

Report for 2005WA116B: Removal of the Human Pathogen *Giardia intestinales* from Ground Water

Publications

- Conference Proceedings:
 - Rust, Colleen, Dirk Schulze-Makuch, Robert S. Bowman, and Diane K. Meier. 2005. Field Testing of a Prototype Filter System for the Removal of the Human Pathogen *Giardia intestinales* from Ground Water "in" EOS Transactions, American Geophysical Union, 86(52), Fall 2005 Annual Meeting Supplement, Abstract H23B-1431.
 - Schulze-Makuch, Dirk, Colleen Rust, Robert S. Bowman, and Diane K. Meier. 2005, Developing a Prototype Filter System for the Removal of Human Pathogens from Drinking Water, "in" Geological Society of America 2005 Annual Meeting, Salt Lake City, Utah, October 16-19, 2005, Abstracts with Programs 37(7) p. 473.
- Other Publications:
 - Rust, Colleen, Dirk Schulze-Makuch, and Robert S. Bowman. 2005. Removal of the Human Pathogen *Giardia intestinales* from Ground Water, presentation at the 2005 National Ground Water Summit, April 17-20, 2005, San Antonio, Texas.

Report Follows

PROBLEM AND RESEARCH OBJECTIVES

Microbial contamination of groundwater is a serious concern worldwide. For many countries, groundwater provides approximately 40% of the potable water used for human consumption. Cyst-forming protozoans such as *Giardia intestinales* and *Cryptosporidium parvum*, viruses such as Hepatitis A, and even pathogenic bacteria such as certain *E. coli* strains can survive for extended periods of time in ground water systems with temperatures of less than 10°C, migrate significant distances, and are relatively resistant to standard municipal water system chlorination practices. Though dormant outside the host, as few as ten cysts can result in a human infection (Casemore et al., 1997).

Pathogenic bacteria, viruses, and protozoans tend to be negatively charged in the pH range of most ground waters. Thus, naturally occurring and modified materials such as surfactant-modified zeolites (SMZ), which have net positive surface charges and hydrophobic properties, are suitable as barriers to impede pathogen migration in aquifer systems (Fig. 1). In our experiments SMZ has been used to remove *E. coli* and the bacteriophage MS-2 from sewage water with a high success rate (*E. coli* 100%, MS-2 > 90%) (Schulze-Makuch et al., 2002; Schulze-Makuch et al., 2003). Testing was conducted both in the laboratory and the field to test the removal efficiency of SMZ for *Giardia intestinales* and its analogs.

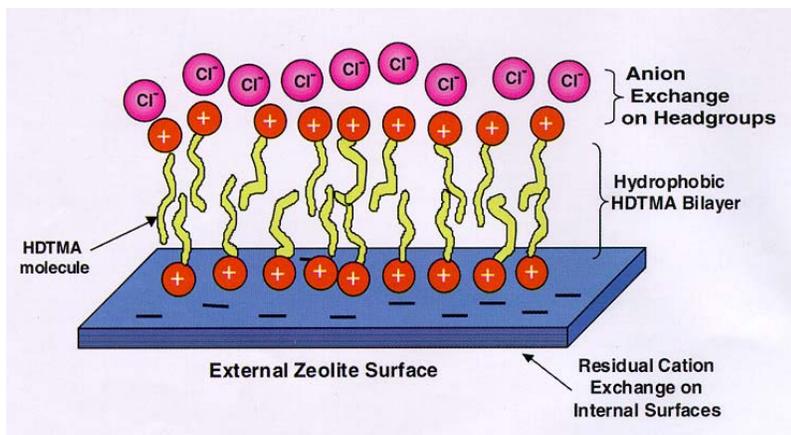


Figure 1: Schematic of the surface of the SMZ from Schulze-Makuch et al. (2002)

The overall goal of this project was to develop and test a zeolite-based filter material for removing *Giardia* and other pathogenic organisms from contaminated drinking water. To achieve this goal we focused on the following specific objectives:

Objective 1: Test three different SMZ formulations, Raw, Cationic, and Hydrophobic SMZ were used in a model aquifer to determine their abilities to remove *Giardia intestinales* and microsphere analogs from water. The outcome of this objective produced quantitative measurements of their removal efficiencies. The coarse-grained Cationic SMZ (1.4-2.4 mm) formulation was further tested at our field site using water amended with microspheres to simulated *Giardia* cyst behavior.

