



Microbiological status of minced meat at retail in Belgrade district

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ABSTRACT

This study was conducted to determine the microbiological status of 390 beef and pork minced meat samples collected over three years from 52 retailers in the territory of Belgrade. The numbers of aerobic colony counts, *Escherichia coli*, and the presence of *Salmonella* spp. are prescribed criteria for this group of meat semi-products. *Salmonella* spp. was confirmed in one sample of minced beef meat (0.8%), while unsatisfactory *E. coli* counts were only determined in pork meat samples (2.7%). In 2021, all samples complied with the microbiological criteria for minced meat. The highest occurrence of positive samples was observed during the III quarter of 2022 (P=0.04) with a frequency of 9.3%. The level of contamination of minced pork with *E. coli* bacteria in the same quarter was significantly higher compared to the II quarter of 2022 (627±75 vs. 292±9 cfu/g, P=0.009). Improvement of process hygiene and revision of process control, along with permanent education of food staff on the principles of GMP and GHP, are necessary for maintaining food safety and public health.

1. Introduction

Foodborne pathogens are a leading cause of disease and death in developing countries. According to the World Health Organization (WHO), the global burden of foodborne diseases is estimated at 600 million people (WHO, 2014). Foodborne diseases are preventable if food protection principles are followed from primary production to the consumer level. Changes in eating habits, mass catering, unsafe food storage conditions, and poor hygiene practices are significant contributors to foodborne illness. The cold chain must be cohesive for temperature-sensitive products and meat semi-products. If only one link is interrupted in the chain, that causes irrevers-

ible damage and these products or semi-products could be unsafe for consumers (Betić, 2019).

In Serbia, food business operators (FBOs) have been obliged, according to the Law of Food Safety (Serbia, 2019), to adhere to good hygiene and good production practices as prerequisites for hazard analysis and critical control point (HACCP) programs. Within the implemented HACCP, the FBOs must also have a self-control plan, proving that they control the system. HACCP, with its prerequisite programs, good manufacturing practice (GMP) and good hygiene practices (GHP), requires the FBOs to identify potential hazards that threaten to endanger the safety of the product in order to eliminate them and keep them under control.

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This study aimed to determine the microbiological risks and compliance with the applied HACCP in minced pork and beef purchased in retail stores and in front of the customer on the spot in the territory of the city of Belgrade.

2. Materials and methods

Sample collection was undertaken from January 2020 to December 2022 (Table 1). During this period, 390 samples were collected at 52 retailers throughout Belgrade, i.e., 127 samples of minced beef and 260 samples of minced pork. Samples were taken according to the self-control plan prescribed by FBOs; the dynamics ranged from monthly to quarterly, depending on the size of the facility. The raw meat came from two different slaughterhouses in Serbia. Minced meat samples (500 g from each cut and minced at the butchery) were also collected in 5 sterile plastic bags from each retailer.

After sampling, the samples were conditionally stored in hand refrigerators (4°C) and transported to the Institute of Meat Hygiene and Technology in Belgrade for immediate analysis. Minced meat was analysed using standard accredited methods, SRPS EN ISO 6579-1:2017 (*Salmonella* spp.), SRPS EN ISO

16649-2:2008 (*Escherichia coli*), SRPS EN ISO 4833-1:2014 (aerobic colony count). Each meat product placed on the market must, during its shelf life, meet the food safety criteria clearly defined by the Regulation in the EU (EU, 2005), with which the legal regulations in our country are harmonized (Serbia, 2010, 2018). Limits for food safety and process hygiene test results for minced meat are shown in Table 2.

The statistical analyses of the results were performed using SPSS 21. The frequency of positive results, considering meat type and sampling years and quarter periods, was calculated using a chi-square test. Differences in *E. coli* contamination levels were analysed with one-way ANOVA.

