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Removal of Mg from spring water using natural clinoptilolite

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ABSTRACT: Natural zeolitic tuff from Brus (Serbia) consisting mostly of clinoptilolite (about 90%) has been investigated for the reduction of the Mg concentration in spring water. The sorption capacity of the zeolite is relatively low (about 2.5 mg Mg g^{-1} for the initial concentration of 100 mg Mg dm^{-3}). The zeolitic tuff removes Mg from water solutions by ion exchange, which has been demonstrated by energy dispersive X-ray analysis (EDS). The extent of ion exchange was influenced by the pH and the initial Mg concentration. Kinetic studies revealed that Lagergen's pseudo-second order model was followed. Intra-particle diffusion of Mg^{2+} influenced the ion exchange, but it is not the rate-limiting step. Rather than having to dispose of the Mg-loaded (waste) zeolite, a possible application was tested. Addition to a wastewater with a low concentration of Mg showed that it could successfully make up for the lack of Mg micronutrient and, accordingly, enabled the growth of phosphate-accumulating bacteria *A. Junii*, increasing the amount of phosphate removed from the wastewater.

KEYWORDS: natural zeolite, Igroš Vidojević deposit, Brus, Serbia, clinoptilolite, Mg removal, bacteria, water softening.

Pollution of water stocks and food with magnesium salts can cause serious health problems such as hypermagnesemia, nausea, low blood pressure and diarrhea (FAO/WHO, 1998). Recommended daily uptake of Mg from both food and drinking water is 65110 mg for children aged up to 10 years, and 360 mg for adults (FAO/WHO, 1998). However, spring waters from mountainous regions formed largely of dolomite rock, such as the area of the city Raška (Southwest Serbia), contain up to 200 mg Mg dm^{-3} (which is characteristic for mineral waters) and, therefore, it is not acceptable for public use (allowed concentration being 50 mg Mg dm^{-3}, Official Gazette SRJ, 1998, 1999). The spring water from abandoned dolomite mines at Potkop near the town of Raška has a stable physico-chemical quality according to experimental data from the Raška water supply, with a negligibly low level of microbiological pollution; this makes the water suitable for the use in a water-supply system (Table 1).

Table 1 shows that high pH and Mg^{2+} concentrations are the only disadvantages of the water. The problem has been overcome by adding acid to decrease the pH and by dilution using the river water (Brvenica) to bring the Mg^{2+} content to an acceptable value. However, the variable quality of the river water necessitated the use of large quantities of coagulants and flocculants to obtain potable water. The spring water from the Potkop shaft is not used for any other purposes and it is remarkably well protected from outside pollution. Therefore, it would be profitable to use its complete output for drinking water, provided the excess of Mg^{2+} could be removed by a suitable method.

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TABLE 1. Maximum values of basic chemical parameters in the water from the Potkop well as analysed in the period 2006–2010.

Parameter	Value/ conc. (mg dm^{-3})	Max. permissible value/conc. (mg dm^{-3})	Parameter	Value (μg dm^{-3})
pH	9.5	6.8–8.5	Mn	0.15
Turbidity	0.5 (NTU)	1.2 (NTU)	Fe	35
$KMnO_4$	8	8	Ni	0.4
Ca^{2+}	10	200	Cu	0.15
Mg^{2+}	90	50	Zn	0.4
SO_4^{2-}	7	250	As	0.22
NO_3^-	5	50	Se	0.26
NO_2^-	0.005	0.03	Mo	0.02
NH_4^+	0.05	0.1	Cd	0.003
PO_4^{3-}	0.002	0.003	Sb	0.01
B	2	300	Ba	0.41
Al	0.45	200	Hg	0.05
Cr	1.2	50	Pb	0.02

In order to reduce water hardness, various methods have been applied such as chemical precipitation, ion exchange, reverse osmosis or the use of electromembrane systems (Marakov, 2007). Most of these methods suffer from high-energy consumption, high cost of operation and/or environmental incompatibility. A simple procedure based on the application of local sources is a necessity for the establishment of a sustainable strategy. In this context ion exchange using natural materials seems to be one of the most promising methods (Caputo & Pepe, 2007). Accordingly, zeolite obtained from waste pumice has recently been investigated for the reduction of the water hardness (Arrigo *et al*., 2007). A capacity of about 56 mg Ca^{2+} and 5 mg Mg^{2+} per g of the zeolite was observed. Moreover, it was reported that the capacity of synthetic zeolites X and Y for water softening can be improved by their treatment with sodium carbonate, whereas zeolite Na-A exhibited high effectiveness under warm conditions (Mao *et al*., 1994).

Natural zeolites are locally available materials. They are low-cost and environmentally friendly and, in the last decade, these materials have been extensively studied for the removal of various metal cations from water solutions (Wang & Peng, 2010). There are several deposits in Serbia with zeolitic tuffs enriched with clinoptilolite, whose 3-D aluminosilicate lattice is characterized by large intersecting open channels occupied by ion-exchangeable cations.