Objective 2: Test the coarse-grained Cationic SMZ (1.4-2.4 mm) formulation, the most promising SMZ formulation (at time of scheduled field test) in the field using water amended with microspheres that simulate *Giardia intestinales*. The outcome of this objective helped the development of a prototype filter system and validation of the laboratory results for removing *G. intestinales* from drinking water.

METHODOLOGY

Three different SMZ formulations were prepared using zeolite from the St. Cloud mine in Winston, NM (Bowman et al., 2001) (Table 1). Five different experimental runs were conducted under varied conditions with microspheres (Polystyrene, 6-7.9 μm) and viable *Giardia* cysts (7-10 μm) for fine cationic SMZ (0.4-1.4 mm, #1440), coarse cationic SMZ (1.4-2.4 mm, #814), hydrophobic SMZ, and raw zeolite, and sand only, and a 10-cm wide barrier of SMZ to test the removal efficiency of SMZ for *Giardia intestinales* using the *Giardia* cysts and microsphere analogs. The Plexiglass model aquifer (dimensions of 2.2 cm wide x 53 cm long) was filled with coarse silica sand (0.210-1.19 mm, 99.4 % silicon dioxide) to mimic realistic natural field conditions (Fig. 2). CaCl_2 water, typical of Western US water chemistries, was used flush the system and to be used as a baseline, and then bromide, microspheres (analog for *Giardia*) and *Giardia intestinales* cysts were used. In all laboratory experiments, the hydraulic gradient was set at 0.07 by controlling the hydraulic head in the inlet and outlet reservoirs. The arrival of the tracer down gradient of the SMZ barrier was compared to the arrival in the absence of the barrier to evaluate the effectiveness of each SMZ formulation.

For the fine cationic SMZ, 5 mL of a 0.1-M potassium chloride solution was injected into the model aquifer and 1-mL samples were taken periodically from Wells 1 & 2. The electrical conductivity of these samples was measured in order to set up appropriate sampling times for the subsequent runs with microspheres and *Giardia*. For all tested filter material, microspheres (1.0% w/v) and *Giardia* cysts (5×10^6 cysts) were injected during two separate runs into the model aquifer and 1-mL samples were taken from Wells 1 & 2. The samples were then analyzed using standard iodine stained cell counting methods with a bright line hemacytometer and a light microscope. Triplicate counts were done on each sample for both the microspheres and the *Giardia* cysts.

The coarse-grained Cationic SMZ (1.4-2.4 mm) formulation was further tested at our field site using water amended with microspheres to simulated *Giardia* cyst behavior. The field site is an existing multiple well site at the University of Idaho in Moscow (Fig. 3). The wells are completed in the Lolo Basalt Formation; a highly heterogeneous and anisotropic fractured basalt aquifer system typical of the subsurface of most of eastern Washington and northeastern Oregon. The injection well (T16D) and pumping / sampling well (V16D) are separated by 7.6 meters (25 feet), are connected by a known E fracture zone at 21.3 meters below ground (70 feet) (Fig. 3).

