Microbiological quality of drinking water in public and municipal drinking water supply systems in Osijek-Baranja County, Croatia

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original scientific paper

Summary

Microbiological and chemical quality of drinking water primarily results from water origin and type of applied water treatment. Since the drinking water is one of the mean way through which many infectious agents can be transmitted to humans causing waterborne diseases, constant monitoring of drinking water quality in water supply systems is needed. This study investigates the microbiological quality of drinking water in twenty five public and municipal water supply systems in the area of Osijek-Baranja County in eastern Croatia. The microbiological analyses were conducted for following parameters: total coliforms, *Escherichia coli*, colony count at 22 °C and 37 °C, enterococci and *Clostridium perfringens*. In most of investigated water supply systems processed groundwater are used, and since increased concentrations of some chemical compounds in water can influence on appearance and growth of microbiological populations, in this study relevant physicochemical parameters were also measured and correlated with obtained values of analyzed microbiological parameters. Five physicochemical indicators were determined: temperature, turbidity, pH value, free residual chlorine and ammonium concentration. Results indicated that 149 of 1503 analyzed drinking water samples were non-compliance with microbiological criteria set by Croatian regulations (88.7 % and 89.2 % of 149 had values of colony count at 22 °C and 37 °C higher than the required). Total coliforms, enterococci and *Escherichia coli* were founded in 3.9 %, 1.1 % and 1 % of non-compliance drinking water samples, respectively. *Clostridium perfringens* was not detected in any of the drinking water samples. Calculating Pearson's coefficients of correlation among analyzed microbiological and physicochemical indicators, very weak correlations were obtained. The highest but negative correlation was observed between appearance of microbiological population and concentrations of free residual chlorine (lower free residual chlorine – higher appearance of microbiological population) and positive correlation between turbidity and appearance of microbiological population (higher turbidity – higher appearance of microbiological population).

Keywords: drinking water, microbiological quality, water supply system, Osijek-Baranja County

Introduction

Drinking water can be produced from surface or groundwater representing one of several means by which different infectious agents or chemical contaminants can be transmitted causing waterborne diseases and certain health problems (Hrudey and Hrudey, 2007; Reynolds et al., 2007; Poma et al., 2012). During the history, different approaches have been used to regulate contaminant appearance in drinking water, especially infectious agents, since drinking water can be directly or indirectly contaminated by human or animal excreta (Baker and Taras, 1981; US EPA, 2000; WHO, 2003). If the contamination is recent, and those contributing to the contamination include carriers of communicable enteric diseases, some of the microorganisms that cause these diseases may be present in the water. Drinking such contaminated water or using it in food preparation may cause new cases of infection. Rather than conducting a formal risk assessment for microbiological contamination of drinking water, regulators worldwide established a Maximum Contaminant Level of zero for pathogens (*Cryptosporidium*, *Giardia lamblia*, *Legionella*, *Escherichia coli*, *Clostridium perfringens,* enteric viruses etc.). Since analytical techniques for pathogens require a high analytical skill, time consuming and often significant expenses, the indicator organisms such as total coliforms are used to show the possible presence of microbiological contamination of the water by human waste (AWWA, 1999; Besner et al., 2011). Indicators are, generally, physical, chemical or microbiological parameters which presence at a level outside of specified limits may involve problems with quality of raw water, indicate the problem in the treatment process or in the integrity of the water supply system (WHO, 2003; Nygård et al., 2007; Levantesi et al., 2012; Douterelo et al., 2013; Malm et al., 2013)

There is a few important parameters influencing the occurrence of microbiological contamination and waterborne infections in a community: (i) the concentration of pathogenic organisms in the water,

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(ii) the virulence of the strain, (iii) the amount of intake of contaminated water, (iv) the infectious dose of the specific pathogen, (v) the susceptibility of individuals, (vi) the incidence of the infection in the community i.e., the number of pathogens being excreted and forwarded to other host (NHMRC and NRMMC, 2011). To prevent or to reduce the appearance of waterborne diseases, many countries provide monitoring programs based on international and/or national regulation standards. In Croatia, drinking water subject to the Regulations of parameters compliance and analysis methods of water for human consumption (MZ HR, NN 125/2013) which has a foothold in Croatian Law on water for human consumption (MZ HR, NN 56/2013) and European Community Directive guidelines (98/83/EC).

