Journal of Hazardous Materials 180 (2010) 640–647

Contents lists available at ScienceDirect

Journal of Hazardous Materials

journal homepage: www.elsevier.com/locate/jhazmat

Removal of bacteriophages MS2 and phiX174 from aqueous solutions using a red soil

Hui Zhang [a,b], Jiabao Zhang [a], Bingzi Zhao [a,*], Congzhi Zhang [a,b]

[a] State Experimental Station of Agro-Ecosystem in Fengqiu, State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, PR China
[b] Graduate School of the Chinese Academy of Sciences, Beijing 100049, PR China

ARTICLE INFO

Article history:
Received 18 January 2010
Received in revised form 5 April 2010
Accepted 20 April 2010
Available online 27 April 2010

Keywords:
Red soil
Virus removal
Equilibrium adsorption
Kinetic adsorption–desorption
Soil sterilization

ABSTRACT

Adsorption and desorption of viruses onto and from an adsorbent may have a dominant role in evaluating removal efficiency of a material. This study evaluated the effectiveness of a red soil from south part of China to remove two viruses, MS2 and phiX174, by adsorption from dilute aqueous solutions using a set of equilibrium and kinetic batch experiments. The effect of presence/absence of autochthonous microorganisms was also investigated. The results showed that when the autochthonous microorganisms were present, the red soil adsorbed more than 99.95% of MS2 and 98.23% of phiX174, in which most of them were inactivated and/or irreversibly adsorbed. Sterilization led to an increase in MS2 adsorption, while decreased the adsorption of phiX174, indicating that sterilization-induced virus adsorption is virus type dependent. Fewer viruses could be desorbed from the sterilized soil as compared to the nonsterilized soil, probably because sterilization led to an increase in the strength of adsorption force between the soil and viruses. Though the overall virus removal efficiency by the red soil was less than the USPEA required value of 99.99%, we suggest the potential of the red soil as a starting material in removing water heavily polluted with viruses.

© 2010 Elsevier B.V. All rights reserved.

1. Introduction

Many reporters have shown that microbiological pathogens, such as bacteria, protozoa, and viruses, are frequently found in wells and drinking water [1,2]. Sources of pathogens include septic tanks, land application of sewage and sewage sludges, domestic landfills, oxidation ponds, and deep well sewage injection [3], as well as runoff and infiltration from animal waste-amended fields [4]. Virus contamination in various water sources has recently caused much more concern because viruses are far smaller (~0.01–0.1 μm) than bacteria and protozoan cysts and thus have more potential mobility during passage through a media and are removed to a lesser extent by filtration [5]. Additionally, viruses pose a public health threat at a very low level. The USEPA states a limit of two virus particles per 10^7 L of water to achieve an annual infection risk per one people of less than 10^{-4} [6].

Disinfection via chlorination is widely used for preventing the spread of infectious diseases, while it showed less effective to remove viral and protozoa pathogens than to remove bacteria [7,8]. Additionally, high dozes of chlorine produce excessive amounts of disinfection by-products such as trihalomethanes, haloacetic acid, and N-chloramines that are highly toxic to aquatic organisms [9,10]. Other treatments such as UV and ozone are either costly and difficult to implement or generate other disinfection by-products including bromate and chlorite [11,12].

Powell et al. [13] reported that activated carbon might be used for virus removal at the heart of point-of-use devices for individual water treatment, and the removal efficiency was strongly determined by the shape of carbon, however, they did not investigate the fate (adsorption or inactivation) of retained viruses. Layered double hydroxides (LDHs) posses positively charged surfaces and high surface area. You et al. [14] showed that LDHs completely removed viruses from water solution, while the removal was mainly due to sorption of virus to LDH surfaces rather than inactivation, indicating that viruses removed by LDHs were capable of releasing to the environment and had potential contamination capacity. The reverse osmosis treatment can also be used to remove pathogens in the water, with high removal (higher than 5.3 log) being achieved in membrane bioreactor-reverse osmosis systems [15]. More recently, You et al. [16] selected zerovalent iron as removal media for viruses and found that most of the viruses removed from solution were either inactivated or irreversibly adsorbed to the iron. They suggested that zerovalent iron might be potentially useful for disinfecting water. However, for the

* Corresponding author at: Institute of Soil Science, Chinese Academy of Sciences, PO Box 821, 71 East Beijing Road, Nanjing 210008, PR China. Tel.: +86 25 8688 1230; fax: +86 25 8688 1000.
E-mail address: bzhao@issas.ac.cn (B. Zhao).

0304-3894/$ – see front matter © 2010 Elsevier B.V. All rights reserved.
doi:10.1016/j.jhazmat.2010.04.084

economically poorer parts of the world where the environmental health impacts are often the most severe and more disinfection materials are often required, to further low the cost and reduce human's dependence on disinfection agent of chlorine is of utmost urgency.

Virus removal has been examined extensively at field sites where treated wastewater effluents are used for groundwater artificial recharge [17,18], as well as in bench scale porous and fractured aquifer models [19,20]. Virus can be removed from water through adsorption and inactivation. The presence of metal oxides, including Fe and Al oxides, has been proven to play an important role in virus adsorption and survival [21,22]. A significant increase in the retention of MS2 was found in sand under the presence of metals and metal oxides [23]. In both field and laboratory experiments, Ryan et al. [24] reported that inactivation of viruses MS2 and PRD1 occurred on the surfaces of Fe oxide-coated quartz sand. We thus hypothesize that red soil is a particularly interesting material as filtration–sorption media to consider for practical application for virus removal because it is usually rich with Fe and Al oxides and clay and, the most important property is that it is widespread in the world, which may low the cost and reduce human's dependence on disinfection agent of chlorine.

Previous studies showed that the inactivation of viruses in groundwater is controlled by a number of factors including virus types, soil characteristics, groundwater quality, and the presence of autochthonous microorganisms [23,25–27]. However, published results on the effect of autochthonous microorganisms on virus inactivation are generally not consistent [28–30]. Ferguson et al. [31] reviewed that up to date, very few studies have examined the impact of such biological factors as antagonism, competition, and predation on inactivation/disintegration when evaluating transport of pathogens in the field.

The purpose of our study was therefore to evaluate the effectiveness of the red soil to remove two viruses, MS2 and phiX174, by adsorption from dilute aqueous solutions. Bacteriophages MS2 and phiX174 were selected as model viruses in this study because they have been used as surrogates in many studies and because of their structure resemblance to many human enteric viruses [22]. Moreover, reproduction of them in the natural environment is very unlikely [3]. A set of equilibrium and kinetic batch experiments were carried out to investigate: (1) adsorption capacity of viruses onto the red soil, and (2) time-dependent adsorption–desorption behavior and subsequently the fate of retained viruses. We also evaluated the effect of presence/absence of autochthonous microorganisms on virus removal. Results of this study provide basic information for possible utilization of the red soil as one of the filtration–sorption materials in water treatment devices.

