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CONDITIONING OF DRINKING WATER ON CONSTRUCTED WETLAND: ELIMINATION OF *Escherichia coli*

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Abstract

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A constructed wetland is, in brief, a water treatment facility. Duplicating the processes occurring in natural wetlands, constructed wetlands are more complex, integrated systems in which water, plants, animals, microorganisms and the environment interact to improve water quality. In local water reservoirs, especially in the countryside, excessive pollution occurs frequently, in particular microbiologic pollution, which is most often a result of poorly safeguarded water protection areas. Agricultural activities can be the cause of increased content of nitrates, pesticide residue and microbiologic pollution. The existing technological solutions (e.g. membrane and absorption filters) are too expensive and difficult to manage in case of small water supply systems. Commonly used chlorination or ozonization do not remove harmful nitrates. Also, nitrates cannot be removed by adsorption agents (activated carbon, quartz sinter, diatomaceous earth, etc.), but they can be removed by reverse osmosis or chemically (ionic exchangers, etc.). Constructed wetlands (CW) are expected to provide a certain solution to all above-mentioned problems. If a suitable mechanical system of particles removal (inorganic and organic particles) is used, there is no need for additional filtering.

Key words: drinking water, constructed wetland, *Escherichia coli*

Introduction

The majority of European countries are facing problems with polluted drinking water (Nixon, 2004). Contaminated drinking water can be a consequence of agriculture, inappropriate deposition of waste, traffic, absent or leaking sewage system, etc. The most frequent contaminants of drinking water are coliform bacteria, nitrates, pesticides and their residues and heavy metals, all of which affect human health (Aslan, 2005; Aslan, Turkman, 2005; Pintar et al., 2001; Cheng et al., 2002; Pintar, 2003; Bruggen et al., 2001). In spite of reduced use of pesticides in agriculture their concentrations in drinking water will remain high for some time as majority of pesticides need long time to break down.

According to drinking water pollution in Slovenia the most problematic are small local water supply systems particularly in agricultural areas. In Slovenia there are 811 small water supply systems which supply altogether 184.207 inhabitants. Small water supply systems are defined as systems which supply between 50 and 1000 people. Water wells which supply less than 50 people are excluded. The most frequent pollutants are microbes from faecal origin, with which drinking water becomes polluted when in contact with human or animal faeces or domestic waste water. The most affected are small supply systems which supply between 50 and 500 inhabitants. In these systems according to national monitoring of drinking water every second sample is microbiologically polluted (Monitoring of drinking water, 2005).

Conventional methods which enable effective elimination of all present pollutants are often too expensive form small water supply systems. In conventional methods for drinking water purification, bigger particles are removed through sand filters, small particles, microbes and pesticide residues through membrane and adsorption filters, and nitrates through reverse osmosis or chemically with ion exchange colons (Pintar et al., 2001; Lecloux, 1999; Vaaramaa, Lehto, 2003; Karakulski et al., 2002; Bentama et al., 2004; Bruggen et al., 2001). The amount of microorganisms in water can be reduced also with chlorination and ozonization. Unpleasant side consequence of chlorination and ozonization is the formation of unwished chemical side products of disinfection (Lecloux, 1999).

For elimination of various pollutants from drinking water there are also some other - alternative methods which mostly base on more natural accessions. For example Hanson et al. (2004) suggests removal of organic, inorganic and bacterial pollution with solar distillation. There are also experiments with biological methods: most frequently different bacterial cultures were studied in terms of nitrate removal (Pintar, 2003; Aslan, 2005; Aslan, Turkman, 2005; Lecloux, 1999). Biological denitrification has a great potential in drinking water purification but its transfer to technology is slow due to possible contamination of cleaned water with bacteria and organic matter remains (Pintar, 2003).

