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Disinfection and removal performance for *Escherichia coli* and heavy metals by silver-modified zeolite in a fixed bed column

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ABSTRACT

This study investigates the simultaneous removal of *Escherichia coli* and metals (Pb, Cd and Zn) in a continuous flow system and provides an insight into the mechanisms involved during bacterial cells kill when in contact with silver-modified zeolite. Results showed complete disinfection and metal removal at 570 min contact time, thereafter *E. coli* breakthrough followed by Cd and Zn at 1080 min. Due to the zeolite's high selectivity for Pb removal, no breakthrough was observed up to 7920 min run time. Column performance was influenced by changes in flow rate and bed height, as breakthrough occurred at 240 min when the flow rate was increased from 2 to 5mL/min and the bed height decreased from 1 to 0.5cm. Morphological characterization of treated cells revealed extensive damage and synthesis of nano- and micro-sized silver particles as a part of their defence mechanism. The treated effluent was passed through a non-modified zeolite column with 98% silver recovery achieved. This study showed the capability of this system to simultaneously handle bacterial and heavy metals contamination while providing an insight into the mechanism of disinfection via complex *E. coli*-silver ion interactions that occur during treatment and demonstrating the potential of silver recovery for reuse.

Keywords: Silver-modified zeolite, *Escherichia coli*, Heavy metals, Disinfection, Adsorption

1. Introduction

Water scarcity and water pollution are major global issues. Rapid population growth and the consequent increase in anthropogenic activities have resulted in high demand for scarce water resources, generation of large volumes of wastewater requiring treatment and diffuse pollution of surface and ground water sources [1-5]. Major pollutants of concern include pathogenic microorganisms and persistent heavy metals such as Pb, Cd, and Zn, which are harmful to humans at levels in excess of permissible limits. For instance, the World Health Organization (WHO) guideline values are 0 CFU/mL for faecal coliforms and 0.01mg/L and 0.003 mg/L for Pb and Cd respectively in drinking water, though there is no specific guideline value for Zn which makes water unpleasant at levels in excess of 3mg/L [6]. Hence, more effective treatment technologies are required for their removal from water and wastewater prior to use, discharge into aquatic and terrestrial environments and/or reuse.

Silver is an effective antibacterial agent, which has been immobilized on different inorganic support materials for a wide range of antimicrobial applications including water and wastewater disinfection [7-9, 10-15]. Zeolites have been used as support materials for silver ions with studies demonstrating the effective elimination of microorganisms from aqueous media by silver-modified zeolites [1,16-21]. Most of these studies have focused on the antibacterial properties of silver with the zeolites acting as inert supports with the exception of Krishnani et al. (2012) who investigated the combined disinfection and ammonia removal performance of silver ion-exchanged zeolites [21]. However natural zeolites possess excellent ion exchange and adsorption properties due to their physical and structural characteristics and have been extensively applied for the removal of metal ions from aqueous solutions with high metal removal efficiencies achieved in batch and column systems [2-4,22-26].

Preliminary results showed that silver-modified zeolite exhibited potential for disinfection and heavy metal removal and non-modified zeolites displayed no antibacterial effect [27]. Consequently, the aim of this study is to (1) investigate the performance of silver-modified zeolites for the removal of *E. coli* and metal ions in a continuous fixed-bed column system including the effects of flow rate and bed height on removal performance; (2) provide an insight into the mechanism of disinfection via morphological characterization of treated cells; and (3) from an environmental and economic point of view, evaluate the potential of silver recovery for reuse.

2. Materials and Methods

2.1. Natural and Silver-Modified Zeolites

The natural zeolites-clinoptilolite (NZ) (particle size: 0.2-0.6mm) purchased from Planet Care Products, UK were washed with deionized water to remove surface dust and dried overnight at 60°C.

Silver modification of NZ was achieved via ion exchange by shaking 100g of NZ in 250mL of 3% (w/v) AgNO_3 . The pH of the solution was adjusted to pH 5 to prevent precipitation of silver ions and the solution was shaken for 24 hours in a dark room. The silver-modified zeolites (SZ) were separated, rinsed with deionized water and dried overnight at 60 °C. The amount of silver adsorbed per gram of zeolite was 43.4 mg Ag/g. The chemical composition of representative fractions of NZ and SZ determined by EDX (Energy Dispersive X-ray Spectroscopy) as reported in our previous study [27] confirmed the presence of silver ions exchanged onto SZ.