 	Hydrophobic	Fine Cationic	Coarse Cationic	Raw Zeolite	Sand Only (no barrier)	Lehner, 2004 Hydrophobic
8 μm Microspheres (Fluorescent Polystyrene)	99.8 %	98.2 %	86.9 %	96.9 %	<i>N/A</i>	5 μm Microspheres 99.2 %
8 μm <i>Giardia</i> Cysts (non-viable)	100.0 %	67.1 %	76.2 %	37.5 %	66.7 %	10 μm Microspheres 67.2 %

Table 1: Removal rates of SMZ formulations during the laboratory experiments

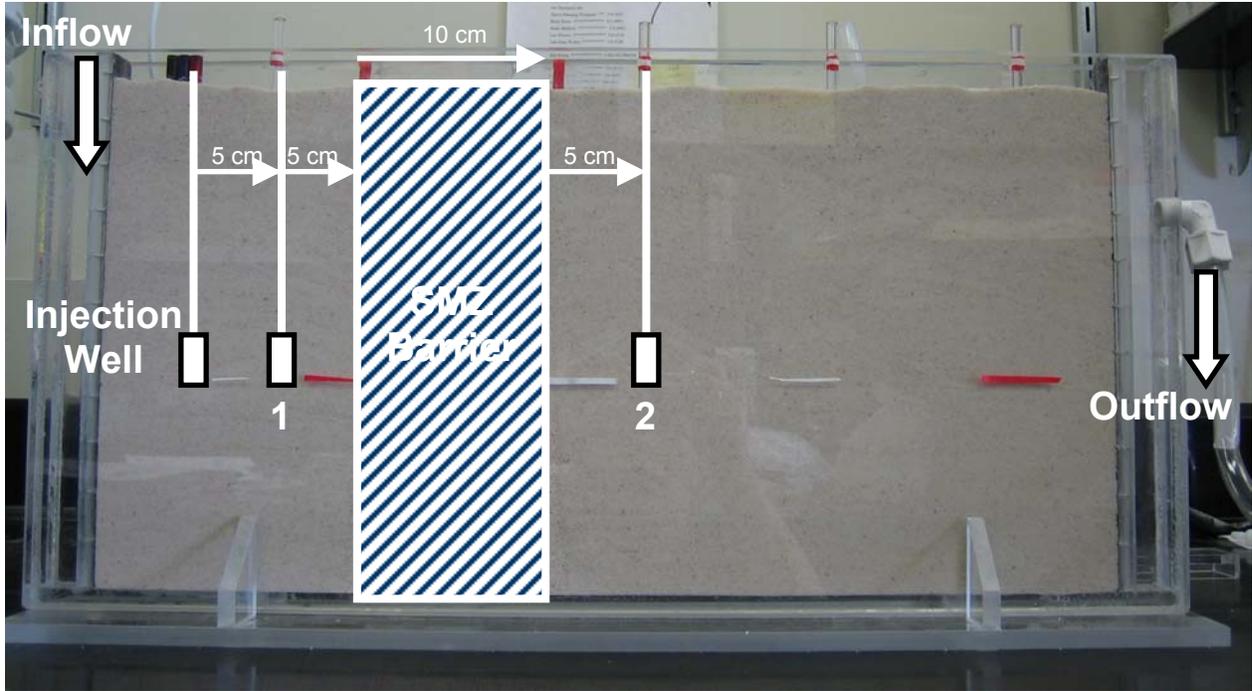


Figure 2: Model aquifer set-up with a 0.07 gradient

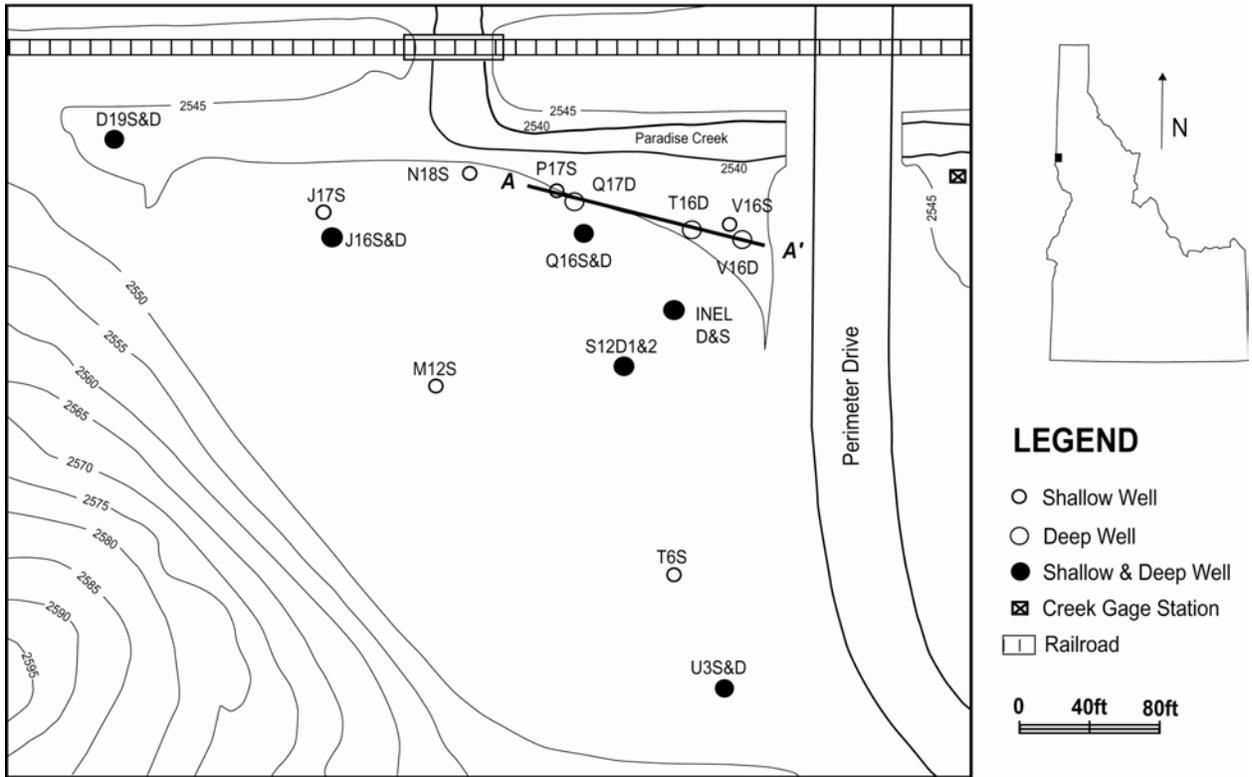


Figure 3: Well Locations at UIGRS (modified from Pardo Figure 1.1)

The SMZ pathogen field filter (Fig. 4) was installed directly in the well bore and the concentrations of microsphere-amended ground water were measured before and after filtration. The pumping well (V16D) was pumped at a constant rate of 0.92 L/sec (13 gpm) to force a gradient through the E Fracture Zone from T16D to V16D. Three slugs were injected an hour after each other at time zero (10 μ m Yellow/Green Fluorescent microspheres plus Bromide), one hour after (6-7.9 μ m Nile Red Fluorescent microspheres), and two hours after (1 μ m Red Fluorescent microspheres). Pumping over an extended period was continued for 13 hours in order to test the lifetime of our prototype filter system. Our tests and results were targeted at developing a prototype filter system for removing a multitude of human pathogens in drinking water.