Purification of drinking water is possible also with the use of community of organisms which is composed from microorganisms, algae, invertebrates and higher plants. Wotton (2002) describes sand filters where community of plants, animals and microorganisms is developed and enables purification of drinking water which is slowly moving through the filter. Wotton (2002) compares reactions in filter with reactions in natural sandy habitats in water environment (sandy beach or river bank). Water purification is happening as in water column as in the sand. Wotton (2002) gives the advantage to open filters which enable the break through of light. Light enables the growth of algae and thus nutrient consumption. Bacteria and invertebrates decompose organic matter in the water and also excrete exopolimeres, which accelerate flocculation and aggregation of particles in the water. After some time on the surface of the sand a mixed layer of sand, organisms and detritus is developed where particular organic matter is captured and colloid and dissolved solids are adsorbed. Mentioned sand filters also enable removal of pathogenic bacteria and viruses (Wotton, 2002).

Very efficient biological systems for water purification are wetlands. Natural or artificial wetlands are also used for purification of water for groundwater recharge (Reilly et al., 2000). To achieve standards for drinking water removal of nitrate is crucial. Reilly et al. (2000) mention that efficiency of nitrate removal in studied system was between 14 and 100%, in average 80% which was enough to reach the criteria for groundwater recharge. The highest contribution to nitrate removal had denitrifying bacteria. Also vegetation and carbon accessibility affected the process. Reilly et al. (2000) also state the deficit of carbon for denitrification is more likely to happen in young wetlands where vegetation is not fully developed and there is little or no detritus than in older wetlands. Short retention times in wetlands (0.3 to 9.6 days) enabled smaller water loses due to evapotranspiration and thus more water for groundwater recharge.

The most wide spread artificial wetlands are constructed wetlands for domestic water treatment (in further text CW). They are used all over the world. Their advantage is simple technology and the possibility of total nitrogen removal with simultaneous course of nitrification and denitrification (Jing et al., 2001; Kuschk et al., 2003; Steinmann et al., 2003; Vacca et al., 2005; Cheng et al., 2002, etc.). Processes of water purification in constructed wetland are affected by environmental factors like temperature, air humidity, solar radiation, etc. Especially in moderate climates there appears season dynamics.

CW enable also the reduction of bacterial and viral pathogens, metals (Cheng et al., 2002) and pesticides (Alvord, Kadlec, 1996). Vega et al. (2003) report about successful reduction of viral concentration from municipal wastewater, and Vacca et al. (2005) mention the reduction of coliform bacteria for two size classes in constructed wetlands as well as in sand filters. The efficiency of bacterial reduction depends on the presence of plants, filtering media and construction conditions (Vacca et al., 2005). Adsorption, desorption and inactivation also play an important role in reduction of microbial population. They depend on specific characteristics of CW, substrates and local clime (Vega et al., 2003). Studies show that bactera in wastewater can bind directly on the surface of plants' roots (Vymazal, 2007).

According to described problematic of drinking water pollution and proved efficiency of constructed wetlands for municipal wastewater treatment pilot CW for drinking water conditioning were set up. Until know constructed wetland have not been researched in detail for drinking water conditioning. Possible achievements of this research could be transferred in technological form suitable for marketing.

Aim of presented research is to find out to what extend we can eliminate different pollutants from drinking water using constructed wetlands. First part of the research includes the reduction of microorganisms. Results which would indicate effective treatment of drinking water on CW would in long term enable use of additional water resources. This is of great importance for areas where drinking water is in shortage.

We expected greater reduction of microorganisms from drinking water in vegetation season of plants in CW. On the other hand smaller but still present reduction was expected for wintertime. Lower temperatures in wintertime should not present bigger problem because flows through CW are high and retention times short. In this conditions water cannot freeze.