2.2. Reagents, Microorganism and Aqueous Solutions

Analytical grade reagents were used to prepare metal stock solutions in deionized water. AgNO_3 was purchased from Sigma Aldrich, UK. $\text{Pb}(\text{NO}_3)_2$, $\text{CdN}_2\text{O}_6 \cdot 4\text{H}_2\text{O}$, and $\text{Zn}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$ were purchased from Fisher Scientific, UK.

Escherichia coli NCTC 9001 from the Public Health England Culture Collection used as an indicator of faecal contamination of water was grown overnight in nutrient broth (Oxoid, UK) at 37°C. The cells were pelleted by centrifugation, rinsed to remove excess media and re-suspended in deionized water at 10^9 CFU/mL.

Aqueous solutions were prepared by diluting *E. coli* and metal stock solutions in deionized water to give initial *E. coli* concentration: 10^8 - 10^9 CFU/100ml and initial metal concentrations: 0.5 mg/L Pb, 0.5 mg/L Cd and 0.5 mg/L Zn. The pH of the feed solution was adjusted to 5 to prevent metal precipitation.

2.3. Fixed-Bed Column Experiments

Experiments were carried out using polyethylene columns of 1.5 cm internal diameter and 30 cm height packed with 1-5g of SZ between two supporting layers of glass wool. The feed solution was pumped into the column in down flow mode using a peristaltic pump at 1-5mL/min. Columns were wrapped with aluminium foil to prevent the formation of black silver oxide on the zeolites due to the light sensitivity of silver ions. Prior to each experiment, deionized water was passed through the column for 30 minutes to ensure that the zeolite particles were properly packed to minimize channelling. The influent was monitored and replaced with fresh stock at regular intervals to maintain microbial concentration levels required. All experiments were conducted at room temperature ($22^\circ\text{C} \pm 2$).

The effects of flow rate (1, 2 and 5mL/min) and bed height (0.5, 1 and 2.5 cm) were investigated until breakthrough and exhaustion of disinfection capacity. Control columns packed with 2g of SZ and NZ were fed with *E. coli* solution at 2ml/min and operated under the same conditions.

Samples were collected periodically for immediate microbiological analysis while samples for metal analysis were syringe-filtered, acidified to pH<2 and refrigerated at 4°C.

2.4. Scanning Electron Microscopy

E. coli cells were examined before and after exposure to SZ. Samples (5µL) were dropped or smeared on pre-sterilized silicon substrates and air dried under sterile conditions with no further fixing done [28]. The samples were painted with high purity silver, gold coated to improve conductivity and analysed using a JEOL JSM-7100F Field Emission Scanning Electron Microscope (SEM) equipped with an Energy Dispersive X-ray Detector (EDX).

2.5. Silver Recovery

The potential of recovering silver ions was achieved by passing the treated effluent through a column packed with 1-12g of NZ at flow rates 1-2mL/min at pH 4 for optimum silver uptake.

2.6. Analytical Methods

The membrane filtration technique was used for enumeration of *E. coli* [29]. A quarter strength Ringer's solution tablets (Oxoid UK) were used to prepare Ringer's solution for serial dilution of water samples prior to enumeration of *E. coli*. M-lauryl sulphate broth (MLSB) (Sigma Aldrich, UK) was used for enumeration of *E. coli* using the membrane filtration technique. Aliquots samples (0.1mL) were serially diluted in Ringer's solution and filtered through sterile 0.45 µm membrane filters. The filters were placed on adsorbent pads saturated with MLSB and incubated

overnight at 37°C. *E. coli* colonies were counted and recorded as colony forming units per 100mL (CFU/100mL).

The concentration of metal ions in solutions was determined by ICP-OES (Perkin Elmer ICP-OES Optima 5300DV)

2.7. Theoretical Calculations

Breakthrough curves of effluent pollutant concentration against time were plotted from the results of the fixed-bed columns experiments. For *E. coli* removal, the amount of *E. coli* removed at breakpoint, X_b (CFU) in the column was calculated using the equation:

$$X_b = Q_v \int_0^{T_b} (C_o - C) dT \quad (1)$$

Where Q_v is the volumetric flow rate (L/min), C_o and C are influent and effluent contaminant concentrations at time T (CFU/L), T_b is the breakthrough time.