In this study we have used the zeolitic tuff from the Igroš Vidojević deposit located in Brus (about 60 km from Raška). Its composition and sorption ability towards particular elements have not been investigated so far. Here, we have investigated this tuff as a water softener in an installation for obtaining potable water. Also, we have considered a possible use of the exhausted zeolite as a support for the phosphate (P)-accumulating bacteria *Acinetobacter junii*. The P-accumulating bacteria immobilized onto naturally occurring materials offer a promising alternative for improving the enhanced biological phosphorus removal from wastewaters (Hrenović *et al*., 2005).

EXPERIMENTAL METHODS

Sorption of Mg

The zeolite (CLI) was obtained from the large sediment accumulation of Igroš Vidojević in Brus (Serbia). Grain sizes in the range 12 mm were chosen for the experiments. In order to improve the tuff's sorption capacity, the as-received sample was pre-treated with an aqueous NaCl solution (2.0 mol dm^{-3}) (Cerjan Stefanović *et al*., 2007). The suspension was magnetically stirred for 24 h at 298 K and the sample (Na-CLI) was then washed in distilled water and dried in an oven at 378 K.

In the pre-treatment, in all sorption and kinetic experiments, and in the post-treatment experiments a (solution volume)/(solid weight) ratio of 100 cm^3:1 g was used. The sorption isotherm was determined at 298K using a batch method. Na-CLI was placed in a solution of $MgCl_2$ of chosen concentration in the range of 20 to 100 mg Mg dm^{-3}. The suspension of Na-CLI and the chosen Mg solution was shaken at about 100 rpm in a thermostated water bath (Memmert WPE 45). The Mg-loaded solid (Mg-CLI) was then recovered by filtration. The rate of sorption was studied at 298 K using solutions with an initial Mg concentration of 20100 mg Mg dm^{-3}. The suspension was shaken at a rate of about 100 rpm for time periods from 20 min to 180 min. The solid was then separated by filtration.

For all sorption and kinetic experiments the pH was adjusted to 8 (*vide infra*) by adding a solution of NaOH. Also, in order to approach similarity between experimentally used solutions and the spring water, a solution of $CaCl_2$ (10 mg dm^{-3}) was added to achieve a Ca/Mg molar ratio of 1:10.

Influence of pH

Batch experiments were conducted at different pH values to establish an optimal value for studying the sorption isotherm. The experiments were performed by placing the Na-CLI in 100 mg dm^{-3} Mg solution. The studied pH range was from 5 to 9 and it was adjusted by using 0.1 mol dm^{-3} solution of HCl or NaOH. The pH measurements were carried out using a pH meter. The suspensions were shaken for 3 h at 298 K.

Post-treatment of the Mg-loaded sample

Desorption. The Mg-CLI sample (containing 2.5 mg Mg g^{-1}) was suspended in a 2 mol dm^{-3} solution of NaCl and then left for 24 h in a thermostatic water bath at 298 K for 24 h. After filtration the Mg content in the filtrate was analysed by atomic absorption spectroscopy. In order to check the possibility of a multiple reuse of Na-CLI, the following experiment was carried out in six adsorption/desorption cycles. The exhausted samples were first treated with 2 mol dm^{-3} solution of NaCl, the suspension was mixed for 2h at 298 K, and then filtered, washed with distilled water and dried at 378 K. The Na-CLI was then treated with a solution containing 90 mg Mg^{2+} dm^{-3} for 3h at 298 K. The concentration of Mg in solutions was checked by AAS.

Influence on growth of A. junni. The culture of the P-accumulating bacteria *A. junii* strain DSM 1532 was obtained from the Deutsche Sammlung von Microorganismen und Zellkulturen GmbH and maintained on nutrient agar (Biolife). The sample of Mg-CLI containing 2.5 mg Mg g^{-1} was crushed to obtain particles between 0.122 to 0.263 mm in size and then sterilized at 120ºC for 3 h.

Cells of *A. junii* were pregrown on nutrient agar plates for 24 h at 30±0.1ºC. The biomass was suspended in a sterile 0.3% NaCl solution and the suspension was allotted to bottles containing 50 cm^3 of sterile synthetic wastewater (composition in mg dm^{-3} of distilled water: Na-propionate 300; $(NH_4)_2SO_4$ 92; $CaCl_2{\cdot}2H_2O$ 37; KCl 19; Na_2HPO_4 92; trace metal solution 10 (solution contained in mg dm^{-3} of demineralized water: $FeSO_4$ 50; $MnSO_4$ 5; $ZnSO_4$ 0.5; $CuSO_4$ 0.5; $CoSO_4$ 0.05; $MoSO_4$ 0.05); pH=7.0±0.2). Into one bottle 0.5 g of Mg-CLI was added, while another bottle without zeolite served as a control. The flasks were sealed with a sterile gum cap with a central hole through which aeration with filtered air (1 dm^3 min^{-1}) was provided. The flasks were incubated at 30.0±0.5ºC in a water bath (Memmert WNB22).

