



MICROBIAL CONTAMINATION OF THE DRINKING WATER DISTRIBUTION SYSTEM IN SUCEAVA COUNTY, ROMANIA -THREE YEARS OF MONITORING (2010-2012)

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Abstract: The microbiological quality of drinking water has attracted great attention worldwide because of implied public health impacts. Monitoring and control technologies are indispensable for the production of safe drinking water. They allow for the surveillance of source water quality and the detection of biological and chemical threats, thus defining the boundary conditions for the subsequent treatment and providing early warning in case of unexpected contamination. The aim of this study was to monitor the water distribution stations in Suceava County and to better understand if the modernization projects were successfully implemented. The results showed that microbiological contamination incidence during three years monitoring was low and in some cases there were no relation between the sample-detected out of line for chlorine analysis and microbiological contamination incidence. This suggests, as a particular case, that underground water in Berchisesti area is microbiologically pure. However, a relatively low and constant incidence of microbiological contamination between 1-3% was recorded during 2010-2012, among the investigated samples. The samples founded out of line for chlorine treatment, meaning insufficient treatment, recorded an average of about 25% during the same period.

Key words: drinking water, microbiological quality, drinking water treatment

1. Introduction

There is no doubt that water quality plays a crucial role on human health. In developing countries, many common and widespread health risks have been found to be associated with drinking water, a large percentage of which are of biological origin [1]. The most important aspect of drinking water in relation to waterborne diseases is its microbiological quality.

Detection of bacterial indicators in drinking water implies the presence of pathogenic organisms. These are considered the source of waterborne diseases that could be fatal [2]. The enteropathogenic bacteria are responsible for a variety of diseases such as cholera, typhoid, dysentery and bacillary dysentery in humans [3]. They belong to the *Enterobacteriaceae* family of organisms that comprise members of the genera Escherichia, Enterobacter, Salmonella and Klebsiella, among others [4]. Commonly, their occurrence in water is due to faecal contamination. The faecal indicator bacterium (Escherichia coli), found in the faeces of all warmblooded animals and some reptiles has been considered as a bioindicator of faecal contamination of drinking water [5]. The major pathogenic responsible for waterborne bacteria diseases are spread by the faecal-oral route, in which water may play an intermediate role. Bacterial contaminants such as E. coli and fecal coliform in drinking water represent an acute health risk. There are many reports, which indicate waterborne disease outbreaks in water meeting the coliform regulations [6], [7], [8], [9]. The European Union Council directive states, according to World Health Organization guidelines, that drinking water should not contain pathogenic microorganism in such concentration able to depreciate human health. To evaluate the efficiency of drinking water treatment against microbial contamination, it is needed to estimate the adequacy of disinfection conditions. such as chlorination however, the chlorination conditions used in water distribution systems are based on the inactivation of several viruses and bacteria but not mycobacterial species or other pathogens. Chlorine disinfection is often combined with storage in a safe container, in order to prevent recontamination, and educational and motivational campains [10].

As to 2011 over 86.200 people live and work in Suceava County. Water is one of the main sources of human exposure to microbiological hazards. Pathogens are introduced into surface waters through different pathways such as industrial effluent, raw and treated sewage, storm water and animal manure runoff, in addition to industrial and household solid waste disposal [11]. Although legislation establishes regulatory standards in terms of fecal indicator bacteria to assess the microbiological quality of water, these do not necessarily predict the presence of pathogens such as parasites and viruses. Better surveillance and management strategies are needed to assess the risk of pathogens' waterborne transmission.

Underground waters usually have a high microbiological quality and can be used directly without any pretreatment prior to human consumption. On the other hand, the surface waters needs to be processed to become potable. This is a complex process and should be carefully monitored to avoid any undesirable incident. Since 2008 Suceava city is supplied with dinking water from two distinct sources Mihoveni station based on surface water and Berchisesti based on underground water. Berchisesti station is situated 26 km away from city and has a capacity of 785 l/s and provides 60% of necessary consuming water of Suceava city. Mihoveni station is situated only 5 km away from the city and has a capacity of 320 l/s. The difference between these two water stations consists in the fact that technological fluxes are different because of the water source is different. At Berchisesti where the water is from underground, there is necessary only a disinfection in a preventive way while at Mihoveni station the surface water need to be well processed including sedimentationfiltration-disinfection the process being much more complex. Environmental health protection requires safe dinking water means free of pathogenic bacteria, and among the pathogens disseminated in water sources, enteric pathogens are the frequently ones most mentioned. Therefore, sources of fecal pollution in waters devoted to human activity must be severely controlled. E. coli O157:H7 is one of entero-pathogens commonly present at very low concentrations in environmental

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waters within a diversified microflora. However, drinking water is not sterile and bacteria can be found in the distribution system and at the tap. Most of these organisms are harmless, but some opportunist pathogens such as *Pseudomonas aeruginosa* and *Aeromonas spp.* may multiply during distribution given suitable conditions [12].

