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Original Scientific Paper**WATER IN FOOD INDUSTRY – MICROBIOLOGICAL REVIEW****Bojan GOLIĆ^{1*}, Drago N. NEDIĆ², Biljana PEĆANAC¹**¹PI Veterinary Institute of the Republic of Srpska “Dr Vaso Butozan” Banja Luka,
Bosnia and Herzegovina²University of Belgrade, Faculty of Veterinary Medicine, Belgrade, Serbia

*Corresponding author: Bojan Golić, bojan.golic@virs-vb.com

Summary

Water is used in different ways in the production of food, thus becoming a part of food. Washing water in the food industry can be a source of bacterial contamination and further endanger the quality and safety of food. The research included samples of water used in the food industry. The study includes 192 samples, of which 75% are from water supply and 25% from wells. Laboratory testing of water was carried out using BAS EN ISO 6222, BAS EN ISO 9308 and BAS EN ISO 7899-2 methods. The aim of the study is to determine the microbiological status of water in the food industry. The study determined 79.69% satisfactory samples and 20.31% unsatisfactory water samples. In relation to the tested parameters, in 12.50% of the samples, an increased number of microorganisms was found at 22°C, while in 10.42% of the samples, an increased number of microorganisms was found at 37°C. When it comes to hygiene indicators, 8.85% of unsatisfactory samples were due to the presence of intestinal enterococci, and 5.73% of unsatisfactory samples were due to the presence of *Escherichia coli* and coliforms. Considering the presence of coliform bacteria, *Escherichia coli* and intestinal enterococci in water used in the food industry, there is a risk of microbiological contamination of food through water. This is particularly significant considering the share of well water in relation to the total water used in the food industry.

Key words: food industry, water, microbiology.

INTRODUCTION

Water is used in different ways in food production, thus becoming part of food (Terplan, 1980). Contamination of food with water-borne microorganisms occurs directly, however, much more often indirectly after the multiplication of these microorganisms on the cleaned surfaces of the equipment used.

The microbiological quality of water is usually defined as the maximum acceptable number or concentration of bacteria that do not pose a health risk (EU, 2020). There is no level of zero probability of microbiological contamination of drinking water. Drinking water distribution and delivery systems cannot be completely sterile, and active growth of microorganisms is considered an indicator of malfunctions in water treatment or distribution units (Gottschal, 1992).

Washing water in the food industry can be a source of bacterial contamination and further endanger the quality and safety of food (Kivaria et al., 2006; Perkins et al., 2009). Also, water used during food handling and processing can be a potential source of microbial contamination with possible negative consequences for food safety.

Production and distribution of biologically safe drinking water can only be achieved by adequate monitoring and control of microbiological status during water treatment and distribution (Prest et al., 2016).

Water supply in food production premises should be subject to risk and hazard assessment to ensure maintenance of adequate water quality during the production process (Dawson, 1998; Dawson, 2000).

Many infectious diseases of animals and humans are transmitted by water contaminated with human and animal excrement, which becomes a source of pathogenic bacteria, viruses and parasites (protozoa, parasite eggs) capable of surviving in different periods and increasing the risk to the health of people (Fridrich et al., 2014; Sasakova et al., 2013).

Water temperature is an essential factor that affects the kinetics of bacterial growth and competition processes. Drinking water temperatures typically range between 3 and 25°C in European countries and vary seasonally within this temperature range even within a single drinking water distribution system (Niquette et al., 2001). Elevated water temperatures are often associated with increased abundance of bacteria in drinking water distribution systems, and with higher numbers of indicator organisms such as coliforms (Francisque et al., 2009; Liu et al., 2013).

To eliminate the risk of disease transmission, water intended for mass consumption is treated and disinfected before use (Edberg et al., 2000). Based on the results, adequate measures can be taken that include prevention of contamination and systemic disinfection. Many pathogenic microorganisms are present only under

certain conditions and, when present, occur in small numbers compared to other microorganisms.

