

## RESEARCH ARTICLE

# Determination of the Microflora of Meatless Cig Kofte in the Black Sea Region of Turkey

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## Abstract

**Objective** Meatless Cig Kofte (MCK) is a traditional product consumed frequently in Turkey. In recent years, MCK shops have been opened and offered to the public. It is important for public health because it is a risky food for microbial contamination. This study was conducted to investigate some microbiological quality of MCK sold in Black Sea region (Trabzon, Giresun, Ordu, Samsun) in Turkey.

**Methods:** In this study, a total of 120 MCK were sampled from different points of sales and examined for the microbiological quality of MCK in the Black Sea Region (Trabzon, Giresun, Ordu and Samsun). Data analysis of the study was conducted using SPSS 24 package program. Statistical significance was evaluated at  $p < 0.05$ .

**Results:** According to the research results, the highest average TMAB (Total Mesophilic Aerobic Bacteria) value of MCK samples was determined in Giresun province (6.04 log cfu/g) and the lowest average value was found in Samsun province. While the average TPAB (Total Psychophilic Aerobic Bacteria) was determined as 1.82 log cfu/g in MCK samples collected from Samsun province, it could not be detected in Trabzon, Ordu and Giresun provinces. The average *S. aureus* value of MCK samples was determined in the highest province of Ordu (4.22 log cfu/g) and the lowest in Trabzon province (1.94 log cfu/g).

**Conclusion:** It was concluded that microbial quality was insufficient and hygiene rules were not taken into consideration in the provinces that were included in this study conducted in the Black Sea Region (Trabzon-Giresun-Ordu-Samsun).

**Key words:** Black Sea Region; meatless cig kofte; microbial quality; public health

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## Introduction

MCK may include bulgur, red pepper, onion, garlic, various spices, tomato paste, tomato sauce, pepper paste, sugar, vinegar, pomegranate syrup, walnut, almond, hazelnut, water and some or all vegetable oils. In addition, when the necessary additives in accordance with the legislation are mixed and kneaded by hand or by machine until it reaches a certain consistency, it becomes ready for consumption without any heat treatment or freezing (TSE, 2018).

MCK, which is one of the ready-to-eat foods, is consumed without heat treatment. Inadequate

personnel hygiene during production, contamination from the tools and equipment, and the additives in the composition of MCK, can cause food poisoning (Ardıç and Durmaz, 2008).

Microbial analyses are performed to determine the quality of food products in terms of public health. There is a risk of spoilage occurrence during storage since no heat treatment and microbial inhibition treatment is applied to ready-to-eat foods until the time to sale (Delikanlı et al., 2014). Species of vegetable origin may include fertilizer, water, soil and the loads of animal-borne microorganisms. It is possible that microorganisms can get contaminated because of the spices used in the production of MCK.

The aim of this study is to investigate the MCK which is offered for sale in the Black Sea Region (Trabzon-Giresun-Ordu-Samsun) in terms of microbial quality. The reliability of MCK, which are liked and consumed by consumers, has been investigated in terms of public health.

## Methods

### Sample collection

In this study, a total of 120 MCK samples (30 grams from each city) were collected from different point of sales in the Black Sea Region (Trabzon, Giresun, Ordu and Samsun) between October, 2018 and January, 2019, and each sample was 200 grams. All MCK samples were carried in a mini cooler and brought to the Giresun University laboratory, and the analyses were initiated within an hour after sample collection (figure 1).



A. MCK Samples in block form B. Packaged MCK samples C. MCK Samples

**Fig. 1.** Samples of MCK, which is a traditional product sold at different points of sales. A. The Block MCK samples in chilled or frozen form. B. MCK samples in packaged form C. Ready-to-eat MCK samples

### Determination of pH

The pH values of the MCKs were determined by using a digital pH meter (OHAUS, Starter 3000, Germany).

### Microbiological analyses

The MCK samples, each of which weighed 10 grams, were brought to the laboratory under aseptic conditions and homogenized with 90 mL Maximum

Recovery Diluent (MRD, Merck, Germany). Dilution of the solution obtained from  $10^{-1}$  dilution up to  $10^{-6}$  was made. The selective agar was inoculated with 0.1 mL of 2 parallels 3 replicas of each dilution. The inoculated petri dishes were incubated under appropriate conditions. The most probable number (MPN) method was used for the detection of *E. coli* and coliform microorganisms (Table 1) (Halkman, 2005; Andrews, 1992).

### Statistical analysis

Data were analyzed using One way ANOVA and Duncan's post hoc tests by using IBM SPSS 24 software (SPSS Inc., Chicago, IL, USA) with a significance level of 5% (Table 1).