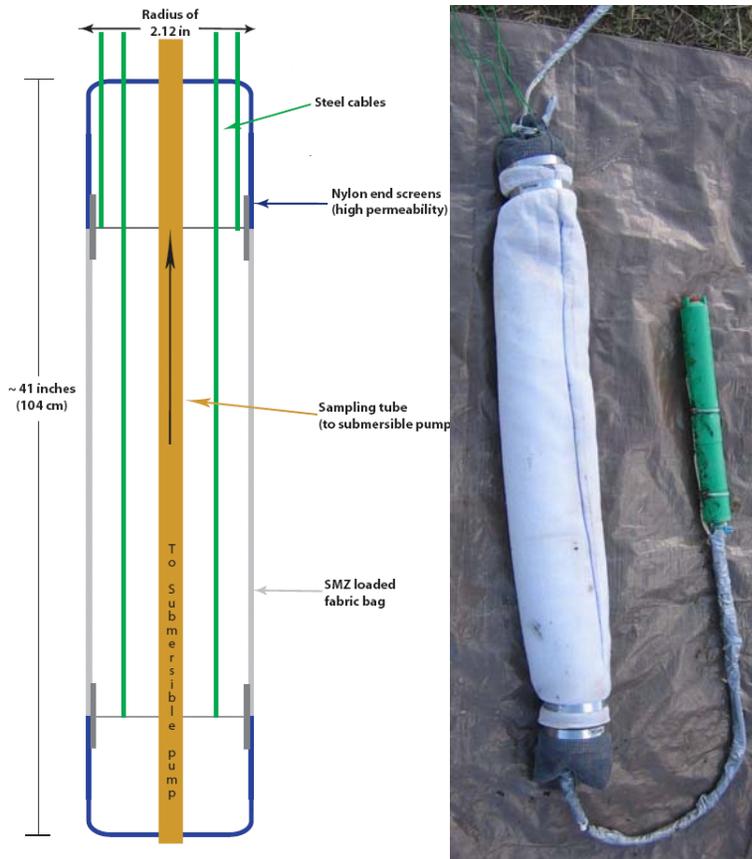


Figure 4: SMZ pathogen filter filled with Coarse Cationic SMZ, with a green electrical submersible sample pump

Figure 5: Microsphere concentrations with Coarse Cationic SMZ (1.4-2.4 mm, #814)

Counting of the field samples is not complete. Every sample taken in the field was filtered on a 0.4 μ m black filter, placed on a glass slide, and counted on a Fluorescent microscope at 100 x with oil.

In addition to microsphere slugs, we investigated the presence of in situ *E. coli* and Total Coliforms in the shallow aquifer. The well site is located in close proximity (Fig. 3) to Paradise Creek which drains the local watershed. Previous physical hydrology studies have suggested (Li, 1991) that Paradise Creek feeds all the UIGRS wells. If this proved to be true we would have a continuous source of natural organic contaminants that could be tested with our SMZ filter. Samples were taken in Paradise Creek, before and after the filter to be incubated and quantified for *E. coli* and Total Coliforms (Fig. 6). After a 24 hour incubation period, no *E. coli* was found before or after the SMZ filter within the aquifer, but it was found in the Paradise Creek samples. Up to > 2000 cells per 100 mL *E. coli* were found in the creek samples. This high concentration may be due to a

rain runoff event that occurred during the field test. Total Coliforms were found in Paradise Creek, before and after the filter. Roughly 50% removal rate was shown by the Coarse Cationic SMZ used in the prototype filter.

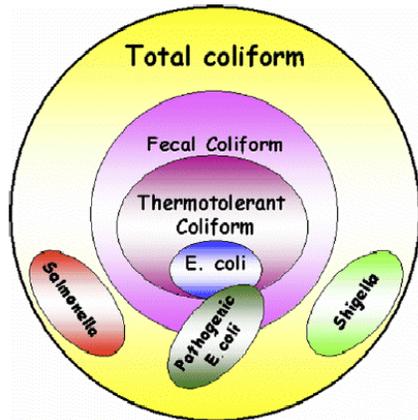


Figure 6: Diagram of the relationship of Total Coliforms and *E. coli* (fecal form).

PRINCIPAL FINDINGS AND SIGNIFICANCE

All formulations of SMZ were effective at removing *Giardia intestinales* cysts from the groundwater, but removal rates were not as high as for bacteria and viruses in the earlier experiments (Schulze-Makuch et al., 2002; Schulze-Makuch et al., 2003). The removal efficiency varied with the particular formulation of the SMZ used (Table 1). The SMZ filtration material with the highest removal rate, shown by our model aquifer runs, was the Hydrophobic SMZ. The field testing using coarse Cationic SMZ was not as effective as shown in earlier laboratory tests.

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