Similarly, the amount of *E. coli* removed at exhaustion of disinfection capacity, X_e (CFU) in the column was calculated using the equation:

$$X_e = Q_v \int_0^{T_e} (C_o - C) dT \quad (2)$$

Where Q_v is the volumetric flow rate (L/min), C_o and C are influent and effluent contaminant concentrations at time T (CFU/L), T_e is the exhaustion time.

For metal removal, the breakthrough capacity, q_b (mg/g) was calculated using the equation:

$$q_b = [Q_v \int_0^{T_b} (C_o - C) dT] / m \quad (3)$$

Where Q_v is the volumetric flow rate (L/min), C_o and C are influent and effluent contaminant concentrations at time T (mg/L), T_b is the breakthrough time and m is the mass of zeolite (g).

The exhaustion capacity, q_e (mg/g) was calculated using the equation:

$$q_e = [Q_v \int_0^{T_e} (C_o - C) dT] / m \quad (4)$$

Where Q_v is the volumetric flow rate (L/min), C_o and C are influent and effluent contaminant concentrations at time T (mg/L), T_e is the exhaustion time and m is the mass of zeolite (g).

The amount of silver released in the treated effluent (mg) was calculated using the formula:

$$X_{Ag} = Q_v \int_0^T C dT \quad (6)$$

Where X_{Ag} is the amount of silver ions released (mg), Q_v is the volumetric flow rate (L/min) and C is the concentration of silver in the effluent at time T (mg/L), T is the time (min).

3. Results and Discussion

3.1. Disinfection Performance

The breakthrough curve for *E. coli* removal in the SZ column and the corresponding amount of silver released in the treated effluent is shown in Figure 1. For the purpose of evaluating the disinfection performance of the system, the breakpoint was selected as the time when viable bacterial cells were first detected in the treated effluent ($C \cdot 1\text{CFU}/100\text{mL}$) and the column was considered to have reached exhaustion when the *E. coli* breakthrough curves reached a plateau ($C \cdot 10^6\text{-}10^7\text{ CFU}/100\text{mL}$).

As illustrated in Figure 1, complete disinfection (100%) was observed up to 570 min of column operation ($V_b=1140\text{mL}$). As the solution was fed into the column, rapid disinfection and metal sorption occurred within the upper layers of the bed as the disinfection and adsorption zones were established. In the disinfection zone, the concentration of silver released into the solution was sufficient to kill 100% of the cells. However, as the disinfection zone travelled downwards towards the end of the bed, the amount of silver being released decreased until breakthrough occurred. Subsequently, there was a rapid increase in the number of viable *E. coli* cells in the effluent and exhaustion was reached after 1020 minutes of operation indicating that disinfection was no longer taking place. As *E. coli* breakthrough and exhaustion occurred, there was a corresponding decrease in the amount of silver released into the solution from 0.23mg/L at breakthrough to 0.050mg/L (Table 3). This result indicates a direct correlation between the amount of silver released and the disinfection efficiency as reported in other studies [17].

In order to develop an understanding of the *E. coli*-metal interactions occurring within the bed, the morphology of *E. coli* cells before and after treatment was examined using SEM as shown in Figures 2(a)-(f). The typical rod-like gram-negative *E. coli* cells (1-2 μm in length) can be observed in Figures 2(a) and (b) prior to exposure to silver ions and Figure 2(c) shows bacterial cells attached to the zeolite particles during treatment. At higher magnification (Figure 2(d)), the characteristic flat blades of the zeolite particle can be observed with extensively damaged cells attached to the surface. Figures 2(e) and (f) show cell debris present in the treated effluent. Bacterial cells came in contact with silver ions present in the zeolite and released in solution, possibly enhanced by electrostatic interaction between the positively charged ions and the negatively charged cell wall [30-31]. These ions significantly damaged the cell wall resulting in the leakage of intracellular cell contents [10, 32-33] while some ions accumulated within the cell, thus interacting with the thiol groups, proteins and enzymes resulting in agglomeration of cell

proteins and condensation of the cell DNA. This inhibited the cell replication ability and disrupted essential cell functions such as the cell respiration resulting in cell death [8,10,34]. The disinfection action of silver ions also involves the generation of destructive reactive oxygen species within the cell catalyzed by the silver ions, which destroy the cell via oxidation [9,34-36].