Methods

The experiment was preformed on pilot constructed wetland which was situated in a small settlement in NE part of Slovenia. Pilot plant was composed of two reed beds and four polishing basins (Figs 1, 2). In both reed beds the substrate was silicate sand. The dimensions of the first bed were 3x3x0.5 m. This bed was designed for rough filtration so the granulation of sand bigger than in second bed which was designed for the main part of drinking water purification. The dimensions of second bed were 7x3x0.5 m. Polishing basins were 0.5x0.5x 0.5 m in size and were designed for additional substrates for drinking water purification. In experiment described here only first bed was filled namely with peat. Due to acidity of peat and filtration elimination of bacteria was expected. Peat has high sorption capacity. Sorption is performed with formation of organic complexes, chelates with heavy metals, kation exchange and formation of hydrogen bonds (Zupančič, 2001), so the peat will also be important in further experiments where elimination of other pollutants will be studied. Additional substrates in polishing basins are meant to be natural and low-cost because sustainable and advantageous solution for drinking water purification for small water supply systems is wanted to be achieved.

Polluted drinking water flowed into the constructed wetland through inflow valve where flow through CW was regulated. Than water was flowing subsurface along the CW and polishing basins. Before the start of the experiment theoretical water retention time was calculated. Actual retention time was defined with the addition of soil in the inflow water and measuring of conductivity on the outflow of both beds and polishing basins of CW. Theoretical retention time was calculated with formula below (Bulc, 1998):

$$\mathbf{t = - Ah/Q} \qquad \mathbf{(1)}$$

t – theoretical retention time (d)
ε – porosity of substrata (m^3/m^3)
A – area of CW (m)
h – water depth (m)
Q – flow (m3/day)

During the experiment culture of *Escherichia coli* was added in the inflow water. Sampling was performed on the inflow, after first reed bed, after second reed bed and on after the polishing basins. In terms of adding of *E. coli* we wanted to reach the values described in report of national monitoring of drinking water in Slovenia in year 2005. In report it is stated that in some small water supply systems there was more than 300 bacteria of *E. coli* per 100 mL (the normative is 0 bacteria of *E. coli*/100 mL).

E. coli is used as indicator organism for coliform bacteria. Coliform bacteria live in digestive organs of mammals and birds. Outside the digestive tract their reproduction is poor. Elimination of indicator organism is important for investigations about elimination of other pathogenic microorganisms. The biggest part of elimination of *E. coli* from drinking water in subsurface flow CW is carried out by sedimentation, predation and natural die-off (Green et al., 1997).

Besides number of *E. coli* water analyses included also other parameters: smell, colour, temperature, pH, turbidity, conductivity, concentrations of nitrites and nitrates but the results are not shown here. The experiment was carried out in summer and wintertime to evaluate the contribution of plant activity and temperatures to efficiency of *E. coli* elimination from drinking water. Analyses were performed according to ISO standards.

The experiment was divided in 2 parts: in the first part the elimination of higher concentrations of *E. coli* was monitored at different flows. In the second part of the experiment we focused on selected flow at which in the first part of the experiment the efficiency was still high. In that flow we tested the elimination of lower concentrations of bacteria.

During spring and summer vertical and horizontal growth of reed was also monitored: density and high of plants in CW was measured. Measurements were carried out once per week on three coincidently chosen squares (Bulc, 1994; Šajn-Slak, 2003).

Results

Calculated theoretical retention times at different flows are given in Table 1. At the flows 5 and 10 L/min also actual retention times were defined. On the basis of these results

T a b l e 1. Theoretical water retention times on the pilot constructed wetland at different flows.

Flow	Theoretical water retention time
1 L/min	66 h
2 L/min	33 h
5 L/min	13 h 12 min
10 L/min	6 h 36 min
18.8 L/min	3 h 40 min