This study investigates during a year-long monitoring campaign the microbiological quality of drinking water in twenty five public and municipal water supply systems in the area of Osijek-Baranja County. The microbiological analyses were conducted for following parameters: total coliforms, *Escherichia coli*, colony count at 22 °C and 37 °C, enterococci and *Clostridium perfringens,* since those are the microbiological parameters that must be subject to check monitoring of drinking water taken from water supply system. Total coliforms are group of closely related bacteria that have been used for many decades as one of the main indicator of microbiological correctness of drinking water. The total coliforms group includes *Citrobacter*, *Enterobacter*, *Klebsiella*, *Escherichia coli* and some species of *Serratia*. They originating from digestive system of animals and they can be founded in different segments of environment including natural waters and waste waters. *Escherichia coli* is the major subset of the fecal coliforms group and specific indicator of recent fecal contamination (AWWA, 1999). Colony counts are enumerations of the general population of heterotrophic bacteria present in water. The enumerations may represent bacteria whose natural habitat is the water environment or those that have originated from soil or vegetation. Historically, these bacteria have been enumerated on bacteriological nutrient-rich media with incubation at 37 °C and 22 °C. It is well recognized, however, that only a small fraction of the viable bacterial population present in water is enumerated by the procedures normally employed. Despite this, monitoring of water supplies for colony count bacteria can be useful for monitoring trends in water quality or detecting sudden changes in quality regarding the treatments that have been used (EA UK, 2002). Practice shows that values of colony count also can be good indicator of water disinfections efficiency. Intestinal enterococci include a number of species that occur in the feces of humans and warm-blooded animals. The main reason for their enumeration is to assess the significance of the presence of coliforms bacteria in the absence of *Escherichia coli*, or to provide additional information when assessing the extent of possible fecal contamination. Enterococci of fecal origin rarely multiply in water and they are generally persisting longer in the environment (EA UK, 2002). *Clostridium perfringers* is a bacterium that is consistently associated with human and animal fecal wastes. Most species of *Clostridium* are environmental bacteria. Many are saprophytic, normally inhabiting soil, water and decomposing plant and animal material. These bacteria will be, therefore, present in surface derived source waters. *Clostridia* can be efficiently removed from water by coagulation and filtration process, but the spores of these bacteria can be resistant to chlorine at concentrations normally used in water treatment. So, lower number of *Clostridia* may occasionally occur in water supplies, but they do not represent a risk to health, since it will not grow to significant numbers, or produce toxins, in water supplies due to unsuitable conditions (AWWA, 1999; EA UK, 2002).

Since some studies emphasized that appearance and growth of microbiological populations in drinking water can be associated with elevated values of some physicochemical parameters (Volk and LeChevallier, 1999; Liguori et al., 2010; Poma et al., 2012), values of analyzed microbiological parameters were correlated with values of five physicochemical indicators (temperature, turbidity, pH value, free residual chlorine and ammonium concentration).

Materials and methods

Sampling area

Osijek-Baranja County (OBC) is located in the eastern Croatia and it is surrounded by Hungary on the North, Serbia on the East and with four other Croatian Counties on South-West (Fig. 1). This County covers an area of 4155 km^2 and has a population of approximately three hundred thousand inhabitants. Almost half of this population lives in town of Osijek and its area, while the other half living in 262 smaller places. The eighty four percent of OBC population obtain drinking water from public water supply systems while six percent of them obtain drinking water from smaller municipal water supply systems.

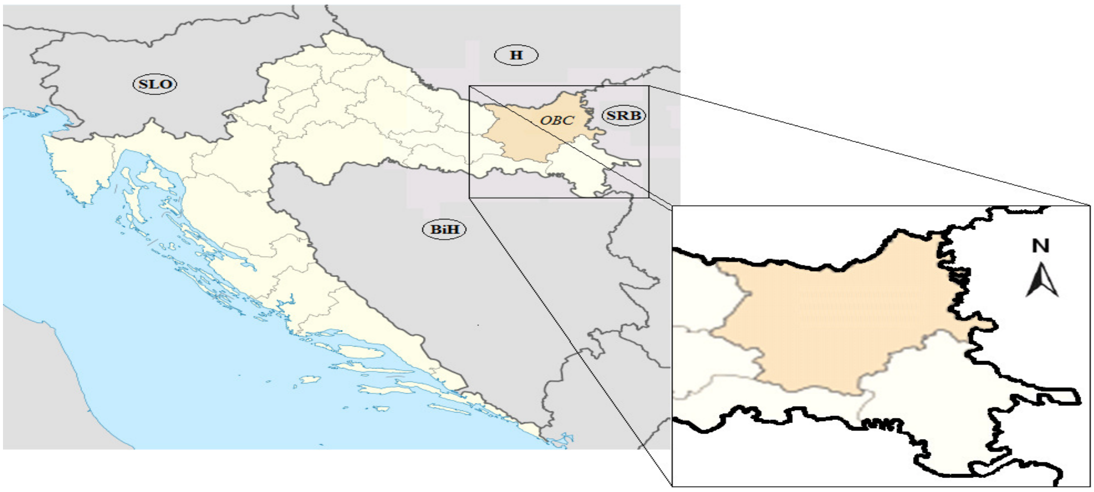

Fig. 1. Map of study area showing the location of Osijek-Baranja County

Sampling, analysis and regulations

This study investigates the microbiological quality of drinking water during a year-long monitoring campaign in the year 2012 which has been distributed through twenty five public and municipal water supply systems in Osijek-Baranja County.
All samples were taken into polyethylene bottles and maintained at 4 °C until analysis. Polyethylene bottles used for the samples collection were rinsed with deionised water before sampling.