All measurements were done in triplicate. Water samples for the measurements of P (PO_4^{3-}) and Mg were filtered through Whatman filter units of pore diameter 0.2 μm. The P concentration in the water was measured spectrophotometrically in a DR/2500 Hach spectrophotometer by the molybdovanadate method (Hach method 8114). The concentrations of Mg were determined by atomic absorption spectrometry (AAS) and the pH value was measured with the WTW 330 pH-meter. The number of viable bacterial cells was determined as colony-forming units (CFUs) grown on the nutrient agar after incubation at 30±0.1ºC for 24 h. Planktonic cells were determined by serial dilutions (10^{-1} to 10^{-8}) of 1 cm^3 of supernatant sample and inoculations of nutrient agar plates. In order to determine the number of immobilized cells, the Mg-CLI was taken from the flask, washed three times with 300 cm^3 of sterile 0.3 wt.% NaCl and aseptically placed into a tube containing 9 cm^3 of 0.3 wt.% NaCl.

The sample was crushed with a sterile glass rod and vigorously shaken on a mechanical shaker (40 Hz/3 min). This procedure (Durham *et al.*, 1994) detaches the immobilized cells from the carrier, so that they remain as individual cells in the suspension. From this suspension serial dilutions were made and nutrient agar plates were inoculated and incubated as already described. After incubation, the bacterial colonies were counted and reported as CFUs per one gram of dry carrier. The Neisser stain was performed to confirm polyP granules in cells of *A. junii*. A direct microscopy (Axiovert 200 MAT; Carl Zeiss MicroImaging, Inc.) was also performed to confirm the immobilization of cells onto the carriers.

Statistical Software 8.0 (StatSoft, Tulsa, USA) was used for statistical analysis. The numbers of bacterial CFU were logarithmically transformed beforehand to normalize distribution and to equalize variances of the measured parameters. The comparisons between the samples were done using the one-way analysis of variance (ANOVA), and subsequently the post-hoc Duncan test was performed for the calculations concerning pair-wise comparisons. Statistical decisions were made at a significance level of $p < 0.05$.

Analytical techniques

All chemicals used were analytical grade reagents. Mg solutions were prepared by dissolving $MgCl_2 \cdot 6H_2O$ (Aldrich) and $CaCl_2 \cdot 2H_2O$ (Aldrich). The metal concentrations in solutions were determined by AAS (Varian Spectra B55 atomic absorption spectrophotometer). At least five measurements were done for each determination. The relative standard deviation was found to be below 2%.

Chemical analysis of the clinoptilolite samples was performed using a scanning electron microscope JEOL JSM-6610LV. The samples were prepared by embedding grains in an epoxy film, polishing and cutting with a fine diamond-grid cut, and then coating the sample by gold. An average elemental composition of the samples was obtained by a data collection at ten different mm^2-sized windows on the sample surface. The accuracy of the method was ±3 %. The loss on ignition was determined by thermal analysis using a SDT Q- 600 simultaneous DSC-TGA instrument (TA Instruments). The sample (mass app. 10 mg) was heated in a standard alumina sample pan, the experiment being carried out under air with a flow rate of 0.1 $dm^3\ min^{-1}$.

The X-ray powder diffraction (XRPD) patterns were recorded at room temperature on a Siemens D-5000 diffractometer using Cu-$K\alpha$ radiation (1.5406 Å). The samples were thorougly homogenized in an agate mortar before the diffraction data collection in order to avoid preferential orientation of the crystallites. The XRPD data were collected in the range from 3 to 60° 2θ in steps of 0.02° 2θ with a scan step time 15 s. The qualitative powder analyses of the collected XRPD patterns were performed using the Crystallographica Search-Match programs (Crystallographica Search-Match, 2003) and quantitative powder X-ray analyses using the TOPAS V2.1 Rietveld refinement program (Topas, 2000).

RESULTS AND DISCUSSION

Zeolite characterization

According to X-ray powder diffraction analysis (Fig. 1), the zeolitic tuff contains ~90% clinoptilolite. Several minor constituents including mica, clay minerals, plagioclase feldspars, K-feldspar, quartz and cristobalite were also identified in the sample. It should be noted that the zeolitic tuff from Ntrista stream location of Petrota village in Greece has a very similar mineral composition (Filippidis, 2010), indicating that the tuffs originate from similar source rocks.

The careful preparation of samples by embedding zeolite grains in an epoxy film, polishing crystallites, cutting them with a fine grid diamond cut enabled an intersection view of the crystallites and allowed a detailed elemental analysis of major mineral phases by EDS. SEM images of a typical grain and a magnified detail on the grain are given in Fig. 2. The shape and colour contrast indicated the presence of different mineral phases which were examined by EDS in detail. The most widespread darker area showing in Fig. 2 belongs to the clinoptilolite phase.