Microorganism	Medium	Incubation conditions	
		°C	Time
TMAB	Plate Count Agar (PCA)	37 °C	24-48 h
Yeast and Mold	Potato Dextrose Agar (PDA)	25-28 °C	4-5 days
<i>S. aureus</i>	Baird Parker Agar (BPA)	37 °C	24 h
TPAB	Plate Count Agar (PCA)	4,5 °C	14 days
Coliform Microorganism	Laurly Sulfate Tryptose	30°C	24 h
<i>E. coli</i>	Laurly Sulfate Tryptose	37 °C	24 h

## Results

As a result of TMAB analysis, significant differences were found in the MCK samples obtained from Trabzon and Giresun provinces. While the samples from Trabzon province gave the lowest value with an average value of 4.66, the samples obtained from Giresun province gave the highest value with an average of 5.44 (Table, 2).

As a result of TPAB analysis, significant differences were found in the MCK samples obtained from Trabzon and Samsun provinces. While the samples from Trabzon province had the lowest value with an average value of 0.65, the samples obtained from Samsun province had the highest value with an average of 2.74 (Table, 2).

As a result of *S. aureus* analysis, there was no significant difference between the MCK samples obtained from Giresun, Ordu and Samsun. While the MCK samples from Trabzon province had the lowest value with an average value of 2.24, the samples obtained from Samsun province had the highest value with an average of 3.72 (Table, 2).

According to yeast and mold analysis results, no statistically significant difference was observed in the samples obtained from 4 provinces. While the MCK

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samples from Trabzon province had the lowest value with an average value of 3.37, the samples obtained from Samsun province had the highest value with an average of 4.52 (Table, 2).

As a result of coliform analysis, significant differences were found in the MCK samples obtained from Trabzon and Ordu provinces. The lowest value in the MCK samples was found in Trabzon province with an average of 2.09 and the highest value was

found in the MCK samples obtained from Ordu province with 40.66 (Table, 2).

According to *E. coli* analysis, significant differences were found in the samples obtained from Trabzon and Ordu provinces. The lowest *E. coli* value was found in the MCK samples from Trabzon with an average of 1.25 and the highest value was found in the MCK samples obtained from Ordu province with 38.90 (Table, 2).

**Table 2.** TMAB, TPAB, *S. aureus* and yeast-mold analysis results of the MCK samples statistical analysis results table

Analysis	TRABZON	GIRESUN	ORDU	SAMSUN
<b>Total Mesophilic Aerobic Bacteria (log cfu/gr)</b>	4.66 ± 1.48 <sup>a</sup>	5.44 ± 0.81 <sup>b</sup>	5.25 ± 1.21 <sup>ab</sup>	5.18 ± 0.46 <sup>ab</sup>
<b>Total Psychophilic aerobic Bacteria (log cfu/gr)</b>	0.65 ± 1.70 <sup>a</sup>	1.83 ± 2.71 <sup>ab</sup>	1.94 ± 2.20 <sup>ab</sup>	2.74 ± 2.34 <sup>b</sup>
<b><i>S. aureus</i> (log cfu/gr)</b>	2.24 ± 2.01 <sup>a</sup>	3.42 ± 1.40 <sup>b</sup>	3.52 ± 1.22 <sup>b</sup>	3.72 ± 1.08 <sup>b</sup>
<b>Yeast and Mold (log cfu/gr)</b>	3.37 ± 2.15 <sup>a</sup>	3.51 ± 2.48 <sup>a</sup>	4.13 ± 2.15 <sup>a</sup>	4.52 ± 1.36 <sup>a</sup>
<b>Coliform (MPN/gr)</b>	4.57 ± 2.09 <sup>a</sup>	25.63 ± 41.34 <sup>ab</sup>	40.66 ± 51.10 <sup>b</sup>	26.23 ± 43.68 <sup>ab</sup>
<b><i>E. coli</i> (MPN/gr)</b>	1.25 ± 3.16 <sup>a</sup>	24.4 ± 41.39 <sup>ab</sup>	38.90 ± 51.46 <sup>b</sup>	25.11 ± 41.42 <sup>ab</sup>

Different letters of mean and standard deviation values in the same line show statistically significant differences between the groups. (p<0.05)

**Table 3.** Maximum and minimum pH values of MCK samples

	Min Value	Max Value
<b>TRABZON</b>	3.50	4.20
<b>GIRESUN</b>	3.89	4.35
<b>ORDU</b>	4.70	5.10
<b>SAMSUN</b>	3.51	5.15

**Table 4.** The number of TMAB, TPAB, *S. aureus* and Yeast-Mold findings of MCK samples sold in Trabzon, Giresun, Ordu and Samsun provinces in the Black Sea Region