The microbiological analysis of collected drinking water samples were conducted as follows:
A. total coliforms and *Escherichia coli* were determined due to HRN EN ISO 9308-1:2000/ 1:2008 method. Croatian Regulations (MZHR, NN 125/2013) does not allowed appearance of coliforms and *Escherichia coli* in drinking water.
B. colony count at 22 °C and 37 °C were determined due to HRN EN ISO 6222:2000 method. Croatian Regulations (MZHR, NN 125/2013) set highest value for colony count at 22 °C as 100/1 ml, and for colony count at 37 °C as 20/1 ml.
C. enterococci were determined due to HRN EN ISO 7899-2:2000 method. Croatian Regulations (MZHR, NN 125/2013) does not allow appearance of enterococci in 250 ml drinking water sample.
D. *Clostridium perfringens* was determined in drinking water samples according to European Council Directive (98/83/EC). Due to Croatian Regulations (MZHR, NN 125/2013) appearance of enterococci in 100 ml of drinking water sample is not permitted.

The physicochemical indicators of collected samples were analyzed as follows:
1) temperature was determined at the site using digital thermometer (Prima Log) due to standard method SM 2550A:1998. Croatian Regulation (MZHR, NN 125/2013) does not set maximum value for temperature of drinking water.
2) turbidity was determined in laboratory using turbidimeter TURB 350 IR (WTW) by HRN EN ISO 7027:2001. According to Croatian Regulations (MZHR, NN 125/2013) maximum permitted value of turbidity for drinking water is 4 NTU.
3) pH values were determined in laboratory electrometrically using Seven Easy S-20K pH meter (Mettler, Toledo) due to HRN EN ISO 10523:2012. According to Croatian Regulations (MZHR, NN 125/2013) pH value of drinking water have to be between 6.5-9.5.
4) free residual chlorine (FRC) was determined at the site using the *N,N*-diethyl-*p*-phenylenediamine (DPD) colorimetric method (colorimetric DPD method; Microquant, Merck) due to HRN EN ISO 7393-2:2001. Croatian Regulations (MZHR, NN 125/2013) set maximum concentration for free residual chlorine in drinking water samples taken from water supply systems at 0.5 mg/l.
5) ammonium concentrations were determined in laboratory using ICS-3000 Ion Chromatography system (Dionex) and method HRN EN ISO 14911:2001. Croatian Regulations (MZHR, NN 125/2013) set maximum concentration for ammonium in drinking water at 0.5 mg/l.

Statistical analysis

The analyzed microbiological parameters of drinking water samples taken from water supply systems in Osijek-Baranja County were statically evaluated using Microsoft Office Excel 2007 (Microsoft Corp., USA). Pearson's product-moment correlation coefficient was used to determine whether statistically significant correlation exist between appearance and growth of analyzed microbiological indicators and measured physicochemical parameters.
Pearson's correlation coefficient is a number between -1 and 1 which measures the degree to which two variables are linearly related. If there is a perfect linear relationship with a positive slope between the two variables, Pearson's correlation coefficient becomes 1. If there is a positive correlation between the two variables, whenever one variable has a high (low) value so does the other. Should there be a perfect linear relationship with a negative slope between two variables, Pearson's correlation coefficient becomes -1. This implies that there is a negative correlation between the two variables. Whenever one variable has a high (low) value the other also has a low (high) value. Pearson's correlation coefficient of value 0 implies that there is no linear relationship between the two variables. The Pearson's correlation coefficient value (r) is determined by Eq. (1).

$$r = \frac{\sum_{i=1}^{n}(X_i - \overline{X})(Y_i - \overline{Y})}{\sqrt{\sum_{i=1}^{n}(X_i - \overline{X})^2 \sum_{i=1}^{n}(Y_i - \overline{Y})^2}} \tag{1}$$

where

$X = \{X_1, X_2, ..., X_n\}$ is related with average values of physicochemical and $Y = \{Y_1, Y_2, ..., Y_n\}$ is related with average values of microbiological parameter for each water supply system and n is the number of water supply system where X or/and Y result was obtained.

Results and discussion

Compliances of drinking water samples from Osijek-Baranja County

A total of 1503 drinking water samples taken from twenty five public and municipal water supply systems in the Osijek-Baranja County were analyzed for six microbiological parameters and results are presented in Fig. 2. As it can be observed, 9.9 % of all analyzed samples were declared as non-compliance (149 of total 1503) since the values of tested microbiological parameters exceeded maximum values set by Croatian Regulations of parameters compliance and analysis methods of water for human consumption (MZ HR, NN 125/2013). Among 149 non-compliance drinking water samples, 88.7 % and 89.2 % exceeded required values regarding the colony count at 22 °C and 37 °C. As it was already emphasized, exceeding values of colony count parameters often indicated the problem of water in distribution system (EA UK, 2002). In our study it was observed that elevated values of colony count at 22 °C and 37 °C usually were detected in large distribution systems (largest has almost 600 km long pipelines) where ends with low water consumptions or with water that stands idle in pipes for long periods are usual. At such circumstances, our analytical result often shows, and calculated values of correlation coefficients present in Table 1 confirmed, that there is no more free residual chlorine present in drinking water sample.
In 3.9 % of 149 non-compliance drinking water samples, presence of total coliforms were detected. Total coliforms can appear in water supply systems due to sudden pipe breaks, develop of biofilm and insufficient or/and batch disinfectant dosage in water. We assumes that for the same reasons in 1.1% and 1% of non-compliance water samples enterococci and *Escherichia coli* were detected since appearance of analyzed microorganisms were never detected at the point where treated water entering into water supply system. *Clostridium perfringens* was not detected in any of analyzed drinking water samples.