The average elemental compositions of the clinoptilolite phase in the as-received (CLI), Na-modified (Na-CLI) and Ca, Mg-loaded samples (Mg-CLI) found by EDS are listed in Table 2. It can be seen that in the clinoptilolite phase K, Ca and Mg ions are dominant whereas the Na content is low. In a preliminary study (not shown) the as-received sample has been found to exhibit a very low ion-exchange ability not only for Mg^{2+} but also towards heavy metal ions. In order to improve the ion exchange capacity the zeolitic tuff was pretreated with NaCl solution. The treatment increased the content of Na (only in the clinoptilolite phase) leading simultaneously to a

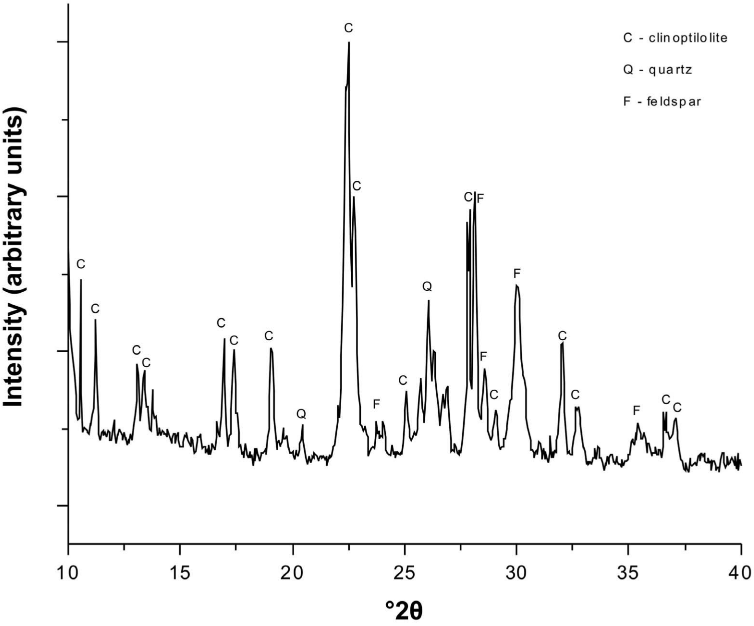

FIG. 1. XRPD pattern of Na-modified zeolitic sample (Na-CLI).

decrease in the amounts of other exchangeable cations. The Si/Al ratio (being 4.8) was rather constant, indicating a uniform distribution of the elements. The Na-CLI was further treated with a solution containing Mg^{2+} and Ca^{2+} ions in concentrations which are similar to those found in the spring water. The treatment led to a change of the composition only in the clinoptilolite phase: an increase in the Mg and Ca content occured with simultaneous decrease of Na whereas the K content remained unchanged. This confirmed that the ion-exchange process in the clinoptilolite lattice occurs mainly via an exchange of Na cations.

The water content in the zeolitic tuff was determined by thermal analysis. Figure 3 shows typical thermograms of Na-CLI and Mg-CLI. A loss of about 13.5 wt.% corresponds to dehydration. The water content increases slightly (14.0 wt.%) after

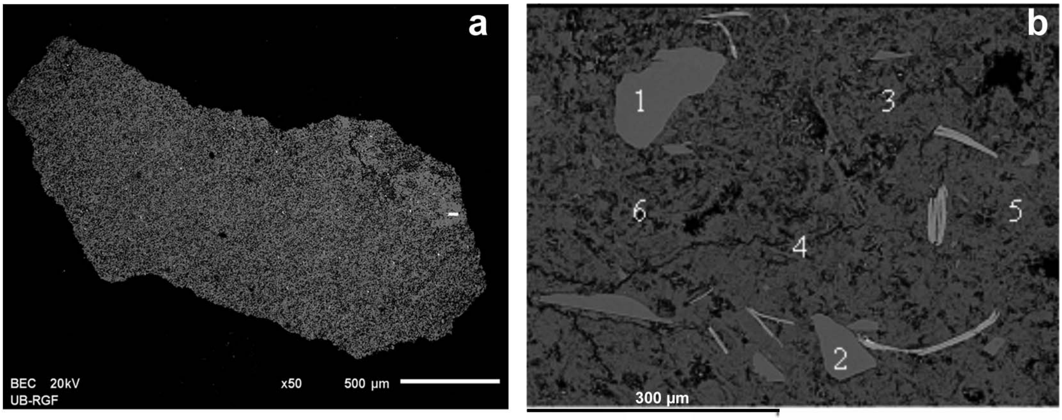

FIG. 2. (a) SEM photomicrographs of polished crystallites of a zeolite grain. (b) The colour contrast shows the presence of different mineral phases: clinoptilolite (dark areas), feldspars (bright) and quartz (very bright).