Indicator organisms are used to assess the microbiological quality of water. The use of indicator bacteria, especially *Escherichia coli* and coliform bacteria, as a factor of assessing the potential presence of waterborne pathogens is essential for public health protection (Hijnen et al., 2000). *Escherichia coli* is a coliform bacterium and is considered the primary indicator of fecal contamination of water. Enterococci include numerous species that occur in the feces of humans and warm-blooded animals (WHO, 2008). The main reason for their determination is to assess the significance of the presence of coliform bacteria in the absence of *Escherichia coli* or to provide additional information when assessing the degree of possible fecal contamination. As such, they are considered secondary indicators of fecal pollution. The bacteria is present in drinking water, even in relatively large numbers (10^3 to 10^6 CFU/ml), without consequences for human health. The presence of bacteria in drinking water is not a problem by itself, as long as no pathogenic microorganisms are present (Hoefel et al., 2005; Hammes et al., 2008; Vital et al., 2012).

The aim of the study is to determine the microbiological status of water used in the food industry in the territory of the Republic of Srpska.

MATERIALS AND METHODS

The study included samples of water used in the food industry. The samples were taken from facilities for processing meat, milk and fish and from catering facilities (ready-to-eat food) from the territory of the Republic of Srpska (Bosnia and Herzegovina). Sampling was done in such a way that all attachments were removed from the tap, the water was allowed to flow for 2-3 minutes, after which the tap was closed and the tap was disinfected with alcohol and flame sterilized, and the water was allowed to flow again for 2-3 minutes. After that, the water was poured into darkened sterile glass bottles with a volume of 1 liter, whereby water in the amount of $\frac{3}{4}$ of the volume of the bottle was poured into them. The samples were transported at a temperature of $5\pm 3^\circ\text{C}$, and the test was performed within 6 hours of sampling.

The study was carried out in year 2022 and included 192 samples, 75% of which were from water supply and 25% from wells.

Laboratory testing of water was carried out by the following methods:

- the number of microorganisms at 22°C and 37°C according to BAS EN ISO 6222 (ISBIH, 2003a),
 - number of intestinal enterococci according to BAS EN ISO 7899-2 (ISBIH, 2003b),
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- the number of coliform bacteria and *Escherichia coli* according to BAS EN ISO 9308 (ISBIH, 2018).

The Rulebook on the Health Safety of Water Intended for Human Consumption (Rulebook, 2017) establishes test parameters and microbiological criteria (Table 1).

Table 1 Test parameters and microbiological criteria

Parameter	Criteria
Number of microorganisms at 22°C	100CFU/1 ml
Number of microorganisms at 37°C	20CFU/1 ml
Intestinal enterococci	0CFU/100 ml
<i>Escherichia coli</i>	0CFU/100 ml
Coliforms	0CFU/100 ml

In our study and in the statistical analysis of the obtained results, we used descriptive statistical parameters (Excel, Microsoft Office 2019) as basic statistical methods. The results of the study are presented in tables and graphics.

RESULTS AND DISCUSSION

The conformity assessment of the samples was carried out in relation to the Rulebook on the Health Safety of Water Intended for Human Consumption (Rulebook, 2017). The test determined 79.69% satisfactory and 20.31% unsatisfactory samples, and the test results by category are shown in Figure 1.

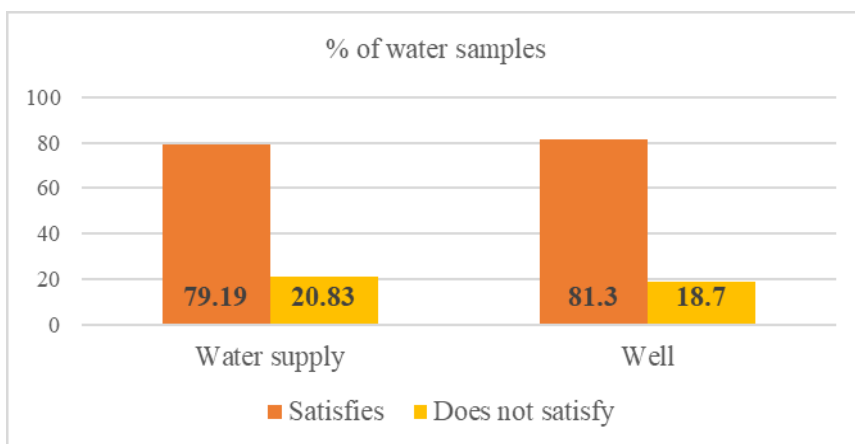


Figure 1 Test results by category in %

During the period from year 2015 until year 2017, Kalaba et al. (2020) found 26.20% unsatisfactory samples, out of 584 water samples. Analysis of water from milk collection points, originating from wells, revealed 63.90% of unsatisfactory samples (Jaki et al., 2010). In the study done by Golić et al. (2021) 88.16% satisfactory and 11.84% unsatisfactory samples were found.