Table 1. Correlation coefficients of analyzed microbiological and physicochemical parameters of drinking water samples taken from water supply systems in Osijek-Baranja County

	Total coliforms	E. coli	Colony Count 37 °C/48h	Colony Count 22 °C/72h	enterococci	average
temperature	0,02	0,004	0,019	0,142	-0,060	-0,004
turbidity	0,521	-0,026	-0,197	-0,062	0,031	0,053
FRC	-0,133	-0,057	-0,037	-0,123	-0,035	-0,077
pH	-0,211	-0,127	0,015	0,245	-0,081	-0,032
ammonium	-0,067	-0,122	0,047	0,120	-0,068	-0,018

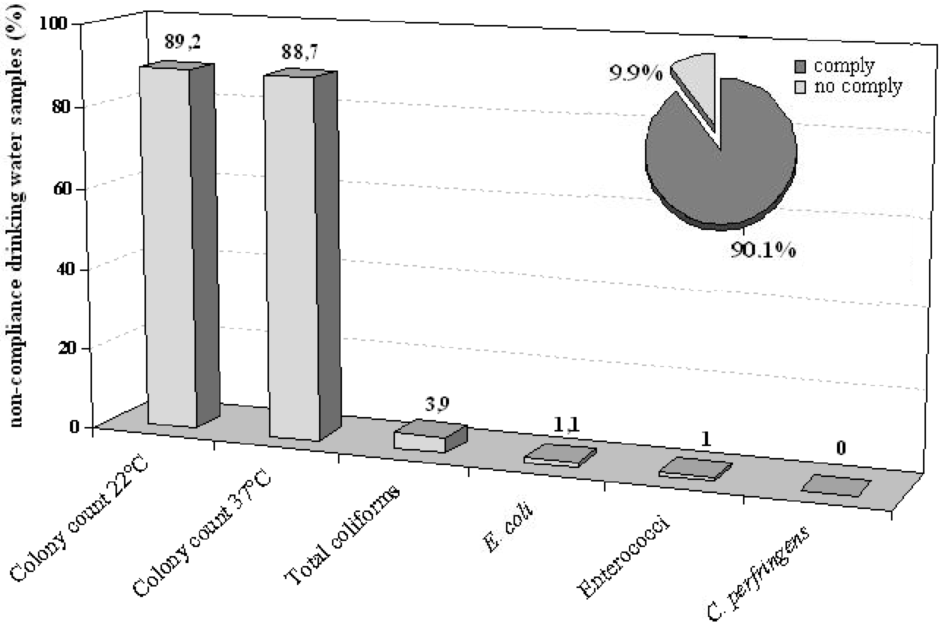

Fig. 2. Percentages of microbiological parameters in total number of microbiological non-compliance drinking water samples taken from water supply systems in Osijek-Baranja County

Correlations between microbiological and physicochemical parameters

In this study the relationship were observed between average values of microbiological parameters (total coliforms, *Escherichia coli*, colony count at 22 °C and 37 °C, enterococci and *Clostridium perfringens*) and average values of five physicochemical indicators (temperature, turbidity, pH value, free residual chlorine and ammonium concentrate) and obtained results are shown on Fig.3-7. The statistical correlation between average values above mentioned parameters were calculated using formula of the Pearson's correlation coefficient and obtained values are shown in Table 1. It must be noted that *Clostridium perfringens* was not detected in any collected drinking water sample, and therefore this microbiological parameter did not correlated with values of measured physicochemical indicators.

Fig. 3a-c presents the relationships between temperature and occurrence of total coliforms, *Escherichia coli*, enterococci and colony counts at 22 °C and 37 °C in drinking water samples taken from water supply systems in Osijek-Baranja County. Average temperature of all drinking water samples was 14.3 °C. The highest measured water temperature was 25.1 °C and this value was recorder during the summer period at water sample take at indoor facility, while lowest measured temperature was 2.9 °C and it was measured in water sample taken at check point placed outdoor during the winter. Our results indicate that increase of temperature had the strongest impact on appearance and growth of heterotrophic bacteria since in most water samples with higher temperature than average value, elevated values of colony count at 22 °C and 37 °C were obtained, and as it can be observed from Table 1, the highest value of correlation coefficient was obtained between temperature and colony count 37 °C (r = 0,142). All other calculated values of correlation coefficients between temperature and examined microbiological parameters have smaller values suggesting that correlation does not exist or is very weak.