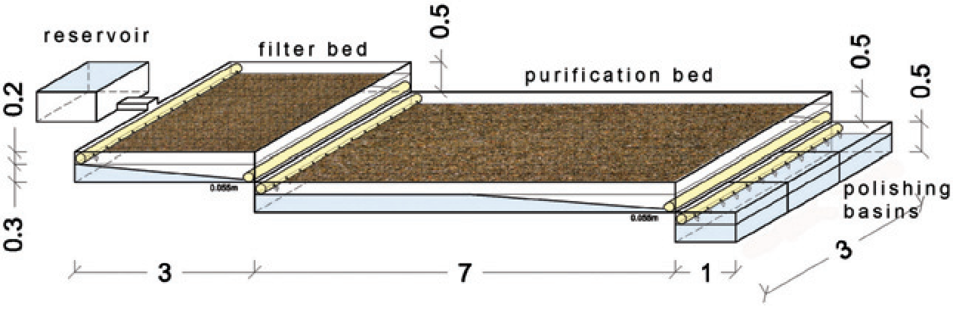

Fig. 1. Scheme of pilot constructed wetland for conditioning of drinking water.

Fig. 2. Constructed wetland for conditioning of drinking water in settlement in NE part of Slovenia.

retention times for other flows were estimated (Table 2). Maximal flow which was possible in our experimental system was 18.8 L/min.

T a b l e 2. Actual and estimated water retention times on pilot constructed wetland at different flows.

Flow	1st reed bed	2nd reed bed	1st polishing basin	Total
1 L/min	10 h	46 h 40 min	cca 50 min	58 h 20 min
2 L/min	5 h	23 h 20 min	cca 50 min	29 h 10 min
5 L/min	2 h	9 h 20 min	cca 10 min	11 h 30 min
10 L/min	1 h	4 h 40 min	cca 10 min	5 h 50 min
18.8 L/min	32 min	2 h 29 min	cca 10 min	3 h 11 min

In preliminary tests higher concentration of *E. coli* was added in the inflow water (approximately 40.000 bacteria/100 mL). The efficiency of bacteria elimination was monitored at different flows according to retention time. Results are shown in Fig. 3.

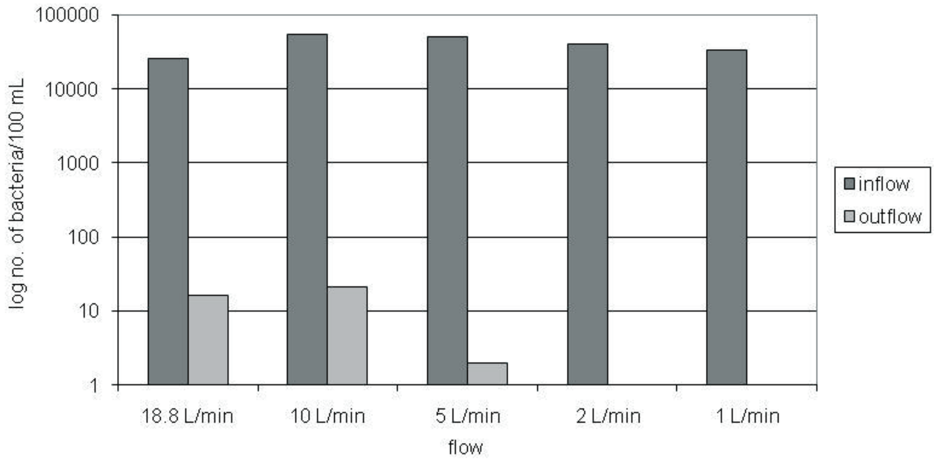

Fig. 3. Changing of number of bacteria (*E .coli*) in the inflow and outflow of constructed wetland at different flows.

Logarithmic reduction of number of bacteria at flows 18.8, 10 in 5 L/min is shown in Table 3. It can be seen from the table that the elimination of *E. coli* was higher at lower flows.

At flows 2 and 1 L/min the elimination of *E. coli* from drinking water on CW was complete. At higher flows bacteria on the outflow have been detected. Higher efficiency of bacteria elimination at lower flows was expected. Retention time at flow 2 L/min was 29 hours and 10 minutes and at flow 1 L/min 58 hours and 20 minutes. Due to still high efficiency in elimination of *E. coli* at flow 5 L/min in further research we focused on men-

T a b l e 3. Logarithmic reduction of bacteria *E. coli* at different flows.