2. Experimental

2.1. Detection and enumeration of intestinal enterococci

We use membrane filtration method as described on ISO 7899:2000. Intestinal enterococci bacteria are able to reduce 2,3,5-triphenyltetrazolium chloride to formazan and hydrolyze aesculin at 44°C \pm 0.5°C for 2h, on media Slanetz and Bartley medium and Bile-aesculin-azide agar. The enumeration of intestinal enterococci is based on filtration of a specified volume of water sample through a membrane filter with a pore size (0.45 um) sufficient to retain bacteria. The filter is placed on a solid selective medium containing sodium azide (to suppress the growth of Gramnegative bacteria) and 2.3.5triphenyltetrazolium chloride, a chlourless dye that is reduced to red formazan by intestinal enterococci. Typical colonies are raised, with a red, maroon or pink color, either in the center of the colony or throughout.

2.2. Detection and enumeration of Escherichia coli and the coliform bacteria

E. coli has been the foremost indicator of faecal contamination in water quality monitoring for many decades. *E. coli* has also been shown to be a significant reservoir of genes coding for antimicrobial drug resistance and therefore is a useful

indicator for resistance in bacterial communities [13]. A membrane filter procedure for enumerating E. coli was developed and evaluated. The method quantifies E. coli within 24h without requiring subculture and identification of isolates. It incorporates a primary selective-differential medium for gramnegative. lactose-fermenting bacteria: resuscitation of weakened organisms by incubation for 2h at 35°C before incubation at 44.5°C for 18 to 22h; and an in situ urease test to differentiate E. coli from thermotolerant, lactose-positive other organisms. Coliforms bacteria are lactasepositive and can produce indole from tryptophan at 44 \pm 0.5°C for 21-24h incubation, on medium containing caseine and agar. Indole in the presence of Kovac's reagent leads to the formation of red nitrozoindol. Red ring appearance certifies that the presence of *E. coli* in the tube.

2.3. Determination of chlorine and total chlorine

ISO 7393-2 Determination with N,Ndiethylphenylene-1,4-phenylenediamine, for rutine control purposes where as a reaction product a red compound result at a pH 6.2-6.5 measured at 510 nm.

3. Results and discussions

The results observed during three years of monitoring are shown in the figures 1, 2 and 3, as it is shown the contamination incidence was below 3%, and the total number of the investigated samples was over 300/quarter (data not shown). A slightly decrease of contamination incidence with E. coli, Enterococcus and both can be observed between 2010 and 2012. However, there could not be established a correlation between the number of samples founded out of line for chloride and a the incidence of microbial

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contamination, despite the fact that chlorine treatment especially for the surface water sources is essential. We also must mention that Suceava area recorded large amounts of rainfall during summer

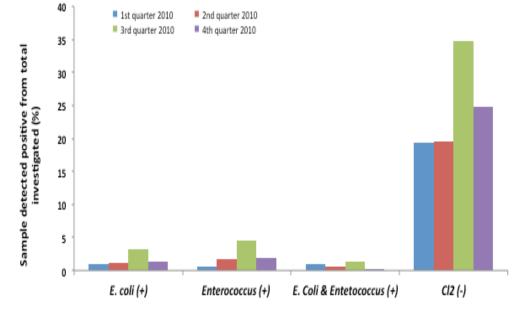


Figure. 1. The chlorine samples out of line and the contamination rate with *E. coli, Enterococcus* and both, on 2010 by quarters

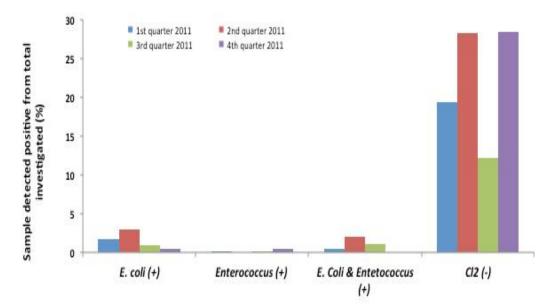


Figure. 2. The chlorine samples out of line and the contamination rate with *E. coli, Enterococcus* and both, on 2011 by quarters