3. Results

During three years, 390 samples were collected from 52 retailers from the territory of the Belgrade district. The mandatory criteria for minced meat were not complied with for 2.1% (8/390) of investigated samples (Table 3). Of this, 0.8% (1/127) of beef minced meat was defective, while 2.7% (7/260) of pork samples tested positive. Considering the meat type ($P=0.22$) and sampling year ($P=0.09$), there was no difference in the occurrence of the non-compliant samples. In beef

Table 1. Sampling scheme during the three years

Year	I quarter	II quarter	III quarter	IV quarter	Total
2020	27	7	21	23	78
2021	25	31	28	30	114
2022	32	59	54	53	198
Total	84	97	103	106	390

Table 2. Limits for minced meat according Regulation EU and Serbian law

Microorganisms	Sampling plan*		Limits		Analytical reference method	Stage where the criterion applies
	N	c	M	M	Analytical reference method	Stage where the criterion applies
<i>Salmonella</i> spp.	5	0	Absence in 10 g		EN ISO 6579	Products placed on the market during their shelf-life
<i>Escherichia coli</i>	5	2	50 cfu/g	500 cfu/g	EN ISO 16649-1 or EN ISO 16649-2	End of the manufacturing process
Aerobic colony count	5	2	5x10 ⁵ cfu/g	5x10 ⁶ cfu/g	EN ISO 4833	End of the manufacturing process

*n = number of units comprising the sample; c = number of sample units giving values between m and M.

minced meat, *Salmonella* spp. was determined during the III quarter of 2022, and was present in 3/5 of the tested meat samples. In contrast, the presence of *E. coli* bacteria in all non-compliant samples was predominant in minced pork meat, in all five examined units of these samples. It was observed that during 2021, all samples met the microbiological criteria for minced meat.

Concerning the results from the research by Mrdovic et al. (2019) in which the presence of *Salmonella* spp. was detected in 1.6% of the examined samples, as was the presence of *E. coli* in 5% of the tested samples, the results in this study showed a lower incidence of *Salmonella* spp. as well as of

E. coli prevalence. On the other hand, the results obtained in this study showed a higher presence of pathogens than in Ireland, where the prevalence of *Salmonella* spp. was 0.1% and *E. coli* occurred in 0.2% of similar products (Ireland, 2013). Regarding data from 2010, published by EFSA and ECDC and originating in 12 countries in the EU, 2.8% of such meat preparations contained *Salmonella* spp., and 0.6% contained *E. coli* (EFSA and ECDC, 2012).

The frequency of positive samples during the quarter periods was statistically significant ($P=0.01$), with the highest occurrence during the III quarter of 2022 (Table 4). In the given period, all samples with

Table 3. The results of microbiological criteria testing for minced meat

	Satisfactory (%)	Unsatisfactory (%)	Total (100%)	P value
Pork meat	256 (97.3)	7 (2.7)	263	0.22
Beef meat	126 (99.2)	1 (0.8)	127	
2020	77 (98.7)	1 (1.3)	78	0.09
2021	114 (100)	0 (0.0)	114	
2022	191 (96.5)	7 (3.5)	198	
I quarter	84 (100)	0 (0.0)	84	0.01
II quarter	95 (97.9)	2 (2.1)	97	
III quarter	97 (94.2)	6 (5.8)	103	
IV quarter	106 (100)	0 (0.0)	106	
Total	382 (97.9)	8 (2.1)	390	

Table 4. The results of microbiological criteria testing of minced meat during the quarter periods of year

		Satisfactory (%)	Unsatisfactory (%)	Total (100%)	P value
2020	I quarter	27 (100)	0 (0.0)	27	0.43
	II quarter	7 (100)	0 (0.0)	7	
	III quarter	20 (95.2)	1 (4.8)	21	
	IV quarter	23 (100)	0 (0.0)	23	
2021	I quarter	25 (100)	0 (0.0)	25	-
	II quarter	31 (100)	0 (0.0)	31	
	III quarter	28 (100)	0 (0.0)	28	
	IV quarter	30 (100)	0 (0.0)	30	
2022	I quarter	32 (100)	0 (0.0)	32	0.04
	II quarter	57 (96.6)	2 (3.4)	59	
	III quarter	49 (90.7)	5 (9.3)	54	
	IV quarter	53 (100)	0 (0.0)	53	