Flow (L/min)	Log removal
18.8	3.21
10	3.41
5	4.41

tioned flow and tested the elimination of lower inflow concentrations of *E. coli*. Results are shown in figure 4. Inflow concentrations were between 47 and 160 bacteria/100 mL in first series and in second between 460 and 500 bacteria/100 mL. Ability of elimination of these concentrations of bacteria is important because these concentrations appear in unsuitable water samples in national water supply systems.

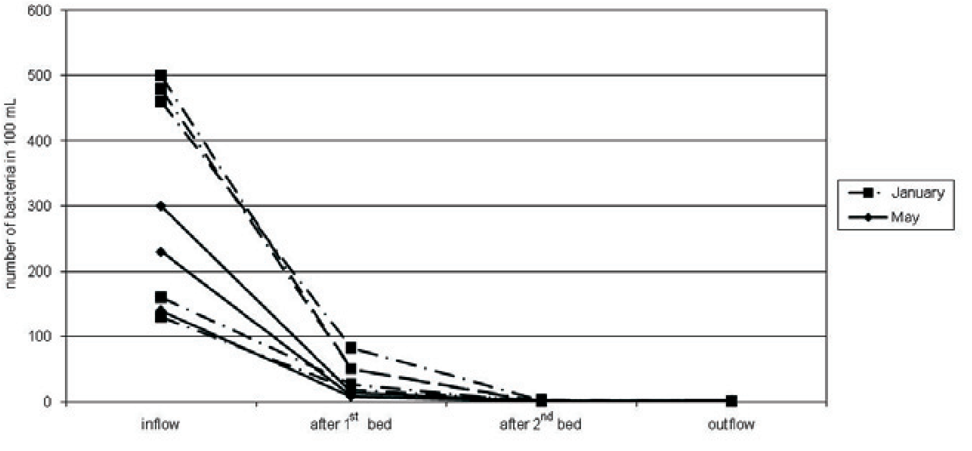

Fig. 4. Changes in bacterial (*E. coli*) concentrations through CW.

Bacterial reduction at flow 5 L/min was effective in lower and also higher inflow concentrations of *E. coli*. Highest attribute to bacteria removal has the first bed of CW. Effect of peat in polishing basin is hard to estimate as water which flew into polishing basin contained just 0 to 4 bactera/100 mL. When flowing through peat concentrations of *E. coli* were between 0 and 2 bactera/100 mL. In Table 4 logarithmic reduction which happened in first bed are shown in winter as in spring time.

Results of measurements of reed's growth are shown in Fig. 5. Reed began to sprout in the beginning of April. Growth was intensive till the end of May: the height of plants was increasing from 8 to 19 cm per week. In June growth slowed down and reached ap-

T a b l e 4. Logarithmic reduction of bacterial number in first bed of CW at flow 5 L/min.

Experiment	January					May		
	1	2	3	4	5	6	7	8
Log reduction after 1st bed	0.84	0.77	0.74	0.98	1.00	1.41	1.24	1.30

proximately 4 cm per week, but in July again 10 cm per week. In July the average height of plants was 135 cm.

Figure 6 shows the increase of plant density on CW. Density increase does not coincide with increase of plant height. Density was increasing till the beginning of May but bigger increase was not detected till mid June. After increase in mid June we detected a reduction in plant density.

