines and increased water usage on the growth of infants in rural Lesotho. *Am J Epidemiol, 135*(6), 659-666.

Esrey, S. A., Potash, J. B., Roberts, L., & Shiff, C. (1991). Effects of improved water supply and sanitation on ascariasis, diarrhoea, dracunculiasis, hookworm infection, schistosomiasis, and trachoma. *Bull World Health Organ, 69*(5), 609-621.

Fewtrell, L., & Colford, J. M., Jr. (2005). Water, sanitation and hygiene in developing countries: interventions and diarrhoea--a review. *Water Sci Technol, 52*(8), 133-142.

Fewtrell, L., & Colford, J. M. J. (2004). *Water, Sanitation and Hygiene: Interventions and Diarrhoea*. Washington, DC: The International Bank for Reconstruction and Development/The World Bank.

Fewtrell, L., Kaufmann, R. B., Kay, D., Enanoria, W., Haller, L., & Colford, J. M., Jr. (2005). Water, sanitation, and hygiene interventions to reduce diarrhoea in less developed countries: a systematic review and meta-analysis. *Lancet Infect Dis, 5*(1), 42-52.

Gundry, S., Wright, J., & Conroy, R. (2004). A systematic review of the health outcomes related to household water quality in developing countries. *J Water Health, 2*(1), 1-13.

Huo, A., Xu, B., Chowdhury, M. A., Islam, M. S., Montilla, R., & Colwell, R. R. (1996). A simple filtration method to remove plankton-associated Vibrio cholerae in raw water supplies in developing countries. *Appl Environ Microbiol, 62*(7), 2508-2512.

Jensen, P. K., Jayasinghe, G., van der Hoek, W., Cairncross, S., & Dalsgaard, A. (2004). Is there an association between bacteriological drinking water quality and childhood diarrhoea in developing countries? *Trop Med Int Health, 9*(11), 1210-1215.

Joint Monitoring Programme Coverage Estimates: Improved Drinking Water. (2006). from http://www.wssinfo.org/en/238_wat_latino.html

Kaiser, N., & Chang, D. (1998). *Report on the Lai Yen Water Filter Project and Sustainability Analysis (Vietnam)*: Samaritan's Purse.

Kaiser, N., Liang, K., Maertens, M., & Snider, R. (2002). *Biosand Household Water Filter Evaluation 2001: A Comprehensive Evaluation of the Samaritan's Purse Biosand Filter (BSF) Projects in Kenya, Mozambique, Cambodia, Vietnam, Honduras and Nicaragua*: Samaritan's Purse Canada.

Kosek, M., Bern, C., & Guerrant, R. L. (2003). The global burden of diarrhoeal disease, as estimated from studies published between 1992 and 2000. *Bull World Health Organ, 81*(3), 197-204.

Lantagne, D. (2004). Trip Report - Dominican Republic, March 2004.

LeChevallier, M. W., Welch, N. J., & Smith, D. B. (1996). Full-scale studies of factors related to coliform regrowth in drinking water. *Appl Environ Microbiol, 62*(7), 2201-2211.

Lee, T.-L. (2001). *Biosand Household Water Filter Project in Nepal.* Unpublished Master's of Engineering, Massachusetts Institute of Technology, Boston.

Liang, J. L., Dziuban, E. J., Craun, G. F., Hill, V., Moore, M. R., Gelting, R. J., et al. (2006). Surveillance for waterborne disease and outbreaks associated with drinking water and water not intended for drinking--United States, 2003-2004. *MMWR Surveill Summ, 55*(12), 31-65.

Maertens, M., & Buller, A. (2006, October 2005). *Kale Heywet Ethiopia Household Water and Sanitation Project Evaluation.* Paper presented at the University of North Carolina Safe Drinking Water, Chapel Hill, North Carolina.

Maier, R. M., Pepper, I. L., & Gerba, C. P. (2000). *Environmental Microbiology*: Academic Press.

Manz, D., Buzunis, B., & Morales, C. (1993). *Nicaragua Household Water Supply and Testing Project.*

McLennan, J. D. (2000). Prevention of diarrhoea in a poor District of Santo Domingo, Dominican Republic: practices, knowledge, and barriers. *J Health Popul Nutr, 18*(1), 15-22.

Meeting the MDG Drinking Water and Sanitation Target. (2006). from http://www.wssinfo.org/pdf/JMP_06.pdf

Moe, C. L., Sobsey, M. D., Samsa, G. P., & Mesolo, V. (1991). Bacterial indicators of risk of diarrhoeal disease from drinking-water in the Philippines. *Bull World Health Organ, 69*(3), 305-317.

Mol, A. (2001). Targeting individuals: away from the concept of community involvement

The success of household sand filtration. *Waterlines, International Journal of Appropriate Technologies for Water Supply and Sanitation, 20*(1).