2. Materials and methods

2.1. Red soil sampling and treatment

The red soil used in the present study was obtained from the Ecological Experimental Station of Red Soil of the Chinese Academy of Sciences located in Yingtan, Jiangxi Province, China (116°55′30″E longitude and 28°15′20″N latitude). The soil was taken from the top 15 cm, transported to laboratory, air-dried, and ground to pass a 20-mesh sieve before use. The red soil is derived from quaternary red clay [32], and its basic properties before and after sterilization treatment (see next paragraph for the description of sterilization treatment) are presented in Table 1.

Table 1
Selected basic properties of the red soil before and after sterilization treatment[a].

		Before sterilization	After sterilization
pH (H_2O)[b]		4.62 ± 0.01	4.36 ± 0.00
OM[c]	g/kg	4.18 ± 0.18	4.36 ± 0.05
DOC[d]	mg/kg	35.77 ± 0.20	66.85 ± 0.24
CEC[e]	mmol/kg	111.96 ± 11.19	101.29 ± 6.27
Free Fe_2O_3[f]	mg/kg	32,758.92 ± 488.00	33,187.92 ± 238.04
Free Al_2O_3[f]	mg/kg	7353.30 ± 213.01	7400.54 ± 109.09
Amorphous Fe_2O_3[g]	mg/kg	3241.33 ± 150.72	3623.86 ± 216.88
Amorphous Al_2O_3[g]	mg/kg	2593.34 ± 156.60	2746.07 ± 124.74
Zero point of charge[h]		2.61	2.65
Specific surface area[i]	m^2/g	31.28	30.21
Sand (2–0.02 mm)[j]	% (wt)	38.47 ± 4.04	ND[k]
Silt (0.02–0.002 mm)[j]	% (wt)	23.68 ± 2.50	ND[k]
Clay (<0.002 mm)[j]	% (wt)	37.46 ± 2.07	ND[k]

[a] The values are means ± standard deviations (n = 3).
[b] 1:2.5 soil to water suspension.
[c] OM – organic matter: dichromate method [33].
[d] DOC – dissolved organic carbon: water extractable (1:2 soil to water).
[e] CEC – cation exchange capacity: ammonium acetate method [34].
[f] Free Fe_2O_3 and free Al_2O_3: dithionite-citrate-bicarbonate method [35].
[g] Amorphous Fe_2O_3, amorphous Al_2O_3: acid ammonium oxalate method [35].
[h] Zero point of charge: potentiometric titration method [36].
[i] Specific surface area: calculated from eight-point N_2 Brunauer–Emmett–Teller (BET) gas adsorption isotherm using an ASAP 2010 surface area analyzer (Micromeritics, Norcross, GA, USA).
[j] Sand, silt, and clay: Pipet method [37].
[k] ND: not determined.

The sterilized soil was autoclaved at 121 °C and 0.105 MPa for 3 h every 24 h, and the sterilization was repeated for 3 times. The air-dried red soil before sterilization contained bacteria, fungi, and actinomycetes of $(1.84 \pm 0.32) \times 10^5$, $(3.48 \pm 0.39) \times 10^2$, and $(2.30 \pm 0.17) \times 10^2$ cfu/g, respectively. After sterilization treatment, no microorganisms were observed in the sterilized soil, indicating that the sterilization method is effective on killing microorganisms in the air-dried soil.

The determination of mineral composition of the red soil before and after sterilization treatment was conducted on clay particles (<0.002 mm), which was obtained from the soil as described by Hseung [36].

2.2. Virus and plaque assay

Both MS2 and phiX174 were obtained from the American Type Culture Collection (ATCC 15597B1 and ATCC 13706B1), and their host bacterium are *E. coli* ATCC 15597 and *E. coli* ATCC 13706, respectively. MS2 is an icosahedral single-stranded RNA phage with a diameter of about 26.0 nm [38] and has an isoelectric point of 3.9 [39]. PhiX174 is a spherical single-stranded DNA phage with a diameter of about 23.0 nm [38] and has an isoelectric point of 6.6 [39]. They were harvested and suspended in PBS solution (0.02 M Na_2HPO_4, 0.10 M NaCl, and 0.003 M KCl, pH 7.5), and then purified by chloroform extraction as described by Blanc and Nasser [40]. Propagation of MS2 and phiX174 and preparation of their stock solution has been described in our previous studies [32,41]. Typical stock concentrations ranged between 10^{11} and 10^{12} pfu/ml for MS2, and between 10^7 and 10^8 pfu/ml for phiX174. All the virus suspension used in the study was prepared from PBS solution, and all the experiments were carried out at 4 °C. The low temperature of 4 °C and PBS solution for virus suspension preparation were generally considered to be favorable for keeping virus alive and has been used in various studies [27,42,43].

Bacteriophages were assayed by the double-layer overlay method [44]. Briefly, 0.1 ml of log-phase host *E. coli* and 0.1 ml of diluted virus sample solution were mixed in a centrifuge tube. After placing the tube in a 37 °C water bath for 20 min (the time necessary for the adsorption of the phage onto its host bacterium), the mixture was combined with molten soft-agar medium (4.5 ml) maintained at 42 °C in a tube and poured onto three separate Petri dishes had already contained 15 ml of solid medium. The plates were solidified for 10 min and incubated at 37 °C 12 h for MS2, 5 h for phiX174.

Viable virus concentration was determined by counting the number of plaques in the host lawn and reported as plaque-forming units per milliliter (pfu/ml). Only dilutions that resulted in 10–300 plaques per plate were accepted for quantification. Each reported virus concentration is the average of three replicate plates.

2.3. Equilibrium adsorption experiments

The equilibrium adsorption experiments were conducted to investigate the adsorption capacity of viruses onto the red soil. Adsorption of virus to both nonsterilized and sterilized red soil was determined at 4 °C to avoid thermal inactivation of the virus. The method consisted of adding virus stock solution to a 50 ml polypropylene centrifuge tube containing 34 g (dry weight) of the soil (solution:soil ratio = 1.0), sealing and mixing for 3 h at 4 °C by shaking the soil-virus suspensions at 300 rpm. The equilibration time of 3 h was selected based on the results of our previous study that showed 3 h to be sufficient for the equilibrium adsorption to reach [41]. The tubes were filled completely (free of headspace) with virus solution and were devoid of trapped air bubbles during filling and capping of the tubes to avoid potential virus inactivation at the air-water interface. Six different virus stock concentrations ranging from 10^2 to 10^8 pfu/ml were used. Each concentration was diluted from the same virus stock solution with PBS solution. The suspension was then centrifuged for 30 min at 12,096 × g at 4 °C and viable virus concentration was assayed. Control tubes received only virus solution and were treated in the same manner as the experimental tubes. There were three replicates for each concentration.

Virus sorption was calculated with the following formula:

$$S = \left[\frac{C_i - C}{M}\right]$$

where C_i, C, and S are, respectively, the concentrations of virus in the control liquid phase (pfu/ml), in the experimental liquid phase (pfu/ml), and adsorbed to the soil (pfu/g), and M is the total mass of soil per unit volume of virus suspension (g/ml) used in each batch experiment. The sorbed-phase concentration calculated using the above expression includes both attached and inactivated virus because the assay quantifies viable viruses only.