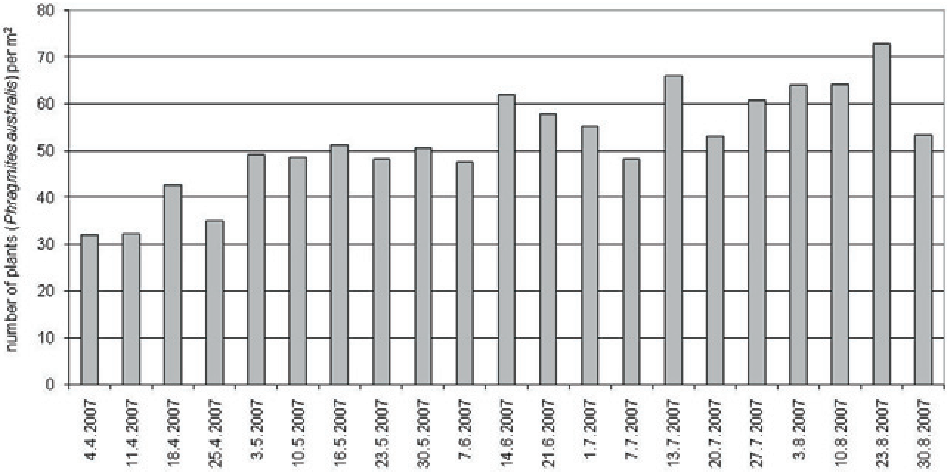

Fig. 5. Density of plants (*Phragmites australis*) on pilot CW.

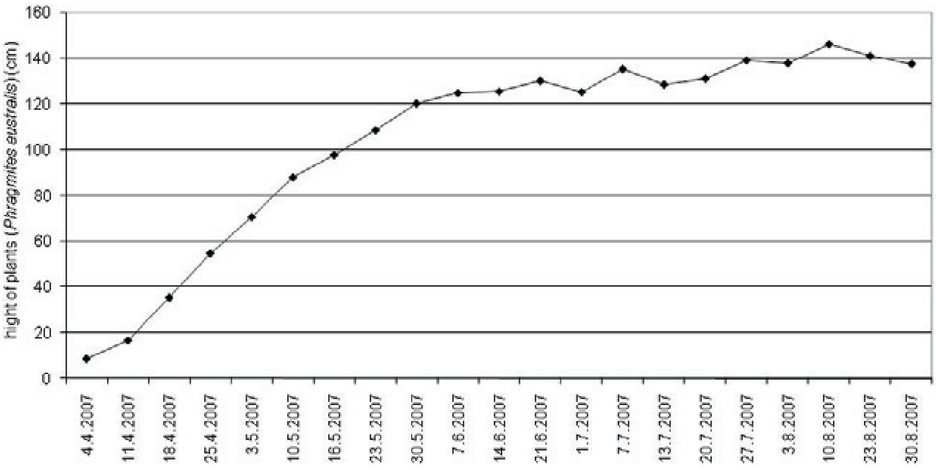

Fig. 6. Height of plants (*Phragmites australis*) on pilot CW.

Discussion and conclusions

For a pilot CW theoretical retention time was calculated (Equation 1). But at flows 5 and 10 L/min retention time was experimentally defined; separately for first and second basin. On the basin of results of measuring actual retention time also retention times for other flows at which experiment was happening were defined. The difference between theoretical and actually measured retention time is smaller at higher flows and gets higher at lower flows. Therefore at flow 18.8 L/min the difference between theoretical and actually measured retention time is 29 min. and at flow 1 L/min 7 h and 40 min. Theoretical calculation of retention time is different from actually measured retention time because the calculation in based on presumption that whole water mass in CW system is equally exchanged. Values of actual retention time are therefore usually much lower to calculated ones. Thus is CW there exist some areas where water mass is not exchanged regularly (Bulc, 1998).

The difference between theoretical and actually measured retention time can be caused by non-homogenous media which causes faster water flow through parts with rougher granulate. Retention time on the pilot plant was monitored soon after the construction and planting of pilot plant. With the growth of plants measures will need to be repeated because root growth influences on hydraulic conductivity of CW (Brix, 1997).

Elimination of *E. coli* from drinking water was 100% at lower flows (2 in 1 L/min), in higher flows the efficiency was reduced. Higher efficiency in *E. coli* elimination at longer retention times is reported also by Green et al. (1997). Inflow concentrations of *E.coli* in the first part of experiment are comparable to concentrations described by Green et al. (1997). Authors report reduction of *E. coli* for 1.51 log at retention time 6 hours, but in our experiment 3.4 log reduction has been detected at very similar retention time (5 hours and 50 minutes, flow 10 L/min). Higher elimination in our experiment could be a consequence of smaller granulate and mineral composition of substratum or higher natural die-off of bacteria.