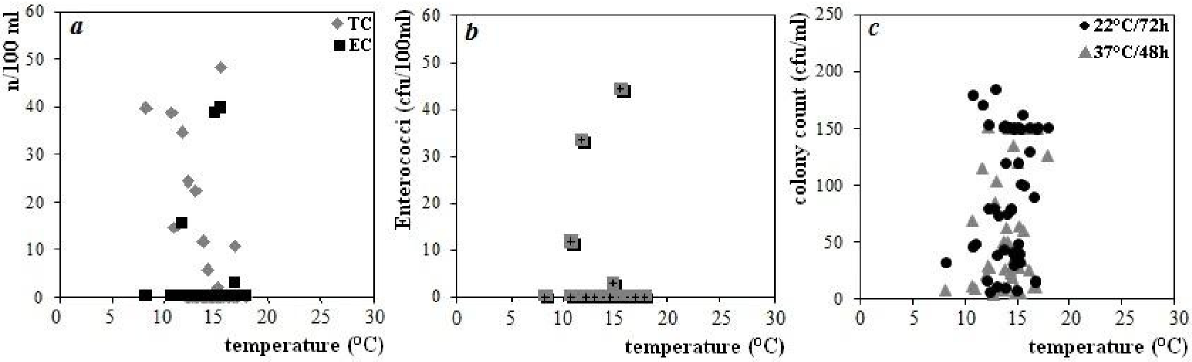

Fig. 3. Relationships between temperature and occurrence of (a) total coliforms (TC) and *Escherichia coli* (EC) and (b) enterococci and (c) colony counts at 22 °C and 37 °C in drinking water samples taken from water supply systems in Osijek-Baranja County

Fig. 4a-c presents the relationships between turbidity and occurrence of total coliforms, *Escherichia coli*, enterococci and colony counts at 22 °C and 37 °C. According to correlation coefficients presented at Table 1, heightened values of turbidity strongly correlated with higher values of total coliforms. It has to be emphasized that usually precipitated iron scale can cause turbidity and during this research, in over 99 % water samples with exceeded the permitted turbidity level, elevated values of total iron were determined although that analyses did not in detail described here. It was suggested that appearance of coliforms, as well as enterococci, in tested water samples is usually caused by sudden problems in integrity of water supply system and formation of biofilms in pipelines (Sartory and Holmes, 1997; Wingender and Flemming, 2011). Biofilm represent a complex organomineral deposits with a diversified microbial community and its formations in water supply systems is significant problem. Formation of biofilm begins with the attachment of free-floating microorganisms in drinking water to a surface of pipes. Results of studies shown that a high concentration of iron, caused by iron pipes corrosion, could also significantly contributed to biofilm formation (Ma et al., 2013), while the disinfection with chlorine appears to be a relatively ineffective procedure against microbes present in biofilms (Mathieu et al., 2009; Gosselin et al., 2013).

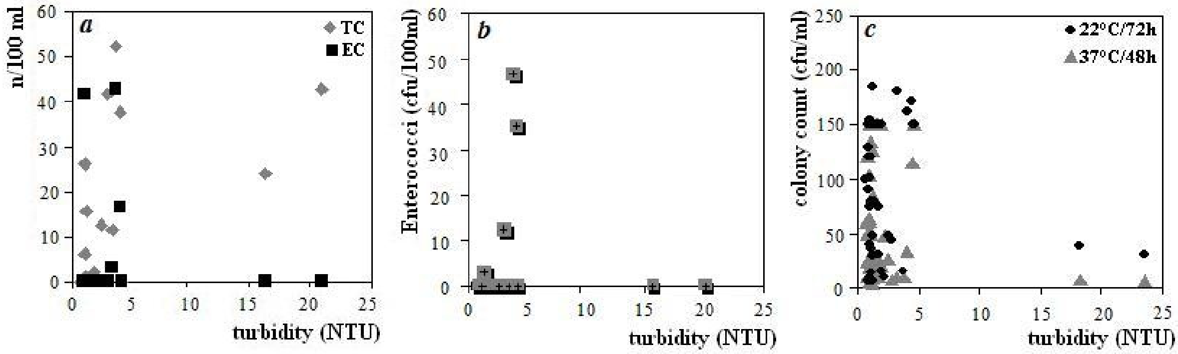

Fig. 4. Relationships between turbidity and occurrence of (a) total coliforms (TC) and *Escherichia coli* (EC) and (b) enterococci and (c) colony counts at 22 °C and 37 °C in drinking water samples taken from water supply systems in Osijek-Baranja County

Fig. 5a-c presents the relationships between free residual chlorine and occurrence of total coliforms, *Escherichia coli*, enterococci and colony counts at 22 °C and 37 °C. Chlorine and its compounds are one of the most widely used disinfectants in drinking water treatments for the last century, and when it is applied, the free residual chlorine is checked in water as the amount of chlorine available to disinfect the water in water supply system. The presence of free residual chlorine in drinking water indicates that a sufficient amount of chlorine was added initially to the water before distribution and it also contribute to water protection from recontamination during storage and distribution (Sartory and Holmes, 1997; AWWA, 1999; Kumpel and Nelson, 2013). The results of our study shows that average value FRC was 0.08 mg/l. The higher determined values were the same as regulation limit and in a many samples absence of FRC were noted. As it was already mentioned, a smaller water supply system in OBC usually has batch disinfection system which usually does not provide enough dosage of disinfectant in water. From data presented at Table 1 it can be observed that free residual chlorine has negative correlation coefficients with all tested microbiological parameter, viz higher concentration of FRC implied absence or sporadic occurrence of bacteria.