TABLE 2. Results of EDS analyses of the as-received (CLI), Na-modified (Na-CLI) and Ca,Mg-loaded Na-CLI (Mg-CLI) in wt.%. The chemical compositions correspond to average values obtained from ten area measurements (12 mm^2). Standard deviations are given in parentheses.

	CLI	Na-CLI	Mg-CLI
SiO_2	67.9 (3)	67.0 (3)	67.6 (2)
Al_2O_3	11.9 (1.3)	12.0 (1.5)	11.6 (1.6)
Na_2O	0.12 (0.2)	4.08 (0.1)	2.64 (0.1)
K_2O	1.03 (0.4)	0.83 (0.2)	0.71 (0.2)
MgO	1.32 (0.2)	1.04 (0.1)	1.87 (0.1)
CaO	4.38 (0.1)	1.54 (0.1)	1.87 (0.2)
H_2O*	13.4	13.5	13.7
**	$Ca_{2.0}K_{0.8}\ Mg_{0.8}Na_{0.05}$ $Al_{6.2}Si_{29.7}O_{72}\ 20H_2O$	$Ca_{0.7}K_{0.2}Mg_{0.7}Na_{1.7}$ $Al_{6.3}Si_{29.6}O_{72}\ 20H_2O$	$Ca_{0.8}K_{0.2}Mg_{1.2}Na_{1.1}$ $Al_{6.4}Si_{29.9}O_{72}\ 20H_2O$

* Estimated by difference

** Oxide formula on basis of 72 framework oxygens

the entry of Mg and Ca into Na- CLI. This could be attributed to a replacement of larger Na cations (102 pm) by smaller ones (Mg – 86 pm, Ca 100 pm).

The effect of pH

The influence of pH of the initial solution on the Mg removal is shown in Fig. 4. The Mg uptake does not change in the pH range 5 to 7. An increase of about 18 % in the Mg uptake is observed when the pH is increased from 7 to 8 whereas raising the pH further does not significantly influence the Mg sorption.

The maximum loading of Mg at a pH of about 8 could be explained by the dependence of the Zeta potential of zeolite on pH. According to Vochten (Vochten *et al*., 1990), the Zeta potential of zeolite becomes more negative with an increase of pH, because of hydroxylation of the zeolite surface.

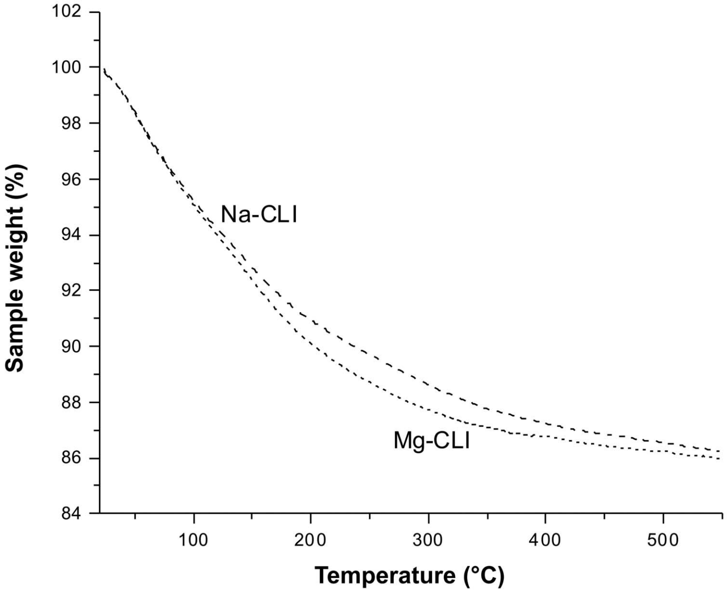

FIG. 3. TG curves of Na-CLI and the exhausted Na-CLI (Mg-CLI).

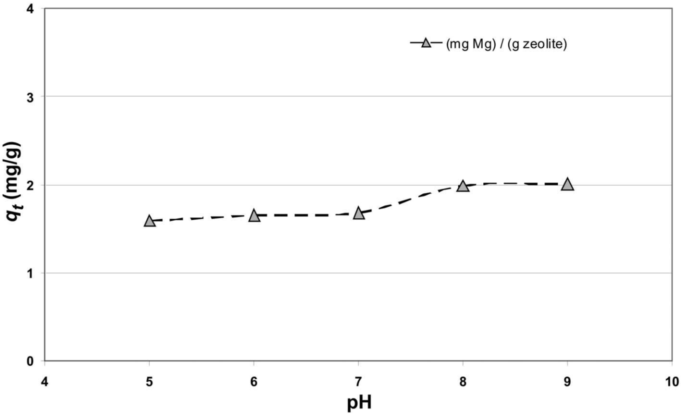

FIG. 4. The effect of pH on the Mg^{2+} uptake.

This results in an increase of the Mg loading. Since the pH of the spring water is about 8, an adjustment of the pH is not necessary.

