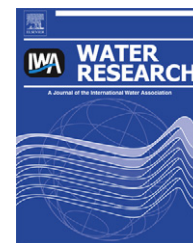


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Virus transport in a discrete fracture

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ABSTRACT

Tracer experiments were carried out in a naturally discrete-fractured chalk core with solute tracers Li^+ and Br^- , and colloidal tracers of two origins—bacteriophages (MS2, ϕX174 and T4) and fluorescent latex microspheres. The colloidal tracers were either ~ 20 nm (MS2, ϕX174 and microspheres) or ~ 200 nm (T4 and microspheres) in size. Both solute and colloidal tracers were injected at a constant flux at the fracture inlet and collected at the outlet to evaluate the form of their breakthrough curves (BTCs). The BTCs of all tracers were compared and analyzed.

The BTC analysis displayed significant differences in recovery as a function of tracer size and type. Even within the same colloid size, transport of the microspheres and bacteriophages was dissimilar, likely due to minor differences in density, surface chemistry and shape. More pronounced peaks and recoveries were observed with ~ 200 nm compared to ~ 20 nm microspheres and phages. Arrival time at the outlet was also size-dependent, with larger microspheres and phages having longer residence times than smaller ones, and solutes being 5–15 times slower than colloids of both sizes. The observed differences were explained by a combination of size and electrostatic interactions that facilitates entrance and transport within the pores in the chalk matrix.

Overall, our results clearly demonstrate that fractures are favorable carriers for viruses of different sizes with different surface properties. The viruses' properties were also shown to govern their transport through the fractures.

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1. Introduction

Groundwater contamination by emerging microbial contaminants is being intensively reported worldwide (e.g., Flynn and Sinreich, 2010; Mohanram et al., 2010). One of the major causes of waterborne diseases are enteric viruses, mainly of fecal origin, which have the capacity to survive in groundwater from days (Pourcher et al., 2007) to months (Gerba and Smith, 2005; Yates et al., 1985) in an infective form (Collins et al., 2006a; Ogorzaly et al., 2010). Bacteriophages have become popular for studies of virus transport in water (e.g., Aronino

et al., 2009; Havelaar et al., 1991; Vorkas and Lloyd, 2000) as they are similar in size, shape, and surface properties to enteric viruses with an RNA genome size similar to those of picorna-, calici- and astroviruses (Carter, 2005). Numerous studies on viral transport through granular media have been published, reporting on the influence of water saturation level, aquatic chemistry (mainly pH and ionic strength), influence of organic matter, and soil properties (e.g., Chu et al., 2003; Han et al., 2006; Jin et al., 2000; Nasser et al., 2002; Torkezaban et al., 2006). However, whereas virus transport in porous media has been intensively studied, that in fractured

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media has garnered much less attention. As fractures are known to be favorable carriers for colloids (e.g., McKay et al., 1993, 2002; Reimus et al., 2003; Weisbrod et al., 2002), the need to explore the transport of viruses in fractured systems is clear.

The major difference between fractured and porous media is the difference in transport mechanisms, due to the substantial size differences between porous and fractured voids. A large fracture aperture combined with potentially high flow velocities results in preferential pathways for colloids (Sirivithayapakorn and Keller, 2003). The role of flow rate in colloid transport through fractures has been highlighted in several studies (e.g., McKay et al., 2002; Weisbrod et al., 2002; Zvikelsky and Weisbrod, 2006). However, fractures are more complex conduits than simply two parallel smooth walls with a given aperture. Rather, the variable aperture, surface roughness, flow pathways, charge variability and dynamic changes of internal fracture properties influence transport and retention (Chrysikopoulos, 1999; Dijk and Berkowitz, 1998; Frazier et al., 2002; McKay et al., 2002; Oron and Berkowitz, 1998).

Several mechanisms might be responsible for the retention of bacteriophages in fractures, including electrostatic or hydrophobic attachment to the fracture wall, gravitational settling, and physical straining (e.g., Huber et al., 2012; Zvikelsky et al., 2008). In size smaller than 1000 nm, Brownian motion is likely to be the more important retention mechanism (Zheng et al., 2009). Shorter residence times for larger colloidal particles migrating through a discrete fracture were reported by Zvikelsky and Weisbrod (2006). Significant differences were also reported between the transport of buoyant (e.g., latex microspheres) and dense (e.g., clay) colloids in the same $\sim 1 \mu\text{m}$ size range (Zvikelsky et al., 2008). Nevertheless, to the best of our knowledge, no one has ever studied whether virus transport is similar to, and affected by the same properties as that of latex microspheres.