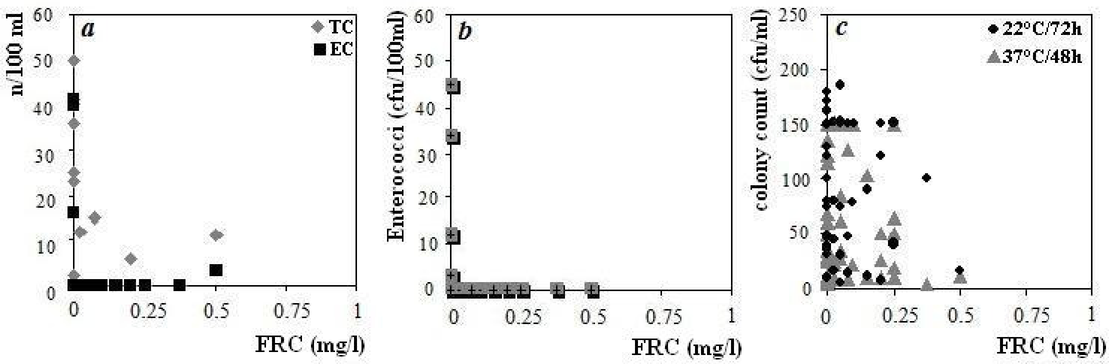

Fig. 5. Relationships between free residual chlorine (FRC) and occurrence of (a) total coliforms (TC) and *Escherichia coli* (EC) and (b) enterococci and (c) colony counts at 22 °C and 37 °C in drinking water samples taken from water supply systems in Osijek-Baranja County

Fig. 6a-c presents the relationships between pH and occurrence of total coliforms, *Escherichia coli,* enterococci and colony counts at 22 °C and 37 °C in drinking water samples taken fom water supply systems in OBC. Although it is not clearly evident at Fig. 6, values of calculated correlation coefficients at Table 1 show that decreasing of pH increased appearance and growth of fecal coliforms. Our results are consistent with results presented by other studies (AWWA, 1999; Barrios et al., 2001; Belenguer et al., 2007) which were investigated impact of pH on human fecal microbial communities and concluded that they rapidly growth at slightly acidic environment. The opposite was observed in the case of heterotrophic bacteria growth since the correlation coefficients among colony count and pH has positive value.

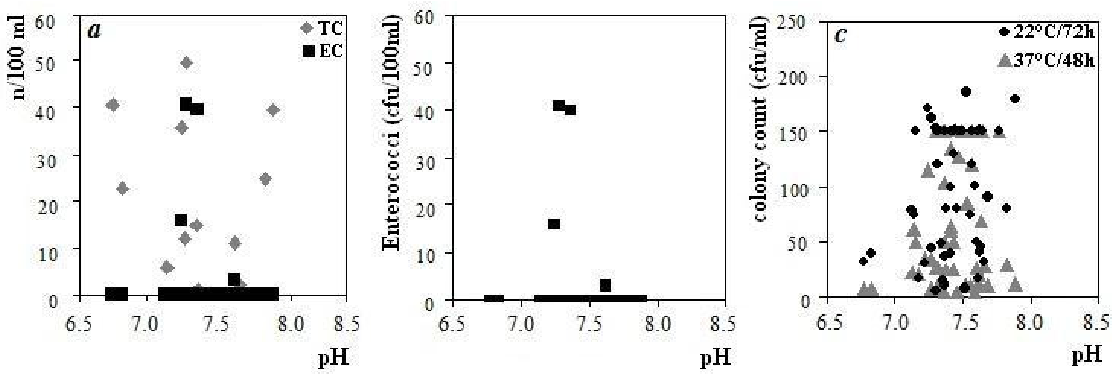

Fig. 6. Relationships between pH and occurrence of (a) total coliforms (TC) and *Escherichia coli* (EC) and (b) enterococci and (c) colony counts at 22 °C and 37 °C in drinking water samples taken from water supply systems in Osijek-Baranja County

Ammonium concentration in drinking water depends of its initial concentration in raw water and applied drinking water treatment. The sudden appearance of ammonium in drinking water samples can be caused by microbiological decay of animal and/or plant proteins and then presence of ammonium in water usually indicates pollution (WHO, 1996; AWWA, 1999). Fig. 7a-c presents the relationships between ammonium concentrations and occurrence of total coliforms, *Escherichia coli* and colony counts at 22 °C and 37 °C in drinking water samples taken from water supply systems in OBC. Due to calculated correlation coefficients presented at Table 1, higher ammonium concentrations in drinking water positively, but very weak, correlated with appearance and growth of heterotrophic bacteria, while negative, but also very weak correlations were observed between ammonium concentration and appearance and growth of total coliforms, enterococci and *Escherichia coli*, although Wilhelm and Malik (1998) investigating frequency of fecal-bacteria occurrence in surface waters, reported the positive correlation between ammonium concentrations and appearance of fecal-indicator bacteria in drinking water samples.