2.4. Kinetic adsorption–desorption experiments

The kinetic adsorption–desorption experiments were conducted to investigate time-dependent adsorption–desorption behavior and subsequently the fate of retained viruses. The adsorption was initiated by adding 2 ml virus stock solution with the concentrations of 10^6–10^7 pfu/ml to a series (42) of 50 ml glass centrifuge tubes containing 2 g (dry weight) of the soil (solution:soil ratio = 1.0), sealing and mixing at 4 °C by shaking the soil-virus suspensions at 300 rpm. At different elapsed times of 1/6, 1/2, 1, 2, 3, 6, 12, and 24 h during the initial 24 h, three tubes were chosen at random. The suspensions were transferred from experiment tubes to polypropylene centrifugation tubes and followed by centrifugation and assay as described above. Subsequently, the used tubes were discarded.

To examine whether the viruses retained by the solid matrix were adsorbed or inactivated, desorption experiments were carried out following the 24 h adsorption by extracting the adsorbed soil with 3% beef extract solution with 0.04 M sodium pyrophosphate of pH 9.4, which has been proven to effectively detach viruses adsorbed to different surfaces [14]. At the end of 24 h adsorption, 26 ml of beef extract solution was immediately added to the additional 18 tubes. The soil-virus suspensions were vortex mixed for a few seconds and kept shaking at 4 °C at 300 rpm, maintaining a pH of 9.4, adjusted as necessary with 1N HCl or 1N NaOH. At different elapsed times of 1/3, 1/2, 1, 2, 5, and 12 h in the next 12 h, three tubes were chosen at random. The suspensions were transferred from experiment tubes to polypropylene centrifugation tubes and followed by centrifugation and assay as described above. The used tubes were discarded after each sampling. Control tubes were performed as the same way as for the experimental tubes.

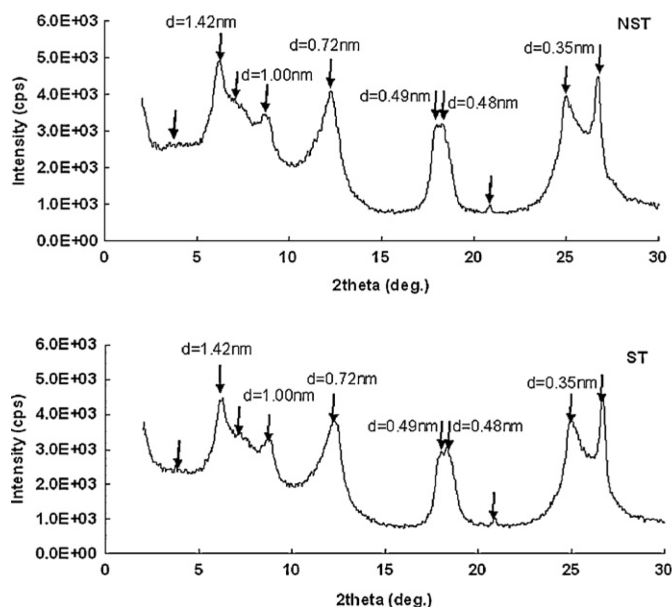

Fig. 1. X-ray diffraction pattern of nonsterilized (NST, top) and sterilized (ST, bottom) red soil. The patterns were obtained using an X-ray diffractometer (XRD) (Philips X'Pert PRO) at ambient temperature with a CuKα radiation source generated at 40 kV and 40 mA (Magixpro, the Netherland).

2.5. Statistical analysis

Analysis of variance (ANOVA) was used to examine the reaction time on virus desorption. Significant differences of means in different reaction time were judged by Duncan multiple comparison tests, with statistical difference level choosing 5%. All statistical analyses were performed using SPSS version15.0.

3. Results and discussion

3.1. Properties of the red soil before and after sterilization

The XRD patterns of the clay particles of the red soil before and after sterilization treatment are illustrated in Fig. 1, which showed that before sterilization there were 10 peaks within the 2 theta of 0–30°. The *d*-value of 0.72 and 0.35 nm, at 12.26° and 24.94°, respectively, matched with standard kaolinite; the value of 1.00 and 0.49 nm, at 8.84° and 18.04°, respectively, with standard hydromica; the value of 1.42 nm at 6.24° with vermiculite, and the value of 0.48 nm at 18.30° with gibbsite. The results indicated that kaolinite, gibbsite, vermiculite and hydromica are the major minerals in the clay fraction of the red soil. After sterilization, the pattern did not show difference from that obtained before sterilization (Fig. 1). Our results are consistent with those reported by previous studies, which showed that the effect of autoclaving on X-ray characteristics of clay minerals was negligible [45,46].

The soil was characterized by low pH and organic matter content, high contents of clay and Fe and Al oxides, and high specific surface area (Table 1). These characteristics are reported to be favorable for virus adsorption [23,26,27,41,47–49]. Sterilization did not seem to significantly affect theses characteristics (Table 1).

Table 2
MS2 and phiX174 batch adsorption data from experiments performed with the nonsterilized red soil.

Control blank concentration (pfu/ml)	Final concentration (pfu/ml)	Fraction adsorbed (%)	Sorption capacity (pfu/g)
MS2			
$(3.06 \pm 0.14) \times 10^2$	0	100	3.06×10^2
$(2.16 \pm 0.06) \times 10^3$	0	100	2.16×10^3
$(4.05 \pm 1.06) \times 10^4$	0	100	4.05×10^4
$(8.30 \pm 2.12) \times 10^5$	$(2.30 \pm 0.52) \times 10^2$	99.97	8.30×10^5
$(2.05 \pm 0.35) \times 10^6$	$(1.02 \pm 0.13) \times 10^3$	99.95	2.05×10^6
$(6.85 \pm 0.78) \times 10^7$	$(1.53 \pm 0.31) \times 10^4$	99.98	6.85×10^7
PhiX174			
$(2.85 \pm 0.55) \times 10^2$	0	100	2.85×10^2
$(2.70 \pm 0.94) \times 10^3$	0	100	2.70×10^3
$(1.36 \pm 0.15) \times 10^4$	$(2.40 \pm 2.45) \times 10^2$	98.23 ± 1.81	1.36×10^4
$(1.28 \pm 0.67) \times 10^5$	$(5.00 \pm 0.71) \times 10^2$	99.61 ± 0.52	1.28×10^5
$(1.25 \pm 0.15) \times 10^6$	$(2.50 \pm 0.71) \times 10^3$	99.80 ± 0.05	1.25×10^6
$(1.43 \pm 0.36) \times 10^7$	$(1.48 \pm 0.35) \times 10^5$	98.96 ± 0.26	1.43×10^7

The values are means ± standard deviations ($n = 3$).

Table 3
MS2 and phiX174 batch adsorption data from experiments performed with the sterilized red soil.