In further experiment we focused on flow 5 L/min where efficiency of bacteria elimination was still high. In drinking water purification it is important that the higher quantities of water are purified in the shortest time as we have to fulfil the needs of consumers. Shorter retention times also mean decrease of evapotranspiration of water from CW. Inflow concentrations were in the first series between 47 and 160 bacteria/100 mL and in second series between 460 and 500 bacteria/100 mL. Oscillations in inflow concentrations were caused by troubles with preparation of exact concentrations of inoculum and by the way inoculum was introduced into the system.

As shown in picture 4 bigger part of reduction is carried out in first bed of CW. Log reductions of *E. coli* at lower inflow concentrations are much lower in comparison with log reductions at higher inflow concentrations of *E. coli*: at flow 5 L/min log reduction in first part of experiment was 4.41, but in second part 1.00 in summer and 1.41 in winter time. Reduction of lower inflow concentrations is thus less intensive as reductions of higher. Although the outflow water contained form 0 to 3 bacteria/100 mL.

As shown in table 3 reductions of bacteria were higher in May and lower in January. Higher reductions in May could be caused by higher temperatures which enable faster course of chemical reactions. Schmoll (2006: 71-76) states that temperature is one of the most important factors affecting inactivation of bacteria in the environment. Laboratory studies, which he describes, show negative correlation between water temperature and survival of coliform bacteria. Also in our experiment reduction was higher in warmer springtime.

Higher elimination of *E. coli* in springtime could be caused also by activity of plants which were in that time in the period of intensive growth. Plant is CW have different physical impacts (they influence on faster sedimentation of particles, reduce flow speed etc.), plants have influence on substratum conductivity, they uptake nutrients, excrete different substances through roots (especially oxygen and also antibiotics and organic matter) and represent the area for growth of microorganisms and protozoa (Brix, 1997). Populations of microorganisms and protozoa can importantly affect inactivation of *E. coli* due to predation. Literature reports different statements according to influence of primary populations of microorganisms on the survival of entric bacteria but the majority of investigations show increase of inactivation level (Schmoll, 2006: 71–76).

In our experiment peat was not shown as effective substratum for elimination of microbial pollution, although in literature reports that peat could be used in this purposes (Zupančič, 2001).

Common reed in pilot plant started to grow in the beginning of April. Till the end of May the height of plants was increasing for 8 to 19 cm per week. In June growth was slowed down and reached approximately 4 cm per week, but in July it increased again to 10 cm per week. Till July plant reached average height 135 cm. Time of the start of common reed growth depends on climate conditions. Also Šajn Slak (2003) reports that common reed started to grow on CW for municipal waste water treatment in NE part of Slovenia in the beginning of April. We expect that plants will still grow through the summer and rich maximum in September or October as reported by Šajn Slak, 2003. In comparison with the growth of plants on CW for municipal wastewater treatment common reed on pilot CW for drinking water conditioning also grew well: in July similar values of plants height were reached as in her investigation reports Bulc (1994).

From the comparison of pictures 5 and 6 it is seen that density of plants increased the most when vertical growth was already stagnating. On the basis of results reported by Šajn Slak (2003) and Bulc (1994) we expect that he number of plants will still increase – approximately till the end of August or September.

So far results of elimination of *E. coli* from drinking water in CW are promising. High elimination efficiencies were reached as in winter as in springtime. Common reed was shown to be suitable plant also for CW for drinking water purification. Results shown are a good basis for further research.

CWs can contribute to improvement of drinking water quality but they cannot assure complete purification. Their use is rational mostly in terms of additional purification or pre-treatment and as preventive protection against different kinds of pollution.

Translated by D. Istenič

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