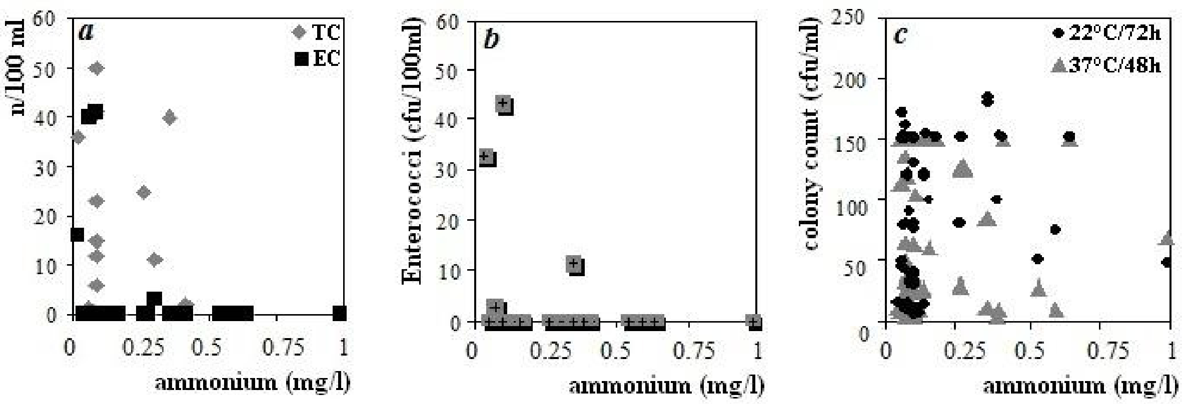

Fig. 7. Relationships between ammonium concentrations and occurrence of (a) total coliforms (TC) and *Escherichia coli* (EC) and (b) enterococci and (c) total colony counts at 22 °C and 37 °C in drinking water samples taken from water supply networks in Osijek-Baranja County

Conclusions

This study investigates the microbiological quality of drinking water during 2012 in twenty five public and municipal water supply systems in Osijek-Baranja County. The following parameters were analyzed: total coliforms, *Escherichia coli*, colony count at 22 °C and 37 °C, enterococci and *Clostridium perfringens,* and obtained values were correlated with five physicochemical parameters (temperature, turbidity, pH value, free residual chlorine and ammonium concentration). The following conclusions have been drawn from this study:

- 149 of 1503 analyzed drinking water samples were non-compliance with microbiological criteria set by Croatian regulation,
- 88.7 % and 89.2 % of 149 non-compliance samples has elevated number of heterotrophic bacteria expressed as number of colony count at 22 °C and 37 °C,
- total coliforms, enterococci and *Escherichia coli* were founded in 59, 15 and 17 of 149 non-compliance drinking water samples, while *Clostridium perfringens* was not detected in any of the drinking water samples,
- the highest but negative correlations were observed among appearance of microbiological population and concentration of free residual chlorine, while strongest positive correlations were determined between turbidity and appearance of microbiological population.

References

AWWA, American Water Works Association (1999): Water Quality and Treatment, New York, USA: McGraw-Hill, Inc., pp 1.24 -2.17.

Baker, M.N., Taras M.J. (1981): The quest for pure water – The history of the twentieth century, Volume I and II, AWWA, Denver, USA.

Barrios, J.A., Jiménez, B., Salgado, G., Garibay, A., Castrejon, A. (2001): Growth of faecal coliforms and Salmonella spp. in physicochemical sludge treated with acetic acid, *Water Sci. Technol.* 44 (10), 85-90.

Belenguer, A., Duncan, S.H., Holtrop, G., Anderson, S.E., Lobley, G.E., Flint, H.J. (2007): Impact of pH on Lactate Formation and Utilization by Human Fecal Microbial Communities, *Appl. Environ. Microbiol.* 73 (20), 6526-6533.

Besner, M.-C., Prévost, M., Regli, S. (2011): Assessing the public health risk of microbial intrusion events in distribution systems: conceptual model, available data, and challenges, *Water Res.* 45, 961-979.

Council Directive 98/83/EC of 2 November 1998 on the quality of water intended for human consumption. Official Journal of the European Communities 5.12.98.

Douterelo, I., Sharpe, R.L., Boxall, J.B. (2013): Influence of hydraulic regimes on bacterial community structure and composition in an experimental drinking water distribution system, *Water Res.* 47, 503-516.