Micro and nano-sized crystalline particles were observed on the surface of the zeolite as shown in Figures 2(c) and (d) and surrounding cell debris retrieved from the treated solution (Figure 2(f)). EDX spot analysis revealed that these particles had high concentrations of silver. The intracellular and extracellular synthesis of silver nanoparticles by bacteria including *E. coli*, as a defense mechanism, has been reported in the literature [8,31,37]. Bio-precipitation of metal ions as crystals or oxides via the reduction of metal ions to less toxic zero-valent species is one of a number of resistance mechanisms microorganisms exhibit in response toxic metal ions [9,38-39]. In extracellular synthesis, the bacterial cells release extracellular polymeric substances which bind to the toxic Ag^+ ions reducing them to less reactive metallic particles Ag^0 [8]. Intracellular synthesis can occur within the reductive environment in the cytoplasm [9]. The silver nanoparticles formed are stabilized by the protein molecules involved in the reduction process [37,40]. However, nanoparticle synthesis is said to occur at lower concentrations of silver when the cell is able to protect itself, while at high silver concentrations the cell is overwhelmed by the attacking ions resulting in death [41]. These silver particles, a by-product of *E. coli*-silver ion interaction during disinfection, most likely were retained within the bed. Moreover, less than 6% of the silver exchanged onto the zeolites was being released during treatment thus suggesting that not all silver ions involved in the disinfection process were being released in the treated solutions. Hence, while some bacterial cells were killed by silver ions released (liquid phase

disinfection), some cells were attached to the particles when they came in contact with silver ions within the zeolite matrix (solid phase disinfection).

3.2. Metal Removal Performance

The metal removal performance of the SZ column was investigated simultaneously and the Pb, Cd and Zn breakthrough curves are shown in Figure 3. The metal breakthrough concentrations for Pb and Cd were selected based on the minimum water quality guideline limit of 0.01 and 0.003mg/L respectively. However there is no guideline limit for Zn, hence breakthrough was said to have occurred when 5% of the influent concentration was measured in the treated effluent [6]. The column reached exhaustion when up to 95% of the influent metal concentration was measured in the treated effluent.

Breakthrough of Cd and Zn occurred after 1080 minutes of column operation and exhaustion occurred after 5220 minutes (Table 1). In each metal adsorption zone, metal removal began within the upper layers of the bed. The adsorption zone was established and moved downwards with breakthrough occurring when the zone reached the end of bed and exhaustion when it moved out of the bed. Metal removal involved the mechanisms of both adsorption and ion exchange. This occurred via bulk diffusion of metal ions from the solution to the zeolite surfaces, intra-particle diffusion of metal ions through the zeolite macro pores and meso pores to the microcrystal surfaces where they replaced some of the exchangeable framework cations, adsorption on the internal surfaces of the zeolites and possibly precipitation on the surface of the zeolite due to increase in pH [2, 22, 42-44]. There was an observed increase in the pH of effluent solutions from 5.5 to 6.30. This was due to zeolite hydrolysis and the uptake of H^+ ions from the solutions by the zeolites as reported in other metal removal studies [2,22,42].

There was no breakthrough of Pb after 7920 minutes of column operation thus indicating the higher Pb selectivity of clinoptilolite [4, 23, 42-43]. The column operation had to be stopped due to bacterial clogging the bed. The loss of disinfection efficiency from *E. coli* breakthrough till exhaustion (570-1020 minutes) resulted in the gradual colonization of the bed by bacterial cells and biofilm formation. Biofilm formation on media during column disinfection with silver nanoparticles on silica beads has been reported in the literature [45]. This phenomenon could also explain the irregular shape of the Cd and Zn breakthrough curves as observed in Figure 3. Ideally, after metal breakthrough, the concentration of metal ions in the treated effluent would increase steadily until exhaustion. However, the fluctuating effluent concentrations and the resultant undulating breakthrough curve could be due to the bio-sorption of metal ions by the biofilm and cell debris at some points in different regions of the bed. In practice, the column operation would be terminated prior to breakthrough of a single pollutant. Since *E. coli* breakthrough occurred earlier, the effects of flow rate and bed height on the performance of the system were investigated until exhaustion of disinfection efficiency.

3.3. Effect of Flow Rate

Figure 4 shows the effect of flow rates on *E. coli* removal and Table 2 provides a comparison of the performance of the column at different flow rates. It can be observed that as the flow rate increased from 1mL/min to 5 mL/min, breakthrough occurred earlier resulting in a shorter column service life.