Control blank concentration (pfu/ml)	Final concentration (pfu/ml)	Fraction adsorbed (%)	Sorption capacity (pfu/g)
MS2			
$(2.16 \pm 0.06) \times 10^3$	0	100	2.16×10^3
$(4.05 \pm 1.06) \times 10^4$	0	100	4.05×10^4
$(8.30 \pm 2.12) \times 10^5$	0	100	8.30×10^5
$(2.58 \pm 0.23) \times 10^6$	$(9.40 \pm 4.10) \times 10^1$	100	2.58×10^6
$(2.71 \pm 0.37) \times 10^7$	$(1.53 \pm 0.47) \times 10^2$	100	2.71×10^7
$(3.81 \pm 0.77) \times 10^8$	$(3.86 \pm 0.88) \times 10^3$	100	3.81×10^8
PhiX174			
$(2.85 \pm 0.55) \times 10^2$	$(9.42 \pm 6.34) \times 10^1$	66.96 ± 22.25	$(1.90 \pm 0.63) \times 10^2$
$(2.70 \pm 0.94) \times 10^3$	$(5.33 \pm 2.26) \times 10^1$	98.02 ± 0.84	$(2.65 \pm 0.02) \times 10^3$
$(2.38 \pm 0.37) \times 10^4$	$(1.05 \pm 0.64) \times 10^3$	95.59 ± 2.69	$(2.27 \pm 0.06) \times 10^4$
$(3.18 \pm 0.52) \times 10^5$	$(1.59 \pm 0.36) \times 10^3$	99.50 ± 0.11	$(3.16 \pm 0.00) \times 10^5$
$(1.25 \pm 0.15) \times 10^6$	$(6.25 \pm 2.16) \times 10^3$	99.50 ± 0.17	$(1.25 \pm 0.00) \times 10^6$
$(1.43 \pm 0.36) \times 10^7$	$(6.06 \pm 4.65) \times 10^5$	95.76 ± 3.25	$(1.37 \pm 4.65) \times 10^7$

The values are means ± standard deviations ($n = 3$).

3.2. Virus adsorption onto the red soil-equilibrium study

The results of adsorption behavior of MS2 and phiX174 at different virus concentrations onto the nonsterilized and sterilized red soil are shown in Tables 2 and 3, respectively. The red soil had high adsorption capacity for MS2, with the virus being completely removed from solution at low initial concentrations (i.e. 3.06×10^2 to 4.05×10^4 pfu/ml); at higher initial concentrations (i.e. 8.30×10^5 to 6.85×10^7 pfu/ml), more than 99.95% of the virus was removed (Table 2). Relative to MS2, the removal efficiency of phiX174 decreased to less than 99.82% at higher initial concentrations, though the values were 100% at two low levels of initial virus concentrations (Table 2). Soil sterilization seemed to increase MS2 adsorption, while significantly decreases the adsorption of phiX174 (Tables 2 and 3). Under the present experimental conditions, MS2 were almost completely adsorbed by the sterilized soil; for phiX174, however, the percentage of virus adsorption decreased from more than 98.23% in the nonsterilized soil to the range of 66.96–99.50% in the sterilized soil (Tables 2 and 3).

Virus adsorption results were described using the Freundlich isotherm, $S = KC^{1/n}$, where S is the quantity of virus adsorbed to soil (pfu/g), C is the concentration remaining in the solution phase (pfu/ml), and K and $1/n$ are constants. Best-fit values of the parameters K and $1/n$ and goodness-of-fit values for nonsterilized and sterilized soils are given in Table 4 for each bacteriophage. The K value indicates adsorptive capacity [50], and the $1/n$ is related to the strength of the adsorption forces between the adsorbate and the adsorbent [13]. The results clearly show that K values were significantly larger for MS2 than for phiX174 regardless of soil sterilization or not, resulting from increased amount of MS2 adsorption and indicating higher adsorptive capacity of MS2, relative to phiX174. Comparing K values between nonsterilized and sterilized red soil, we found that sterilization increased K value for MS2, while decreased for phix174 (Table 4), further indicating the virus-dependent adsorption response to sterilization. In addition, increase in values of $1/n$ was found after soil sterilization for both MS2 and phiX174, suggesting that sterilization might lead to an increase in the strength of adsorption force.

Virus removal efficiency decreased with increase in initial virus concentration is not difficult to understand, as with more virus introduced, adsorption sites were more fully saturated with virus,

Table 4
Estimated Freundlich equation parameters for virus adsorption onto nonsterilized and sterilized red soil.

	MS2	
	Nonsterilized	Sterilized
K	1760.8	31,275.2
$1/n$	1.0811	1.1544
r^2	0.9739	0.8699
	PhiX174	
	Nonsterilized	Sterilized
K	183.8	14.3
$1/n$	0.9845	1.1207
r^2	0.8939	0.8368

The equilibrium adsorption time was 3 h.

resulting in less efficient sorption, which can be expected from simple adsorption theory [51]. In contrast, with less virus application, it is assumed that a greater relative percentage of the applied virus was adsorbed.

Our results of the more MS2 removal than phiX174 are in agreement with many reported results. Under the condition of our experimental pH of 7.5, MS2 would be more negatively charged and more favorable for adsorption on the positively charged Fe or Al oxides in the red soil than phiX174 because of the lower isoelectric point of MS2, relative to phiX174. Chattopadhyay and Puls [52] also explained that the lower sorption of phiX174 compared to MS2 was due to its higher isoelectric point. In addition, MS2 was more hydrophobic than phiX174, and exhibited more hydrophobic interaction with soil surface than phiX174 [53,54]. Further, viruses containing RNA were more sensitive to inactivation to copper or iron ions than those containing DNA [55], which might also contribute to the more inactivation of MS2, relative to phiX174, during the process of adsorption onto the red soil. Chu et al. [21] has testified that the presence of iron oxides was responsible for sorption and inactivation of MS2, while not as evident as that for phiX174.

The fact that soil sterilization resulting in the increase in MS2 removal (including adsorption and inactivation) using batch experiments is consistent with the results presented in our previous study conducted on Ferriudic Cambolsols [41]. However, the results of the decrease in phiX174 removal by sterilization indicate that sterilization-induced adsorption/inactivation is strongly virus type dependent. A possible explanation for the observed increase in MS2 removal while decrease in phiX174 removal by soil sterilization is the increase in dissolved organic carbon (DOC) from sterilization. Results in Table 1 show that after sterilization DOC was about 1.9 times greater when compared with that before sterilization. The increased DOC might promote hydrophobic interactions between MS2 and grain surfaces [56], and inhibit phiX174 adsorption by competing for binding sites [22], because MS2 is a relative hydrophobic virus and phiX174 is a relative hydrophilic virus. Zhuang and Jin [57] suggested that if the dominant mechanism controlling organic matter–virus interaction was electrostatic, virus transport would be promoted, and if the dominant mechanism was hydrophobic, virus transport would be retarded, suggesting that the former resulted in reduced virus adsorption and the later increase in virus adsorption.

Many previous studies have got some contrary results. For example, Ottosson and Stenström [29] found that MS2 had a significantly higher decay rate in the tyndallized sediments than in the non-tyndallized sediments. Alvarez et al. [28] got similar results for virus in groundwater, showing that inactivation of MS2 and poliovirus in the groundwater filtered through a 0.2-μm pore filter was slightly faster than in raw groundwater. However, Nasser et al. [58] and Davies et al. [59] reported more rapid inactivation of MS2 and PRD1 in nonsterilized as compared to sterilized soils. Babich and Stotzky [60] found no significant differences in bacteriophage φ11M15 between natural, autoclaved, or filtered lake waters.