Effect of initial concentrations on the Mg loading

Sorption curves are presented as the time functions for variable initial concentrations in Fig. 5.

The curves show that the Mg loading increases with an increase of initial concentrations. Moreover, the uptake of Mg increases rapidly in the first 100 min and, after that, rises gradually up to the equilibrium amount. The Mg content in Mg-loaded zeolite obtained from the initial concentration of 100 mg Mg dm^{-3} after 24 h is less then 10% greater than after 3h. This indicates that the ion exchange reaches equilibrium very slowly. It can also be seen that the sorption capacity of Na-CLI towards Mg^{2+} ions is rather low (about 2.5 mg g^{-1}). As a comparison, sorption capacity of the zeolite for Zn^{2+} is 5.8 mg g^{-1} (Stojaković *et al.*, 2011b) and 8.3 mg g^{-1} for Cu^{2+} (Stojaković *et al.*, 2011a). A possible reason could be the size of the hydrated ions (Huheey *et al.*, 1993). In an aqueous solution, Zn^{2+} is mainly tetrahedrally coordinated by water molecules (Douglas *et al.*, 1994) and its radius is 74 pm. Furthermore, octahedrally coordinated Cu^{2+} (87 pm) can rather easily adjust its coordination geometry to the smaller planar one (71 pm) due to the Jahn-Teller effect (Cotton *et al.*, 1995). In contrast, the small Mg^{2+} ion has a large hydration

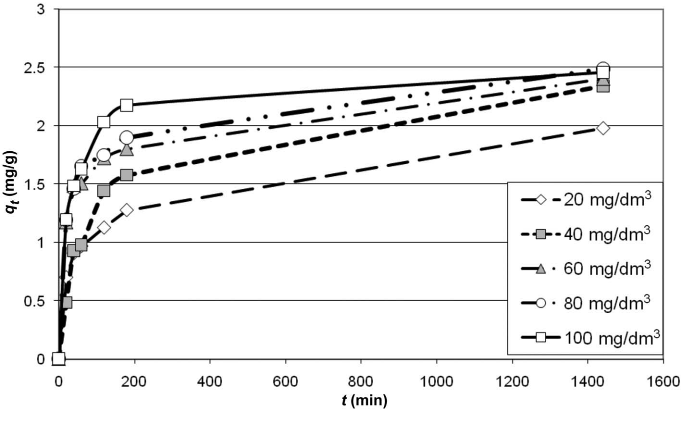

FIG. 5. Amounts of the removed Mg^{2+} by Na-CLI for different initial concentrations of the solution at 298 K.

volume and its hydration shell can be reduced much less easily (Arrigo *et al.*, 2007), which prevents the entrance of $Mg^{2+}(aq)$ into the clinoptilolite channels. Regarding the results obtained it can be presumed that at the beginning the ion exchange increases with the initial concentration since there are enough available sites for ion exchange at the surface of the clinoptilolite crystals. As the surface sites become occupied, a slow filling of the sites inside the channels begins until equilibrium is reached.

Kinetics study

Study of the Mg uptake with time was conducted at 298K for initial concentrations of 20, 40, 60, 80, and 100 mg dm^{-3} over the time intervals of 20, 40, 60, 120 and 180 min. Two basic kinetic models were applied to describe the obtained experimental data. First is the Lagergren model for the first order rate of the sorption process (Ho, 2004):

$$dq_t/dt = k_1\,(q_e - q_t) \tag{1}$$

where q_e (mg g^{-1}) stands for the capacity of the adsorption at equilibrium and k_1 (min^{-1}) is the first order rate constant for the adsorption process.

In order to distinguish between the kinetic equations based on the concentration of the solution from the adsorption capacity of solid bodies, the Largengren first-rate equation is frequently called a pseudo-first-order reaction (Ho, 2006). Integration of equation (1) over the limits $t = 0$ to $t = t$ and $q = 0$ to $q = q_e$, gives the expression:

$$\log\,(q_e \quad q_t) = \log q_e \quad k_1 t/2.303 \tag{2}$$

The graph of log $(q_e - q_t)$ *vs.* t should be a straight line if the experimental data are in agreement with the kinetic model.

The other model which has also been used in the present study can be described by an equation of the pseudo-second order as follows (Ho & McKay, 1998):

$$dqt/dt = k_2\,(q_e - q_t)^2 \tag{3}$$

where k_2 (g mg^{-1} min^{-1}) stands for the adsorption rate constant of the pseudo-second order. Integration within the limits stated for equation 2 gives the following expression:

$$t/q_t = (1/k_2 q_e^2) + (1/q_e)t \tag{4}$$

The plot for the relationship of t/q_t *vs.* t will be a straight line if the experimental data are in agreement with the corresponding kinetic model, and the values of q_e and k_2 could be evaluated from the slope and intersection of the plot. Calculated parameters are given in Table 3.