EA UK, Environment Agency of United Kingdom (2002): The Microbiology of Drinking Water (2002) - Part 1 - Water Quality and Public Health. http://www.environment-agency.gov.uk/aboutus/default.aspx. Accessed: 1.10.2013.

Gosselin, F., Madeira, L.M., Juhna, T., Block, J.C. (2013): Drinking water and biofilm disinfection by Fenton-like reaction, *Water Res.* 47, 5631-5638.

Hrudey S.E., Hrudey E.J. (2007): Published case studies of waterborne disease outbreaks-evidence of a recurrent threat, *Water Environ. Res.* 79, 233-245.

Kumpel, E., Nelson, K.L. (2013): Comparing microbial water quality in an intermittent and continuous piped water supply, *Water Res.* 47, 5176-5188.

Law on water for human consumption, Zakon o vodi za ljudsku potrošnju (in Croatian), (NN 56/2013).

Levantesi C., Bonadonna, L., Briancesco, R., Grohmann, E., Toze, S., Tandoi, V. (2012): Salmonella in surface and drinking water: Occurrence and water-mediated transmission, *Food Res. Int.* 45, 587-602.

Liguori, G., Cavallotti, I., Arnese, A., Amiranda, C., Anastasi, D., Angelillo, I.F. (2010): Microbiological quality of drinking water from dispensers in Italy, *BMC Microbiol.* 10 (19) 1-5

MZ RH, Ministarstvo zdravlja Republike Hrvatske (Ministry of Health of the Republic of Croatia): Pravilnik o parametrima sukladnosti i metodama analize vode za ljudsku potrošnju, (Regulations of parameters compliance and analysis methods of water for human consumption), (NN 125/2013).

Ma, H., Darmawan, E.T., Zhang, M., Zhang, L., Bryers, J.D. (2013): Development of a poly(ether urethane)system for the controlled release of two novel anti-biofilm agents based on gallium or zinc and its efficacy to prevent bacterial biofilm formation, *J. Control. Realese* 172, 1035-1044.

Malm A., Axelsson G., Barregard, L., Ljungqvist, J., Forsberg, B., Bergstedt, O., Pettersson, T.J.R. (2013): The association of drinking water treatment and distribution network disturbances with Health Call Centre contacts for gastrointestinal illness symptoms, *Water Res.* 47, 4474-4484.

Mathieu, L., Bouteleux, C., Fass, S., Angel, E., Block, J.C. (2009): Reversible shift in the α-, β- and γ-proteobacteria populations of drinking water biofilms during discontinuous chlorination, *Water Res.* 43, 3375-3386.

NHMRC and NRMMC, National Health and Medical Research Council and Natural Resource Management Ministerial Council (2011): Australian Drinking Water Guidelines, Chapter 5 – Microbial quality of drinking water. http://www.nhmrc.gov.au/guidelines/publications/eh52. Accessed: 1.10.2013.

Nygård, K., Wahl, E., Krogh, T., Tveit, O.A., Bøhleng, E., Tverdal, A., Aavitsland, P. (2007): Breaks and maintenance work in the water distribution systems and gastrointestinal illness: a cohort study, *Int. J. Epidemiol.* 36, 873-880.

Poma, H.R., Gutiérrez Cacciabue, D, Garcé, B., Gonzo, E.E., Rajal, V.B. (2012): Towards a rational strategy for monitoring of microbiological quality of ambient waters, *Sci. Total Environ.* 433, 98-109.

Reynolds, K.A., Mena, K.D., Gerba, C.P. (2007): Risk of waterborne illness via drinking water in the United States, *Rev. Environ. Contam. Toxicol.* 192, 117-158.

Sartory, D.P., Holmes, P. (1997): Chlorine sensitivity of environmental, distribution system and biofilm coliforms, *Water Sci. Technol.* 35 (11-12), 289-292.

Volk, C.J., LeChevallier, M.W. (1999): Impacts of the Reduction of Nutrient Levels on Bacterial Water Quality in Distribution Systems, *Appl. Environ. Microbiol.* 65 (11), 4957-4966.

US EPA, United States Environmental Protection Agency (2000): The History of Drinking Water Treatment. www.epa.gov/safewater/consumer/pdf/hist.pdf. Accessed: 1.10.2013.

WHO, World Health Organization (1996): Ammonia in Drinking-water: Guidelines for drinking-water quality, 2nd ed. Vol. 2. Health criteria and other supporting information. World Health Organization, Geneva, Switzerland.

WHO, World Health Organization (2003): Assessing Microbial Safety of Drinking Water. Improving Approaches and Methods. IWA publishing, London, pp. 47-110.

Wilhelm, L.J., Malik, T.L. (1998): Fecal-indicator bacteria in surface waters of Santee River Basin and coastal drainages, North and South Carolina, 1995-98: U.S. Geological Survey Fact Sheet FS-085-98, 6 p.

Wingender, J., Flemming, H.C. (2011): Biofilms in drinking water and their role as reservoir for pathogens, *Int. J. Hyg. Environ. Health* 214, 417-423.

Received: November 18, 2013
Accepted: December 23, 2013