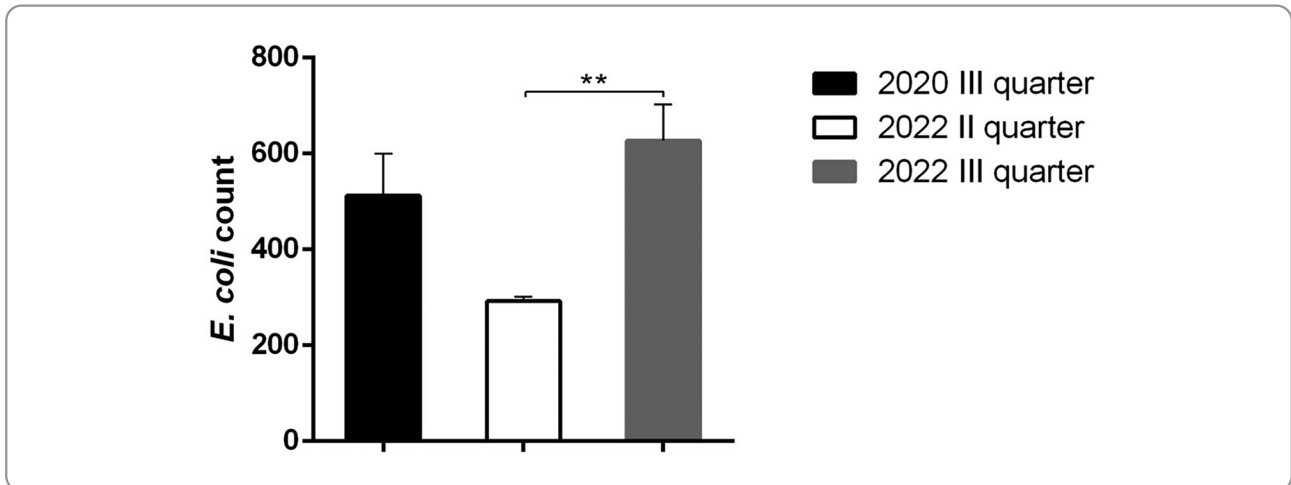


Figure 1. Level of contamination of minced pork with *Escherichia coli*

E. coli contamination were pork. Considering the contamination level of pork meat with *E. coli*, positive samples from the III quarter had a higher count than those from the II quarter of 2022 (627 ± 75 vs. 292 ± 9 cfu/g, $P=0.009$) (Figure 1). However, the same pork meat sampled in the III quarter of the 2020 year, in addition to a high mean *E. coli* contamination level (512 ± 88 cfu/g), also had a high level of aerobic colony count ($1.02 \pm 0.06 \times 10^6$ cfu/g). Consequently, most of the samples that did not comply with the required criteria were sampled during the III quarter, which was in the summer period of the year. The summer period is more challenging for maintaining the cold chain, which must not be interrupted at any time along the meat distribution chain, in the continuum from slaughterhouse to consumer (Nastasijević *et al.*, 2017).

4. Conclusion

This study showed the presence of *Salmonella* spp. in one sample of minced beef meat and *E. coli* in seven samples of minced pork meat. FBOs must improve process hygiene and review process control, and permanent education of food staff on the principles of GMP and GHP, like the pre-requisite programs of HACCP, is necessary. Particular attention should be given to the storage of foods. Fresh meat is highly perishable and has a short shelf life, which means the time available for product distribution is also short. If fresh meat and meat products are not kept at adequate, controlled temperatures, they can be excellent environments for pathogenic and other bacterial growth.

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