It is evident that the model for the pseudo-second-order process is in better agreement with the experimental data than the model for the pseudo-first-order one. This was to be expected since the pseudo-second-order model has been preferentially used in the studies of sorption of metal ions by natural zeolites (Ho, 2006; Rajić *et al.*, 2009, 2010).

Diffusion effects. In order to investigate the effect of diffusion on the sorption process the data were interpreted via the Weber-Morris model for mass transfer (Weber & Morris, 1963). This model describes the intraparticle diffusion using the relationship presented in equation 5:

$$q_t = k_d t^{1/2} + I \tag{5}$$

where q_t (mg g^{-1}) stands for adsorption capacity, t (min) – time, k_d (mg g^{-1} $min^{-1/2}$) is the rate of the intra-particle diffusion and I (mg g^{-1}) is the intercept. If the graph of q_t *vs.* $t^{1/2}$ is a straight

TABLE 3. Parameters calculated from the kinetic models for pseudo-first and pseudo-second-order reactions applied to the uptake of Mg^{2+} by Na-CLI at variable initial concentrations of the Mg solutions.

	Lagergen model of the pseudo-first order		Lagergen model of the pseudo-second order		
Mg (mg dm^{-3})	k_1 (min^{-1})	R^2	k_2 (g $mg^{-1}min^{-1}$)	q_e (mg g^{-1})	R^2
20	4.61×10^{-4}	0.764	8×10^{-3}	2.05	0.9951
40	6.91×10^{-4}	0.776	6×10^{-3}	2.44	0.9960
60	4.61×10^{-4}	0.620	1.3×10^{-2}	2.45	0.9984
80	6.91×10^{-4}	0.631	1.2×10^{-2}	2.54	0.9985
100	4.61×10^{-4}	0.474	1.8×10^{-2}	2.49	0.9996

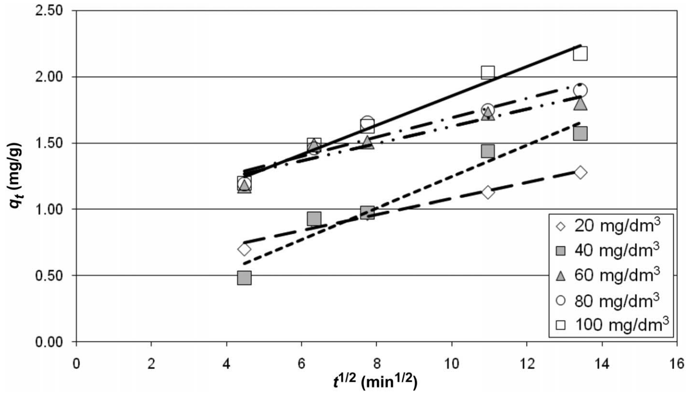

FIG. 6. Dependence of the sorption capacity q_t *vs.* $t^{1/2}$ for the initial concentrations of 20, 40, 60, 80 and 100 Mg dm^{-3} at 298 K.

line the intra-particle diffusion is incorporated in the sorption process. In addition, if the straight line passes through the coordinate intersection, then the intra-particle diffusion is the rate determining step (Poots *et al*., 1976).

Figure 6 shows that the plots are straight lines. They have been analysed by linear regression and it was found that the intercept *I* (equation 4) is greater than zero (not shown). This indicates that in the present study the intra-particle diffusion occurs but it is not the rate-limiting step.

Regeneration of the exhausted clinoptilolite

The possibility of regeneration of the exhausted zeolite and its reuse was investigated by multiple adsorption/desorption cycles. Amounts of the Mg^{2+} uptake per cycle are given in Fig. 7.

It is evident that the uptake of Mg by Na-CLI increases slightly after the third cycle (for about 10%), which could be attributed to an exchange of Ca^{2+} by Na^+ in repeated regenerations (confirmed by EDS analysis). Also, it can be noticed that a complete desorption of Mg from Mg-containing zeolite is possible. This indicates not only that the sorbent can be reused but also the sorbed Mg can be recovered.

Influence of the exhausted zeolite on the growth of A. junii

The influence of the addition of Mg-CLI to wastewater with a low content of Mg (0.17 mg Mg/mg P) is presented in Table 4. In a control reactor, significantly lower multiplication of *A. junii* was observed, resulting in a lower number of total

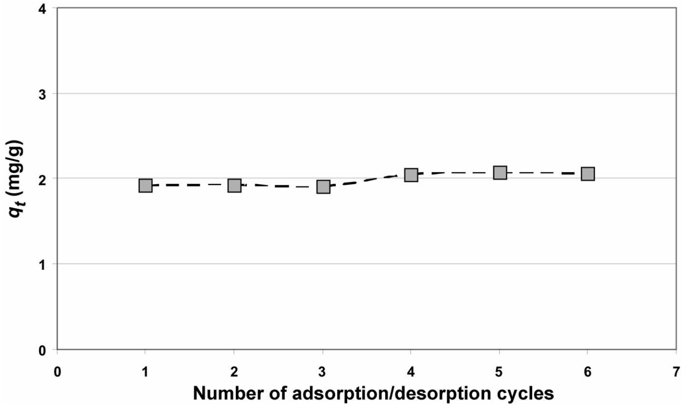

FIG. 7. Amounts of Mg^{2+} uptake (mg g^{-1}) after several adsorption/desorption cycles.