3.3. Time-dependent virus adsorption–desorption and fate of adsorbed viruses-kinetic study

The reaction time was considered as one of the most important aspects for selecting adsorbents for their potential application in removing viruses from aqueous phases. The effect of reaction time on virus sorption by the red soil, followed by desorption from the soil is shown in Fig. 2. From the adsorption side (left side) of the curve, the general trend of more MS2 removal by adsorption, relative to phiX174, regardless of soil sterilization was observed. In addition, sterilization resulted in increase in MS2 adsorption but not for phiX174. At the end of adsorption period (i.e. after 24 h adsorption), the percent of MS2 adsorbed (including adsorbed and

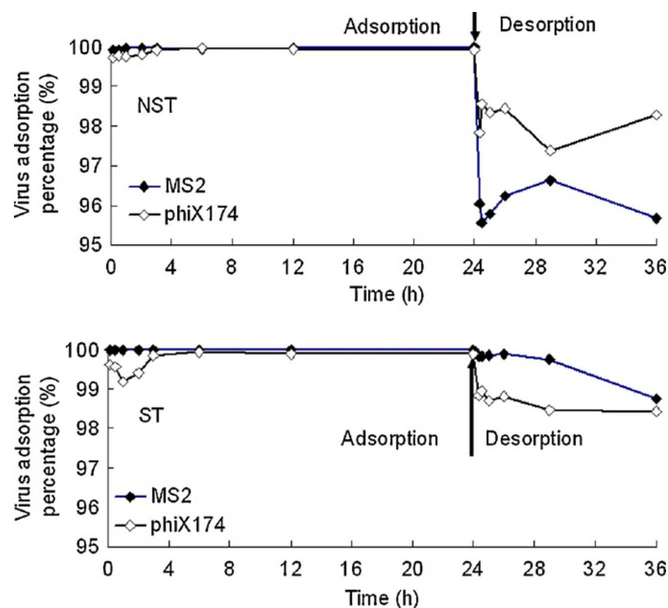

Fig. 2. Percentage of virus adsorption during adsorption and desorption versus reaction time for nonsterilized (NST, top) and sterilized (ST, bottom) red soil. Desorption was initiated after 24 h (see arrow). Standard deviations are not shown because they are covered by the symbols.

inactivated) onto nonsterilized and sterilized soil, respectively, was 99.98% and 100%; for phiX174, the values were 99.92% and 99.87%, respectively. These results were consistent with those we obtained in the above equilibrium study. Although in this study virus inactivation was considered as constant, several studies have indicated that virus inactivation is a time-dependent process [61–63], which should be considered in the further studies.

Removal of MS2 was a rapid progress regardless of sterilization treatment or not, with quasi-equilibrium obtaining with the first 1 h of sorption (Fig. 2). However, 3 h was required for phiX174 to reach quasi-equilibrium (Fig. 2), suggesting that more time is needed to remove phiX174 from an aqueous solution when comparing with MS2.

The calculated amount of adsorbed viruses in adsorption experiment contained both adsorbed and inactivated viruses. It is expected that the adsorbed viable viruses had potential to cause environmental pollution again if the soil conditions changed and met the requirements of virus detachment such as increased pH from fertilization or other pollutants. The desorption side (right side) of the curve (Fig. 2) showed that the general trend of more adsorbed viruses being removed from nonsterilized than from sterilized soil was observed. This can be explained by the increase in the strength of adsorption force from sterilization, which was reflected by increase in the values of $1/n$ calculated from the Freundlich isotherm from the equilibrium study described above. In addition, adsorbed MS2 was more easily to be desorbed than phiX174 in the nonsterilized soil, while the trend was opposite in the sterilized soil (Fig. 2).

The amount of virus desorbed at the end of each desorption (extraction) time was quantified using the results obtained after 24 h adsorption as the final total adsorbed, and subsequently the percentage of total amount desorbed from and retained on the red soil was calculated, based on both the total adsorbed and initial input concentration. These results are presented in Tables 5 and 6. The calculation based on total adsorbed and on initial input concentration was quite similar (Tables 5 and 6). Thus, one set of calculation is capable of describing both sets.

An average of 4.02% of adsorbed MS2 and 1.86% of adsorbed phiX174 were detached from the nonsterilized red soil by beef

Table 5
Mass balance of introduced virus on nonsterilized red soil following 24 h adsorption and subsequent desorption at different reaction time using beef extraction solution.

Reaction time (h)	Input amount (pfu/g)	Total amount adsorbed (pfu/g)	Total amount extracted (pfu/g)	Total amount extracted (percent of adsorbed) (%)	Total amount retained (percent of adsorbed) (%)	Total amount extracted (percent of input) (%)	Total amount retained (percent of input) (%)
MS2							
1/3	3.43×10^7	3.43×10^7	$(1.36 \pm 0.07) \times 10^6$a	3.97 ± 0.19	96.03 ± 0.19	3.97 ± 0.20	96.03 ± 0.20
1/2	3.43×10^7	3.43×10^7	$(1.52 \pm 0.42) \times 10^6$b	4.44 ± 1.22	95.56 ± 1.22	4.43 ± 1.22	95.57 ± 1.22
1	3.43×10^7	3.43×10^7	$(1.45 \pm 0.29) \times 10^6$a	4.22 ± 0.84	95.78 ± 0.84	4.22 ± 0.84	95.78 ± 0.84
2	3.43×10^7	3.43×10^7	$(1.29 \pm 0.36) \times 10^6$a	3.77 ± 1.04	96.23 ± 1.04	3.77 ± 1.04	96.23 ± 1.04
5	3.43×10^7	3.43×10^7	$(1.16 \pm 0.05) \times 10^6$a	3.37 ± 0.16	96.63 ± 0.16	3.37 ± 0.16	96.63 ± 0.16
12	3.43×10^7	3.43×10^7	$(1.49 \pm 0.13) \times 10^6$a	4.33 ± 0.38	95.67 ± 0.38	4.33 ± 0.37	95.67 ± 0.37
Average				4.02 ± 0.74	95.98 ± 0.74	4.02 ± 0.74	95.98 ± 0.74
PhiX174							
1/3	7.25×10^6	7.24×10^6	$(1.58 \pm 0.16) \times 10^5$ab	2.18 ± 0.22	97.82 ± 0.22	2.17 ± 0.22	97.83 ± 0.22
1/2	7.25×10^6	7.24×10^6	$(1.05 \pm 0.29) \times 10^5$b	1.44 ± 0.40	98.56 ± 0.40	1.44 ± 0.40	98.56 ± 0.40
1	7.25×10^6	7.24×10^6	$(1.20 \pm 0.13) \times 10^5$b	1.65 ± 0.19	98.35 ± 0.19	1.65 ± 0.19	98.35 ± 0.19
2	7.25×10^6	7.24×10^6	$(1.12 \pm 0.27) \times 10^5$b	1.55 ± 0.38	98.45 ± 0.38	1.55 ± 0.38	98.45 ± 0.38
5	7.25×10^6	7.24×10^6	$(1.89 \pm 0.15) \times 10^5$a	2.61 ± 0.21	97.39 ± 0.21	2.61 ± 0.21	97.39 ± 0.21
12	7.25×10^6	7.24×10^6	$(1.24 \pm 0.07) \times 10^5$b	1.71 ± 0.09	98.29 ± 0.09	1.71 ± 0.09	98.29 ± 0.09
Average				1.86 ± 0.46	98.14 ± 0.46	1.86 ± 0.46	98.14 ± 0.46