However, increased flow rate did not have a significant effect on the amount of *E. coli* removed at breakthrough $((3.80-4.48) \times 10^9$ CFU). In the bed operated at 5mL/min, Cd breakthrough was observed after 120 minutes of contact as there was insufficient time for intra-particle diffusion of

metal ions into the pores of the zeolite resulting in earlier breakthrough [46]. Hence, the lower the flow rate, the better the metal removal performance of the column [25].

The effect of flow rate on the amount of silver released into the solutions was also investigated and the results are shown in Table 3. It can be observed that total the amount of silver released into the treated effluent varied at different flow rates. As the flow rate increased, the residence time of the solution within the bed decreased, hence the amount of silver released from the zeolites decreased. The breakthrough performance of the column is quite important, as the column would not be operated beyond that time. The flow rate did not significantly affect the disinfection performance of the system under the experimental conditions of this study but affected the metal removal performance. Therefore it must be optimized to ensure that both contaminants are removed effectively.

3.4. Effect of Bed Height

The removal performance of the column at different bed heights Z (0.5-2.5cm) until exhaustion of disinfection capacity was investigated. *E. coli* breakthrough curves are shown in Figure 5 and Table 3 provides a performance comparison of the column at different bed heights.

The results indicate that the service life (breakthrough and exhaustion times) and volumes treated increased with increased bed height as it took a longer time for the disinfection zone to move down and out of the bed. Furthermore, the amount of *E. coli* removed by the column at breakthrough increased from 1.90×10^9 CFU to 9.98×10^9 CFU with an increase in bed height from 0.5 to 2.5 cm (Table 2). This was due to the greater surface area for bacterial attachment and increased availability of silver ions for disinfection as the mass of zeolites increased [17]. Similarly, as shown in Table 3, the amount of silver released into the treated solutions increased with an increase in bed height. At breakthrough, 11.38mg of silver was released from the 2.5 cm

bed, which was more than 25 times the amount released from the 0.5cm bed (0.41mg). The bed height of the column had a greater effect on the removal of metal ions as breakthrough of Pb, Cd and Zn was observed in the 0.5 cm bed at 720, 240 and 480 minutes respectively (not shown), thus indicating that the bed height was insufficient to achieve optimum contaminant removal. This was due to the decrease in available sorption sites for metal removal and insufficient residence time for intra-particle diffusion of metal ions within the zeolite pores. In shorter beds, mass transfer is governed by axial dispersion resulting in decreased diffusion of metal ions [46].

3.5. Silver Recovery

The results of this study have established a correlation between the disinfection efficiency and the amount of silver released in the treated solutions. From an environmental and economic point of view, it is important to close the loop and ensure that there is minimal wastage of silver. Hence, preliminary experiments were undertaken to recover silver from the SZ-treated effluents. This was achieved using beds of non-modified zeolites with a view to conditioning them for use. In the different columns tested 97-98% silver recovery was achieved (Table 4). Furthermore, conditioning the zeolite to a homo-ionic form may slightly enhance the removal efficiency and could result in 100% recovery.

4. Conclusions

The SZ column effectively removed *E. coli* and metal ions from aqueous solutions. In the main SZ column, *E. coli* breakthrough occurred after 570 minutes as the amount of silver released in the effluent decreased, while Cd and Zn breakthrough occurred later after 1080 minutes. Pb breakthrough was not observed during the service life of the column. However, the column had to be stopped after 7920 minutes of operation due to bed clogging (biofilm formation) with loss of disinfection efficiency. The varying *E. coli* and metal breakthrough times observed indicate

the formation of different overlapping contaminant removal zones making up a complex mass transfer zone within the bed. The simultaneous removal of *E. coli* and metal ions involved complex mechanisms including disinfection, ion exchange, adsorption and bio-sorption. SEM micrographs revealed extensively damaged cells and cell debris after treatment and the synthesis of silver micro and nano-sized particles on the zeolite surface and in solution as a defense mechanism. Column service life increased with a decreased flow rate and increase in bed height with the latter having a greater effect on the column performance.

In NZ columns 97-98% recovery of silver was achieved thus demonstrating the possibility of silver recovery for potential reuse. Overall, the results indicate that this system could potentially be developed and effectively applied as a one-step treatment unit process capable for the removal of both bacteria and heavy metals from contaminated aqueous streams.