Momba, M. N., & Kaleni, P. (2002). Regrowth and survival of indicator microorganisms on the surfaces of household containers used for the storage of drinking water in rural communities of South Africa. *Water Res, 36*(12), 3023-3028.

Mutanda, L. N., Kinoti, S. N., Gemert, W., & Lichenga, E. O. (1984). Age distribution and seasonal pattern of rotavirus infection in children in Kenya. *J Diarrhoeal Dis Res, 2*(3), 147-150.

Ortiz, G. M., Stauber, C., Aiken, B., & Sobsey, M. (2007).

Palmateer, G., Manz, D., & Jurkovic, A. (1999). Toxicant and parasite challenge of Manz intermittent slow sand filter. *Environmental Toxicology, 14*, 217-225.

Payment, P., Trudel, M., & Plante, R. (1985). Elimination of viruses and indicator bacteria at each step of treatment during preparation of drinking water at seven water treatment plants. *Appl Environ Microbiol, 49*(6), 1418-1428.

Pinfold, J. V., Horan, N. J., & Mara, D. D. (1991). Seasonal effects on the reported incidence of acute diarrhoeal disease in northeast Thailand. *Int J Epidemiol, 20*(3), 777-786.

Pruss, A., Kay, D., Fewtrell, L., & Bartram, J. (2002). Estimating the burden of disease from water, sanitation, and hygiene at a global level. *Environ Health Perspect, 110*(5), 537-542.

Pruss-Ustun, A., & Corvalan, C. (2006). How Much Disease Burden can be Prevented by Environmental Interventions? *Epidemiology, 18*(1), 1-12.

Rabe-Hesketh, S., & Skrondal, A. (2005). *Multi-leveling and longitudinal modeling using Stata*. College Station: Stata Pres.

Reller, M. E., Mendoza, C. E., Lopez, M. B., Alvarez, M., Hoekstra, R. M., Olson, C. A., et al. (2003). A randomized controlled trial of household-based flocculant-disinfectant drinking water treatment for diarrhea prevention in rural Guatemala. *Am J Trop Med Hyg, 69*(4), 411-419.

Rowland, M. G. (1986). The Gambia and Bangladesh: the seasons and diarrhoea. *Dialogue Diarrhoea* (26), 3.

Samaritan's Purse Canada Website. from http://www.samaritanspurse.ca/ourwork/water/feature-Kenya.aspx

Sattar, S. (1998). *Study of the Virus-Removing Activity of the Manz Intermittently Operated Slow Sand Filter for Drinking Water*. Ottawa, Ontario, Canada: The International Development Research Centre.

Slow Sand Filtration. (1991). New York: American Society of Civil Engineers.

Snider, R. (1998). *A Final Report on the Biosand Water Filter (BWF) Project in East Africa (Western Kenya)*: Samaritan's Purse.

Sobsey, M. D. (2002). Managing Water in the Home: Accelerated Health Gains from Improved Water Supply. *World Health Organization*.

Sobsey, M. D., Handzel, T., & Venczel, L. (2003). Chlorination and safe storage of household drinking water in developing countries to reduce waterborne disease. *Water Sci Technol, 47*(3), 221-228.

Stauber, C. E., Elliott, M. A., Koksal, F., Ortiz, G. M., DiGiano, F. A., & Sobsey, M. D. (2006). Characterisation of the biosand filter for E. coli reductions from household drinking water under controlled laboratory and field use conditions. *Water Sci Technol, 54*(3), 1-7.

Trevett, A. F., Carter, R., & Tyrrel, S. (2004). Water quality deterioration: a study of household drinking water quality in rural Honduras. *Int J Environ Health Res, 14*(4), 273-283.

The UN Millennium Development Goals (website). from http://www.un.org/millenniumgoals/index.html

Varghese, A. (2002). *Point-of-Use Water Treatment Systems in Rural Haiti: Human Health and Water Quality Impact Assessment.* Unpublished Master's of Engineering, Massachusetts Institute of Technology, Boston.

Vyas, S., & Kumaranayake, L. (2006). Constructing socio-economic status indices: how to use principal components analysis. *Health Policy Plan, 21*(6), 459-468.

Weber-Shirk, M. L., & Chan, K. L. (2007). The role of aluminum in slow sand filtration. *Water Res, 41*(6), 1350-1354.

White, G. F., Bradley, D. J., & White, A. U. (2002). Drawers of water: domestic water use in East Africa. 1972. *Bull World Health Organ, 80*(1), 63-73; discussion 61-62.

WHO. (2006). from http://www.who.int/household_water/en/

Wright, J., Gundry, S., & Conroy, R. (2004). Household drinking water in developing countries: a systematic review of microbiological contamination between source and point-of-use. *Trop Med Int Health, 9*(1), 106-117.

Yao, K. M., Habibian, M. T., & O'melia, C. R. (1971). Water and waste water filtration: concepts and applications. *Environmental Science and Technology, 5*(11), 1105-1112.