The values are means ± standard deviations ($n = 3$). Values within a column followed by the same letter for each virus are not significantly different at $p < 0.05$. Duncan test.

extract solution at 6 reaction times in the following 12 h desorption period; for the sterilized red soil, the values lowed to 0.33% and 1.30% for MS2 and phiX174, respectively (Tables 5 and 6). Therefore, for the nonsterilized red soil, an average of 95.98% of adsorbed MS2 and 98.14% of adsorbed phiX174 were either inactivated or irreversibly adsorbed, which was noninfective; for the sterilized soil, the values raised to 99.67% for MS2 and 98.70% for phiX174 (Tables 5 and 6). Our ANOVA analysis further showed that very few cases of virus recovery from adsorbed state were time-dependent, and the actual difference in virus recovery at 6 reaction times (i.e. 1/3, 1/2, 1, 2, 5, and 12 h following 24 h adsorption) was minor, indicating that virus desorption almost completed within the first reaction time (i.e. 20 min) under the present experimental conditions, regardless of virus type and soil sterilization treatment. The results suggested that sterilization soils were more favorable for retaining the adsorbed viruses.

Our results of rapid virus adsorption are in agreement with many previous reports. You et al. [14] reported that MS2 sorption by Mg–Al LDH 2 completed with the first 1 h of sorption and, results from Stagg et al. [64] showed that during the first 45 min, about 80% bacteriophage MS2 was adsorbed on the clay. Powell et al. [13] results showed that 90% of MS2 had been adsorbed by powdered activated carbon within the first 2 h. You et al. [16] demonstrated that removal of MS2 and phiX174 by commercial iron was rapid. The red soil's characteristics of rapid virus removal by inactivation and/or irreversible adsorption combined with its high adsorption capacity suggest that the red soil may be a promising sorbent to be used. The soil inherent property of rich with Fe and Al oxides and high content of clay may contribute to this.

Metal and metal oxides could result in orders of magnitude greater adsorption rates, even though in trace amounts [22]. You et al. [16] indicated that metal oxides would cause viruses to disintegrate and become noninfective. Many other studies had also reported that the presence of metal or metal oxides played an important role in virus inactivation [21,65]. On the other hand, the clay minerals of the red soil used in the present study have large

Table 6
Mass balance of introduced virus on sterilized red soil following 24 h adsorption and subsequent desorption at different reaction time using beef extraction solution.

Reaction time (h)	Input amount (pfu/g)	Total amount adsorbed (pfu/g)	Total amount extracted (pfu/g)	Total amount extracted (percent of adsorbed) (%)	Total amount retained (percent of adsorbed) (%)	Total amount extracted (percent of input) (%)	Total amount retained (percent of input) (%)
MS2							
1/3	1.73×10^7	1.73×10^7	$(2.56 \pm 0.64) \times 10^4$a	0.15 ± 0.04	99.85 ± 0.04	0.15 ± 0.04	99.85 ± 0.04
1/2	1.73×10^7	1.73×10^7	$(2.68 \pm 0.87) \times 10^4$a	0.16 ± 0.05	99.85 ± 0.05	0.16 ± 0.05	99.85 ± 0.05
1	1.73×10^7	1.73×10^7	$(2.19 \pm 0.42) \times 10^4$a	0.13 ± 0.02	99.87 ± 0.02	0.13 ± 0.02	99.87 ± 0.02
2	1.73×10^7	1.73×10^7	$(1.93 \pm 0.07) \times 10^4$a	0.11 ± 0.00	99.89 ± 0.00	0.11 ± 0.00	99.89 ± 0.00
5	1.73×10^7	1.73×10^7	$(3.97 \pm 0.60) \times 10^4$a	0.23 ± 0.03	99.77 ± 0.03	0.23 ± 0.03	99.77 ± 0.03
12	1.73×10^7	1.73×10^7	$(2.13 \pm 0.30) \times 10^5$b	1.23 ± 0.17	98.77 ± 0.17	1.23 ± 0.17	98.77 ± 0.17
Average				0.33 ± 0.41	99.67 ± 0.41	0.33 ± 0.41	99.67 ± 0.41
PhiX174							
1/3	1.17×10^7	1.17×10^7	$(1.36 \pm 0.41) \times 10^5$a	1.17 ± 0.35	98.83 ± 0.35	1.17 ± 0.35	98.83 ± 0.35
1/2	1.17×10^7	1.17×10^7	$(1.22 \pm 0.32) \times 10^5$a	1.05 ± 0.28	98.95 ± 0.28	1.05 ± 0.28	98.95 ± 0.28
1	1.17×10^7	1.17×10^7	$(1.50 \pm 0.34) \times 10^5$a	1.29 ± 0.29	98.71 ± 0.29	1.29 ± 0.29	98.71 ± 0.29
2	1.17×10^7	1.17×10^7	$(1.37 \pm 0.08) \times 10^5$a	1.18 ± 0.07	98.82 ± 0.07	1.18 ± 0.07	98.82 ± 0.07
5	1.17×10^7	1.17×10^7	$(1.77 \pm 0.14) \times 10^5$a	1.52 ± 0.12	98.48 ± 0.12	1.52 ± 0.12	98.48 ± 0.12
12	1.17×10^7	1.17×10^7	$(1.82 \pm 0.44) \times 10^5$a	1.56 ± 0.37	98.44 ± 0.37	1.56 ± 0.37	98.44 ± 0.37
Average				1.30 ± 0.29	98.70 ± 0.29	1.30 ± 0.29	98.70 ± 0.29

The values are means ± standard deviations ($n = 3$). Values within a column followed by the same letter for each virus are not significantly different at $p < 0.05$. Duncan test.

surface areas, which might also contribute to high sorption capacity for viruses [25,27].

4. Conclusions

In this study, we demonstrated that the red soil with high contents of Fe and Al oxides and clay from south part of China had relatively high capacity for virus adsorption, and further demonstrated that most of the adsorbed viruses were inactivated and/or irreversibly adsorbed, meaning that most of the adsorbed viruses were noninfective. The effect of soil sterilization on virus adsorption was virus type dependent, while soil sterilization consistently decreased virus desorption, suggesting that soil microbial composition might has potential influence on virus removal. It must be emphasized that the red soil, which is widespread disturbed and as a natural material, is an inexpensive and low-risk material for the human health. Though the overall virus removal efficiency by the red soil obtained from the present study was less than the USPEA required value of 99.99%, especially when the solution virus concentration was more than 10^4 pfu/ml, the red soil can be used as a promising adsorbent for virus pollution control. In reality red soil would be more effective to remove viruses because real virus concentrations in the environment are generally lower than 10^5 pfu/ml [16]. However, for practical use, further investigations are needed to examine the impact of some environmental factors such as the presence of various colloids or some anions and cations on virus removal, and to evaluate virus removal by the red soil under flow conditions. Information from these investigations will provide theoretical basis for effectively evaluating the red soil as a potential virus adsorbent.