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Figure Captions

Figure 1. Breakthrough curve for *E. coli* removal in SZ column and silver released in the treated effluent ($C_o = 10^9$ CFU/100mL *E. coli*, 0.4mg/L Pb, 0.5mg/L Cd, 0.5mg/L Zn; pH=5; $Q_v = 2$ mL/min; Z= 1cm)

Figure 2. SEM Images of *E. coli* cells (a, b) prior to treatment (c, d) interaction with SZ (e, f) debris after treatment

Figure 3. Breakthrough curves for metals removal in SZ column ($C_o = 10^9$ CFU/100mL *E. coli*, 0.4mg/L Pb, 0.5mg/L Cd, 0.5mg/L Zn; pH=5; $Q_v = 2$ mL/min; Z= 1cm)

Figure 4. Effect of flow rate on *E. coli* breakthrough in SZ column ($C_o = 10^9$ CFU/100mL *E. coli*, 0.4mg/L Pb, 0.5mg/L Cd, 0.5mg/L Zn; pH=5; Z= 1cm)

Figure 5. Effect of bed height on *E. coli* breakthrough in SZ column ($C_o = 10^9$ CFU/100mL *E. coli*, 0.4mg/L Pb, 0.5mg/L Cd, 0.5mg/L Zn; pH=5; $Q_v = 2$ mL/min)

Tables

Table 1. Experimental results for metals removal in SZ column ($C_o = 10^9$ CFU/100mL *E. coli*, 0.4mg/L Pb, 0.5mg/L Cd, 0.5mg/L Zn; pH=5)

Metals	Breakthrough			Exhaustion		
	T_b (min)	V_b (mL)	q_b (mg/g)	T_e (min)	V_e (mL)	q_e (mg/g)
Cd	1080	2160	0.571	5220	10440	2.510
Zn	1080	2160	0.568	5220	10440	2.439

Table 2. Experimental results for *E. coli* removal in SZ column ($C_o = 10^9$ CFU/100mL *E. coli*, 0.4mg/L Pb, 0.5mg/L Cd, 0.5mg/L Zn; pH=5)

		Breakthrough				Exhaustion		
Z (cm)	m (g)	Q _v (mL/min)	T _b (min)	V _b (mL)	X _b (CFU)	T _e (min)	V _e (mL)	X _e (CFU)
Effect of flow rate								
1	2	1	1320	1320	3.80x10 ⁹	3000	3000	8.60x10 ⁹
1	2	2	570	1140	3.39x10 ⁹	1020	2040	5.95x10 ⁹
1	2	5	240	1200	4.48x10 ⁹	300	1500	5.47x10 ⁹
Effect of bed height								
0.5	1	2	240	480	1.90x10 ⁹	900	1800	7.11x10 ⁹
1	2	2	570	1140	3.39x10 ⁹	1020	2040	5.95x10 ⁹
2.5	5	2	2100	4200	9.98x10 ⁹	2880	5760	1.32x10 ¹⁰

Table 3. Effect of experimental conditions on the amount of silver released during *E. coli* removal in SZ column ($C_o = 10^9$ CFU/100mL *E. coli*, 0.4mg/L Pb, 0.5mg/L Cd, 0.5mg/L Zn; pH=5)

Z (cm)	m (g)	Q_v (mL/min)	Breakthrough			Exhaustion		
			T_b (min)	C_c (mg/L)	X_{Ae} (mg)	T_e (min)	C_c (mg/L)	X_{Ae} (mg)

Effect of flow rate								
1	2	1	1320	0.203	2.535	3000	0.047	2.684
1	2	2	570	0.203	3.012	1020	0.050	3.100
1	2	5	240	0.078	1.459	300	0.058	1.793
Effect of bed height								
0.5	1	2	240	0.188	0.411	900	0.033	0.501
1	2	2	570	0.203	3.012	1020	0.050	3.100
2.5	5	2	2100	0.469	11.372	2880	0.063	11.787

Table 4. Silver Recovery in NZ column (pH=4)

Z (cm)	m (g)	Q_v (mL/min)	Ag from SZ (mg/L)	V (mL)	% Recovery
1	2	2	2.651	1200	98.56
2.5	5	2	2.432	1200	97.57
12	24	1	2.767	360	98.25

Figure 1

Figure 3

Figure 4

Figure 5

Highlights

- Silver-modified zeolite column simultaneously removed *Escherichia coli* and metals
- Complete removal of *E. coli* achieved after 570 min followed by Cd and Zn (1080 min)
- No Pb breakthrough observed during the service life of the column (7920 min)
- SEM revealed damaged and lysed cells and *E. coli*-synthesized Ag particles
- 97-98% Ag recovery achieved in the non-modified natural zeolite column