Transport and attachment of viruses to fractured media is complex, and tracers' ability to capture this behavior is the subject of debate in the literature. Recent studies (Asraf-Snir and Gitis, 2011), however, suggest that solute tracers cannot be used to predict the transport of colloidal particles. Those authors showed that the difference in diffusion coefficients between the angstrom-scale solute tracers (10^{-10} m) and the colloids (ranging between 10^{-8} and 10^{-5} m) influences the residence time. Specifically, the residence times of bacteria and viruses were lower than those of the solute tracers as their diffusion was small relative to the diffusion of solutes. The higher diffusion coefficient of smaller colloids (vs. larger colloids) and subsequently higher Brownian motion and interactions with fracture walls also explain the lower recovery of small ($0.02 \mu\text{m}$) vs. large (0.2 and $1.0 \mu\text{m}$) colloids in discrete fractures (Zvikelsky and Weisbrod, 2006). Considering the complexity of the pore network structure and the fact that only the small colloids can diffuse into the matrix pores, it is likely that some of the small colloids diffuse into the matrix in irreversible manner (Zvikelsky and Weisbrod, 2006).

The current study was performed to compare transport and retention of tracers in a naturally discrete fracture. Transport of three viruses, with different sizes, surface charges, and hydrophobicities was explored, together with the transport of latex microspheres and inorganic solutes (Li^+ and

Br^-). The main objective was to explore the similarities between virus and latex microsphere transport in fractures and to establish the impact of virus size and surface properties on their transport.

2. Materials and methods

2.1. Experimental setup

The study was performed in a naturally fractured chalk core drilled from Eocene chalk of the Avdat formation in the Negev region of Israel. The discrete fracture crossed the entire core length, 43 cm long and 18 cm in diameter. The core was wrapped with Teflon[®] tape before being fixed with epoxy cement (Duralite[®]) inside a PVC casing (Fig. 1). Teflon inlet and outlet chambers were attached to each side of the flow boundaries of the fracture, while the other two boundaries were sealed with epoxy cement to create no-flow boundaries. The chamber's volume was relatively large (~ 20 mL) to prevent clogging. The chalk matrix is negatively charged at the experimental conditions (ζ -potential of -20.2 at pH 7; Zvikelsky and Weisbrod, 2006). Nevertheless, as surfaces of similar fractures were observed to be highly heterogeneous and include a variety of surface minerals (Weisbrod et al., 1999, 2000), it is impossible to define the exact surface charge of the fracture throughout.

The core was saturated under vacuum (Zvikelsky and Weisbrod, 2006) using degassed artificial rainwater (ARW) and flow was maintained for several months prior to the experiments to ensure stable conditions and minimum or no air bubbles entrapment. The chemical composition of the ARW before addition of the tracers was (in mg/L): 12.2 Ca^{2+} , 12.7 Cl^- , 13.8 SO_4^{2-} , 13.1 Na^+ , 3.5 Mg^{2+} , 35 HCO_3^- , 15.3 NO_3^{2-} , pH 7.2 ± 0.1 , ionic strength 0.0023 M, similar to the average chemical composition of rainwater in the northern Negev desert. Dissolution was found to be insignificant throughout the pretreatment (continuous flow of ARW for over a year) and all experimental stages. The same ARW has been used in previous studies carried out in similar fractured cores (Tang and Weisbrod, 2009, 2010; Zvikelsky and Weisbrod, 2006; Zvikelsky et al., 2008).

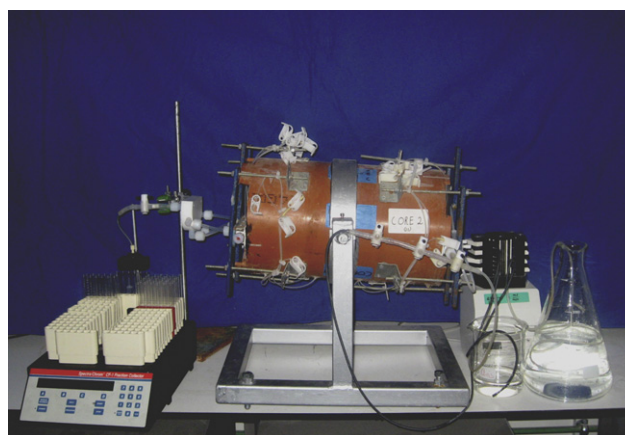


Fig. 1 – A picture of the experimental setup used. The fractured core is inside the PVC pipe.

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