Acknowledgements

The work was financially supported by the National Natural Science Foundation of China (grant number 40971129) and National Natural Science Foundation of Jiangsu Province (grant number BK2007535).

References

[1] M. Abbaszadegan, P. Stewart, M.W. LeChevallier, A strategy for detection of viruses in groundwater by PCR, Appl. Environ. Microbiol. 65 (1999) 444–449.
[2] M. Abbaszadegan, P. Stewart, M.W. LeChevallier, J.S. Rosen, C.P. Gerba, Occurrence of Viruses in Groundwater in the United States, Interim Report, American Water Works Service Company, Voorhees, New Jersey, 1998.
[3] Y. Jin, M. Flury, Fate and transport of viruses in porous media, Adv. Agron. 77 (2002) 39–102.
[4] S.W. McMurry, M.S. Coyne, E. Perfect, Fecal coliform transport through intact soil blocks amended with poultry manure, J. Environ. Qual. 27 (1998) 86–92.
[5] D.M. Quanrud, S.M. Carroll, C.P. Gerba, R.G. Arnold, Virus removal during simulated soil-aquifer treatment, Water Res. 37 (2003) 753–762.
[6] U.S. EPA, National primary drinking water regulations: Ground Water Rule, proposed rules, Federal Register 65, Washington, D.C., 2000, pp. 30193–30274.
[7] P. Payment, R. Armon, Virus removal by drinking water treatment processes, Crit. Rev. Environ. Contr. 19 (1989) 15–31.
[8] R.J. Bull, C.P. Gerba, R.R. Trussell, Evaluation of the health risks associated with disinfection, Crit. Rev. Environ. Contr. 20 (1990) 77–113.
[9] J.S. Jensen, G.R. Helz, Rate of reduction of N-chlorinated peptides by sulfite: relevance to incomplete dechlorination of wastewaters, Environ. Sci. Technol. 32 (1998) 516–522.
[10] W.A. MacCrehan, J.S. Jensen, G.R. Helz, Detection of sewage organic chlorination products that are resistant to dechlorination with sulfite, Environ. Sci. Technol. 32 (1998) 3640–3645.
[11] W.A. MacCrehan, M. Bedner, G.R. Helz, Making chlorine greener: performance of alternative dechlorination agents in wastewater, Chemosphere 60 (2005) 381–388.
[12] P.R. Gogate, A.B. Pandit, A review of imperative technologies for wastewater treatment II: hybrid methods, Adv. Environ. Res. 8 (2004) 553–597.
[13] T. Powell, G.M. Brion, M. Jagtoyen, F. Derbyshire, Investigating the effect of carbon shape on virus sorption, Environ. Sci. Technol. 34 (2000) 2779–2783.
[14] Y. You, G.F. Vance, D.L. Sparks, J. Zhuang, Y. Jin, Sorption of MS2 bacteriophage to layered double hydroxides: effect of reaction time, pH, and competing anions, J. Environ. Qual. 32 (2003) 2046–2053.
[15] A.M. Comerton, R.C. Andrews, D.M. Bagley, Evaluation of an MBR–RO system to produce high quality reuse water: microbial control, DBP formation and removal, Water Res. 39 (2005) 3982–3990.
[16] Y. You, J. Han, P.C. Chiu, Y. Jin, Removal and inactivation of waterborne viruses using zerovalent iron, Environ. Sci. Technol. 39 (2005) 9263–9269.
[17] R. Anders, C.V. Chrysikopoulos, Virus fate and transport during artificial recharge with recycled water, Water Resour. Res. 41 (2005) W10415, doi:10.1029/2004WR003419.
[18] C. Masciopinto, R. Lamantia, C.V. Chrysikopoulos, Fate and transport of pathogens in a fractured aquifer in the Salento area, Italy, Water Resour. Res. 44 (2008) W01404, doi:10.1029/2006WR005643.
[19] C.V. Chrysikopoulos, C. Masciopinto, R. Lamantia, I.D. Manariotis, Removal of biocolloids suspended in reclaimed wastewater by injection in a fractured aquifer model, Environ. Sci. Technol. 44 (2010) 971–977.
[20] R. Anders, C.V. Chrysikopoulos, Transport of viruses through saturated and unsaturated columns packed with sand, Transp. Porous Med. 76 (2009) 121–138.
[21] Y. Chu, Y. Jin, M.V. Yates, Virus transport through saturated columns as affected by different buffer solutions, J. Environ. Qual. 29 (2000) 1103–1110.
[22] J.F. Schijven, S.M. Hassanizadeh, Removal of viruses by soil passage: overview of modeling, processes, and parameters, Crit. Rev. Environ. Sci. Technol. 30 (2000) 49–127.
[23] Y. Chu, Y. Jin, M. Flury, M.V. Yates, Mechanisms of virus removal during transport in unsaturated porous media, Water Resour. Res. 37 (2001) 253–263.
[24] J.N. Ryan, R.W. Harvey, D. Metge, M. Elimelech, T. Navigato, A.P. Pieper, Field and laboratory investigations of inactivation of viruses (PRD1 and MS2) attached to iron oxide-coated quartz sand, Environ. Sci. Technol. 36 (2002) 2403–2413.
[25] C.P. Gerba, Applied and theoretical aspects of virus adsorption to surfaces, Adv. Appl. Microbiol. 30 (1984) 133–168.
[26] C.P. Gerba, S.M. Goyal, I. Cech, G.F. Bogdan, Quantitative assessment of the adsorptive behavior of viruses to soils, Environ. Sci. Technol. 15 (1981) 940–944.
[27] Y. Chu, Y. Jin, T. Baumann, M.V. Yates, Effect of soil properties on saturated and unsaturated virus transport through columns, J. Environ. Qual. 32 (2003) 2017–2025.
[28] M.E. Alvarez, M. Aguilar, A. Fountain, N. Gonzalez, O. Rascon, D. Saenz, Inactivation of MS-2 phage and poliovirus in groundwater, Can. J. Microbiol. 46 (2000) 159–165.
[29] J. Ottosson, T.A. Stenström, Growth and reduction of microorganisms in sediments collected from a greywater treatment system, Lett. Appl. Microbiol. 36 (2003) 168–172.
[30] D.E. John, J.B. Rose, Review of factors affecting microbial survival in groundwater, Environ. Sci. Technol. 39 (2005) 7345–7356.
[31] C. Ferguson, A.M.D.R. Husman, N. Altavilla, D. Deere, N. Ashbolt, Fate and transport of surface water pathogens in watersheds, Crit. Rev. Environ. Sci. Technol. 33 (2003) 299–361.
[32] Q.Y. Wang, B.Z. Zhao, J.B. Zhang, Y.M. Wang, H. Zhang, X.M. Chen, J.D. Gong, Virus-adsorbing behavior of soil and its significance in natural disinfection, Acta Pedol. Sin. 44 (2007) 808–816 (in Chinese with English abstract).
[33] D.W. Nelson, L.E. Sommers, Total carbon, organic carbon and organic matter, in: A.L. Page, R.H. Miller, D.R. Keeney (Eds.), Methods of Soil Analysis, Part 2, American Society of Agronomy, Madison, WI, 1982, pp. 539–579.
[34] G.W. Thomas, Exchangeable cations, in: A.L. Page, R.H. Miller, D.R. Keeney (Eds.), Methods of Soil Analysis: Part 2, Chemical and Microbiological Properties, Agronomy No. 9, second ed., American Society of Agronomy and Soil Science Society of America, Madison, WI, 1982, pp. 159–165.
[35] B. Van Raji, M. Peech, Electrochemical properties of some oxisols and alfisols of the tropics, Soil Sci. Soc. Am. Proc. 36 (1972) 587–593.
[36] Y. Hseung, Soil Colloid, vol. 2, Science Press, Beijing, 1985, pp. 1–18 (in Chinese).
[37] Institute of Soil Science, Chinese Academy of Sciences, Methods of Analysis on Soil Physical Properties, Science Press, Beijing, 1978, pp. 26–65 (in Chinese).
[38] J.H. Strauss, R.L. Sinsheimer Jr., Purification and properties of bacteriophage MS2 and of its ribonucleic acid, J. Mol. Biol. 7 (1963) 43–54.
[39] K.S. Zerda, Adsorption of viruses to charge-modified silica, Ph.D. dissertation, Department of Virology and Epidemiology, Baylor College of Medicine, Houston, TX, 1982.
[40] R. Blanc, A. Nasser, Effect of effluent quality and temperature on the persistence of viruses in soil, Water Sci. Technol. 33 (1996) 237–242.
[41] B.Z. Zhao, H. Zhang, J.B. Zhang, Y. Jin, Virus adsorption and inactivation in soil as influenced by autochthonous microorganisms and water content, Soil Biol. Biochem. 40 (2008) 649–659.
[42] S.S. Thompson, M. Flury, M.V. Yates, W.A. Jury, Role of the air–water–solid interface in bacteriophage sorption experiments, Appl. Environ. Microbiol. 64 (1998) 304–309.
[43] Y. Jin, Y. Chu, Y. Li, Virus removal and transport in saturated and unsaturated sand columns, J. Contam. Hydrol. 43 (2000) 111–128.
[44] M.H. Adams, Bacteriophages, Wiley Intersciences, New York, 1959, pp. 443–457.
[45] G. Stotzky, L.E. Demumbrum, Effect of autoclaving on X-ray characteristics of clay minerals, Soil Sci. Soc. Am. Proc. 29 (1965) 225–227.
[46] G. Stotzky, L.T. Rem, Influence of clay minerals on microorganisms, Can. J. Microbiol. 12 (1966) 547–563.
[47] W.D. Burge, N.K. Enkiri, Adsorption kinetics of bacteriophage ϕX-174 on soil, J. Environ. Qual. 7 (1978) 536–542.
[48] R.S. Moore, D.H. Taylor, M.M. Reddy, L.S. Sturman, Adsorption of reovirus by minerals and soils, Appl. Environ. Microbiol. 44 (1982) 852–859.