TABLE 4. Performance of reactors containing only *A. junii* (control - CR) and *A. junii* with addition of Mg-loaded clinoptilolite (Mg-CLI) after 24h of incubation. [c_0 CFU (10^9 L^{-1})] = 1.12±0.34; [c_0 P-PO_4 (mg L^{-1})] = 19.75±0.06.

Parameter	Control	Mg-CLI
Final pH	7.07±0.02	7.62±0.02^A
Immobilized cells (10^8 CFU g^{-1})	–	6.37±1.90
Planktonic cells (10^9 CFU L^{-1})	8.15±0.63	46.10±2.69^A
Total cells (10^9 CFU L^{-1})	8.15±0.63	46.13±2.68^A
CFU final/CFU start	6.74±1.94	48.48±6.82^A
P uptake rate (10^{-10} mg P CFU^{-1})	1.48±0.11	1.62±0.08
P removed (%)	6.08±0.02	37.72±0.49^A

^{A} – significantly different values when compared to control.

cells and a consequently lower percent of P removal from wastewater when compared to the reactor containing Mg-CLI. The bacterial population in the control reactor consisted of planktonic cells in supernatant and immobilized cells onto Mg-CLI. The number of bacteria immobilized on Mg-CLI was slightly higher than the number of *A. junii* immobilized on the original zeolitic tuff (6.14×10^8 CFU/g) and lower than on Mg-rich zeolitic tuff originating from Turkey (95.22×10^8 CFU/g) (Hrenović *et al.*, 2009). Previous investigation has shown that the immobilization of *A. junii* onto the zeolitic tuff is a relatively slow process: for a 3 h contact only 2% of the total bacterial population was immobilized onto the Mg rich zeolitic tuff (Hrenović *et al.*, 2009). As a comparison, the immobilization of cyanobacteria onto Greek zeolitic tuff proceeds significantly faster: for a 1 h contact up to 92% of the total bacterial population was immobilized (Filippidis 2010; Filippidis *et al.*, 2010). The immobilization of the bacteria has been attributed mainly to surface precipitation due to the presence of Broensted acidic sites and the Lewis basic sites of the HEU-type zeolite (Filippidis 2010; Filippidis *et al.*, 2010).

Improved efficiency of P removal in the reactor containing Mg-CLI was the result of bacterial metabolism, since it has been found that the zeolite itself adsorbed only 1.06% of P from wastewater. The percentage of P removal showed a significantly positive correlation with the number of total cells of *A. junii* (R = 0.999), while the correlation with the P uptake rates was not significant (R = 0.772), suggesting that the P removal from wastewater was the function of increased bacterial biomass. This observation is in agreement with a previous one (Hrenović *et al.*, 2005), where the increased concentration of Mg^{2+} originating from natural zeolitic tuff increased the yield of biomass of P-accumulating bacteria in a pure culture, which resulted in improved P removal from wastewater.

At critically low concentration of Mg (<0.64 mg Mg/mg P; Hrenović *et al.*, 2010) in the wastewater, the Mg-CLI successfully replaced the lack of this micronutrient, enhanced the growth of *A. junii* and consequently increased the P removal from wastewater.

CONCLUSIONS

The zeolitic tuff from the locality of Igroš near Brus (Serbia) exhibits a relatively low affinity towards Mg^{2+} ions (max. 2.5 mg Mg per 1 g zeolite). The ion exchange process follows pseudo-second-order kinetics and the capacity is highest at a pH of about 8 which corresponds to that of the spring water which was the subject of the present investigation.

In spite of the low capacity of the zeolite it could be used as a softener in smaller drinking water plants. A flow rate of 0.5 dm^3 s^{-1} is sufficient for drinking water supplying up to 200250 inhabitants, taking into account that 150–200 dm^3 per inhabitant is the daily consumption. Therefore, an installation consisting of two filters of 1 t each should be sufficient to improve the quality of the spring water for supplying a village or a group of households. This could significantly decrease the cost of currently used water softeners based on synthetic polymers as well as their impact on the environment. The zeolitic tuff from Igroš is a potentially useful material, especially considering its low cost. The exhausted zeolite can be used as a support for the immobilization of P-accumulating bacteria involved in the process of the enhanced biological phosphorus removal from wastewaters. At the same time the exhausted zeolite can serve as a source of Mg for P-accumulating bacteria during the treatment of wastewater which has an unfavourable Mg/P ratio.

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