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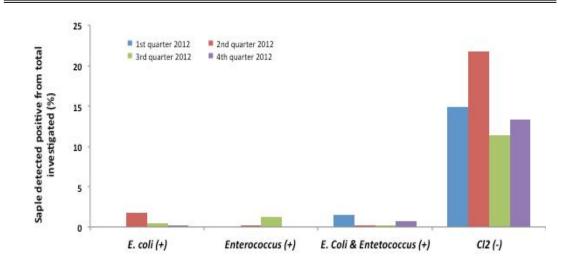


Figure. 3. The chlorine samples out of line and the contamination rate with *E. coli, Enterococcus* and both, on 2012 by quarters

on 2010 and spring of 2011 that might have a significant influence on the contamination rate observed in the analyzed samples. As it is shown in the figures a relatively low rate of contamination incidence has been recorded during this time interval. On the 2011 (first, second and third quarter) the contamination incidence with Enterococcus was almost none, among the

4. Conclusion

The quality of drinking water and possible associated health risks vary throughout the world. Microbial contamination of drinking water remains a significant threat and constant vigilance is essential, even in the most developed countries. Delivering safe and acceptable water, therefore, is a key target in improving public health in many developing countries. Overall, however, it is obvious that the supply and maintenance of safe drinking water remain key requirements for public health. Continuous and rigorous monitoring is required to avoid any large-scale contamination incidence.

analyzed samples. On the other hand on 2012 (second, third and fourth quarter) the contamination incidence with *E. coli* was very low. These results suggests that the modern technology and an accurate monitoring at the main water stations Berchisesti and Mihoveni from Suceava County, were essentially to assure the high water quality for the consumers.

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6. References:

 SUTHAR S, CHHIMPA V, SINGH S., Bacterial contamination in drinking water: a case study in rural areas of northern Rajasthan, India, *Environ Monit Assess.*, **159**, 43-50, (2008)
YASSIN M.M., ABU-AMR S.S., AL-NAJAR H.M., Assessment of

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microbiological water quality and its relation to human health in Gaza Governorate, *Gaza Strip. Public Health*, **120**, 1177–87, (2006)

[3]. ASHBOLT NJ., Microbial contamination of drinking water and disease outcome in developing regions, *Toxicology* **198**, 229–38, (2004)

[4]. CRICHTON PB., Enterobacteriaceae: Escherichia, Klensiella, Proteus and other genera. In: Colle JG, Fraser AG, Marmion BP, Simmons A, editors. *Mackie & McCartney Practical Medical Microbiology*. 14th ed. New York: Churchill Livingstone, (1996)

[5]. ENRIQUEZ C, NWACHUKU N, GERBA CP., Direct exposure to animal enteric pathogens, *Rev. Environ. Health*, **16**, 117–31, (2001)

[6]. PAYMENT P., RICHARDSON L., SIEMIATYCKI J., DEWAR R., EDWARDES M., FRANCO E., A randomized trial to evaluate the risk of gastrointestinal disease due to consumption of drinking water meeting current microbiological standards, *Am. J. Public Health*, **81**, 703–708, (1991)

MOORE AC, HERWALDT BL, [7]. **CRAUN** CALDERON RL, GF. HIGHSMITH AK. JURANEK DD. Surveillance for waterborne disease States. 1991-1992, outbreaks--United MMWR CDC Surveill Summ., 19, 42(5):1-22, (1993)

[8]. REYNOLDS K.A., MENA K.D., GERBA C.P., Risk of waterborne illness via drinking water in the United States, *Rev. Environ. Contam. Toxicol.* **192**,117-58, (2008)

[9]. CRAUN G.F., BRUNKARD J.M., YODER J.S., ROBERTS V.A., CARPENTER J., WADE T., CALDERON R.L., ROBERTS J.M., BEACH M.J., ROY S.L., Causes of outbreaks associated with drinking water in the United States from 1971 to 2006, *Clin. Microbiol. Rev.* 23, 507-28, (2010)

[10]. ARNOLD B., COLFORD J., Treating water with chlorine at point-of – use to improve water quality and reduce child diarrhea in developing countries: A systematic review and meta-analysis, *Am. J. Trop. Med. Hyg.*, **76**, 354-364, (2007)

[11]. VAN DEN BERG H., LODDER W., VAN DER POEL W., VENNEMA H., DE RODA HUSMAN A.M., Genetic diversity of noroviruses in raw and treated sewage water, *Res. Microbiol.*, **156**, 532– 40, (2005)

[12]. HUNTER P., Waterborne Disease. Epidemiology and Ecology, Chichester: Wiley, (1997)

[13]. BUCKNELL, D.G., GASSER, R.B., IRVING, A., WHITHEAR, K., Antimicrobial resistance in Salmonella and Escherichia coli isolated from horses, *Austrian Veterinary Journal*, **75**, 355-356, (1997)

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