[49] R.S. Moore, D.H. Taylor, L.S. Sturman, M.M. Reddy, G.W. Fuhs, Poliovirus adsorption by 34 minerals and soils, Appl. Environ. Microbiol. 42 (1981) 963–975.
[50] I.H. Suffet, M.J. McGuire, Activated Carbon Adsorption of Organics from Aqueous Phage, vol. 1, Ann Arbor Publishers, Ann Arbor, New York, 1981.
[51] K.A. Barrett, M.B. McBride, Phosphate and glyphosate mobility in soil columns amended with roundup, Soil Sci. 172 (2007) 17–26.
[52] S. Chattopadhyay, R.W. Puls, Adsorption of bacteriophages on clay minerals, Environ. Sci. Technol. 33 (1999) 3609–3614.
[53] P.A. Shields, S.R. Farrah, Determination of the electrostatic and hydrophobic character of enteroviruses and bacteriophages, in: Abstr. Q-82. Program Abstr. 87th Annu. Meet. Am. Soc. Microbiol., American Society for Microbiology, Washington, D.C., 1987.
[54] C.D. Lytle, L.B. Routson, Minimised virus binding for test barrier materials, Appl. Environ. Microbiol. 61 (1995) 643–649.
[55] J.L. Sagripanti, L.B. Routson, C.D. Lytle, Virus inactivation by copper or iron ions alone and in the presence of peroxide, Appl. Environ. Microbiol. 59 (1993) 4374–4376.
[56] R.C. Bales, S.R. Hinkle, T.W. Kroeger, K. Stocking, Bacteriophage adsorption during transport through porous media: chemical perturbations and reversibility, Environ. Sci. Technol. 25 (1991) 2088–2095.
[57] J. Zhuang, Y. Jin, Virus retention and transport through Al-oxide coated sand columns: effects of ionic strength and composition, J. Contam. Hydrol. 60 (2003) 193–209.
[58] A.M. Nasser, R. Glozman, Y. Nitzan, Contribution of microbial activity to virus reduction in saturated soil, Water Res. 36 (2002) 2589–2595.
[59] C.M. Davies, M.R. Logan, V.J. Rothwell, M. Krogh, C.M. Ferguson, K. Charles, D.A. Deere, N.J. Ashbolt, Soil inactivation of DNA viruses in septic seepage, J. Appl. Microbiol. 100 (2006) 365–374.
[60] H. Babich, G. Stotzky, Reductions in inactivation rates of bacteriophages by clay minerals in lake water, Water Res. 14 (1980) 185–187.
[61] R. Anders, C.V. Chrysikopoulos, Evaluation of the factors controlling the time-dependent inactivation rate coefficients of bacteriophage MS2 and PRD1, Environ. Sci. Technol. 40 (2006) 3237–3242.
[62] C.V. Chrysikopoulos, E.T. Vogler, Estimation of time dependent virus inactivation rates by geostatistical and resampling techniques: application to virus transport in porous media, Stochastic Environ. Res. 18 (2004) 67–78.
[63] Y. Sim, C.V. Chrysikopoulos, One-dimensional virus transport in porous media with time dependent inactivation rate coefficients, Water Resour. Res. 32 (1996) 2607–2611.
[64] C.H. Stagg, C. Wallis, J.L. Melnick, Inactivation of clay-associated bacteriophage MS-2 by chlorine, Appl. Environ. Microbiol. 33 (1977) 385–391.
[65] J.G. Atherton, S.S. Bell, Adsorption of viruses on magnetic particles-I, adsorption of MS2 bacteriophage and the effect of cations, clay and polyelectrolyte, Water Res. 17 (1983) 943–948.