Looking at the total number of analyzed samples, the obtained results are significantly less favorable than the results of Golić et al. (2021) for water originating from the water supply, while the situation is reversed when it comes to well water samples. The obtained results are in accordance with the results of Kalaba et al. (2020). The obtained results are unexpected, that is, they indicate a potential illogicality, because it is to be expected that there will be more satisfactory results for water from the water supply compared to well water, given that the water supply is under regular daily supervision, and the wells periodically. A possible explanation for these results is that when it comes to the water supply system, that is, the public water supply system, the distributor is responsible for the microbiological safety of the water to the point of distribution, and from it to the point of use, the water user is responsible, which may indicate possible poor quality or malfunction of installations from the point of distribution to the point of use and the consequent contamination of water.

Table 2 shows the unsatisfactory test results of the total number of samples in relation to the test parameters, and Table 3 shows the unsatisfactory test results of the samples by category in relation to the test parameters.

Table 2 Unsatisfactory samples in relation to test parameters

Parameter	Unsatisfactory samples in %
Number of microorganisms at 22°C	12.50
Number of microorganisms at 37°C	10.42
Intestinal enterococci	8.85
<i>Escherichia coli</i>	5.73
Coliforms	5.73

Table 3 Unsatisfactory samples by category in relation to test parameters

Parameter	Unsatisfactory samples in %	
	Water supply	Well water
Number of microorganisms at 22°C	11.81	6.25
Number of microorganisms at 37°C	11.81	6.25
Intestinal enterococci	6.94	14.58
<i>Escherichia coli</i>	5.56	6.25
Coliforms	5.56	6.25

According to WHO (2008), *Escherichia coli* is the only true indicator of fecal contamination, as it is exclusively of intestinal origin and is found in feces. Its presence indicates mostly fresh fecal contamination and thus indicates serious deficiencies in the protection of the specific water source, water treatment and its hygienic safety. The number of microorganisms at 22°C and at 37°C represents the general population of heterotrophic bacteria present in water supplies. They can represent bacteria whose natural habitat is the aquatic environment or those originating from soil or vegetation. The number of heterotrophs includes all microorganisms capable of growing in or on solid nutrient-rich agar. Two temperatures and two incubation times are used for the analyses, at 37°C for 48 hours to encourage the growth of bacteria of mammalian origin and at 22°C for 72 hours to encourage the growth of bacteria originating mainly from environmental sources. The test results indicate that the causes of microbiological deterioration of water come from both animals and the environment. When it comes to wells, contamination from animals is very possible due to inadequate drainage of waste and fecal water and the consequent contamination of groundwater. The obtained results are in accordance with the results of Kalaba et al. (2020) and indicate significant fecal contamination of water, especially intestinal enterococci. In relation to the results of Golić et al. (2021), a significantly higher percentage of unsatisfactory samples was determined. Observing the obtained results, it is observed that the presence of indicator organisms is greater in well water, while the number of microorganisms at 22°C and at 37°C is higher in water from water supply. Although the presence of coliform bacteria does not always indicate a threat to public health, their detection is a useful indication that water treatment operations should be investigated (Edberg, 2000).

CONCLUSION

Considering the presence of coliform bacteria, *Escherichia coli* and intestinal enterococci in water used in the food industry, there is a risk of microbiological contamination of food through water. This is particularly significant considering the share of well water in relation to the total water used in the food industry. For this reason, it is necessary to make effort to achieve the best possible quality of drinking water.

Conflict of interest statement: The authors declare that there is no conflict of interest.

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