	TMAB (log cfu/gr)	TPAB (log cfu/gr)	<i>S. aureus</i> (log cfu/gr)	Yeast and Mold (log cfu/gr)
<b>TRABZON</b>	Minimum Value	ND	ND	ND
	Maximum Value	5.98	5.51	4.77
	Average Value	5.46	ND	1.94
<b>GIRESUN</b>	Minimum Value	4.35	ND	ND
	Maximum Value	7.84	6.61	4.43
	Average Value	6.04	ND	2.07
<b>ORDU</b>	Minimum Value	ND	ND	ND
	Maximum Value	7.38	6.13	4.46
	Average Value	5.81	ND	4.22
<b>SAMSUN</b>	Minimum Value	3.71	ND	ND
	Maximum Value	5.98	5.55	5.38
	Average Value	5.15	1.82	3.96

\* ND: not Detected

**Table 5.** The number of Coliform and *E. coli* analysis results of the MCK samples obtained from Trabzon, Giresun, Ordu and Samsun in the Black Sea Region

		Microorganism	
		<i>E. coli</i> (MPN/g)	Coliform (MPN/g)
TRABZON	Minimum Value	ND	ND
	Maximum Value	15	4.3
	Average Value	1.25	2.09
GIRE SUN	Minimum Value	ND	ND
	Maximum Value	110	110
	Average Value	24.40	25.63
ORDU	Minimum Value	ND	ND
	Maximum Value	110	110
	Average Value	38.90	40.66
SAMSUN	Minimum Value	ND	ND
	Maximum Value	110	110
	Average Value	25.11	26.23

\* ND: not Detected

### Discussion

The aim of this study is to examine the microbiological quality of MCK offered for sale in different provinces of the Black Sea region (Samsun, Ordu, Giresun and Trabzon) and to make an assessment in terms of Public Health. The results of 120 MCK samples collected for this purpose are presented in tables. The results of microbial analysis of 120 MCK samples are given in Tables.

It has been determined that the most important factor in *S. aureus* transmission to food in cases of food-borne intoxications is human (Akpınar et al., 2019).

*S. aureus* can be found in raw and processed foods, meat and meat products, salads, skin and nasal mucosa of healthy human (Erol, 2007). The presence of *S. aureus* in foods is due to the poor hygienic practices of the staff, cross contamination during preparation or improper storage (Pamuk et al., 2013). These bacteria can cause meningitis, septicemia, inflamed wounds and joint rheumatism in humans (Akpınar et al., 2019). High risk conditions during food preparation should be well explained to working personnel (Pamuk et al., 2013).

The number of *S. aureus* in MCK samples from Trabzon, Giresun, Ordu and Samsun provinces was determined as follows: 1.94 log cfu/gr, 2.07 log cfu/gr, 4.22 log cfu/gr, 3.96 log cfu/gr (as mean values). It can be stated that *S. aureus* values of the MCK samples collected from Trabzon and Giresun provinces are low, and *S. aureus* values of the MCK samples collected from Ordu and Samsun provinces are high when compared to the average *S. aureus* results (5.03x10<sup>3</sup> log cfu/g) of the research on the

MCK samples collected from Diyarbakır province by Vural and Yeşilmen (2003).

The temperature range for the development of *S. aureus* is 6.7-47.8°C and the optimum is 35-37°C. *S. aureus* can be easily eliminated during pasteurization or cooking. Also, the optimal pH range for the development of *S. aureus* is 6.0-7.1, and most of the strains can reproduce at pH values between 4.4-10 (Erol, 2007).

The minimum and maximum pH values of Trabzon, Giresun, Ordu and Samsun provinces were as follows; 3.50-4.20, 3.89-4.35, 4.70-5.10 and 3.51-5.15 (Table 3). In a study by Kurt et al. (2019), the pH of MCK was found out to be in the range of 3.99-4.84. Compared to the minimum pH value (3.99) found by Kurt et al. (2019), it was found that the MCK samples collected from Trabzon, Giresun and Samsun provinces have lower pH values, and the MCK samples from Ordu province have higher pH values. When compared to the pH values of this present study, it can be stated that the maximum pH values (4.84) found in the MCK samples by the same researchers are higher than the samples collected from Trabzon and Giresun provinces, and Ordu and Samsun samples have higher pH values.

In a study where Cerit et al. (2014) analyzed MCK obtained from Sakarya province, the pH value was found in the range of 4.8-5.2. When compared with the results of their study, it can be stated that the minimum (4.8) and maximum (5.2) values were higher than the pH values of the samples collected from all the provinces examined in this research.

The pH values of the research results are within the range of the pH value (3.5-5.5) given for the

Tartar a la turca (Meatless) at the Turkish Standards Institute (TSE, 2018).

Determination of TMAB and yeast-mold presence in foods reveals the microbial quality of food (Delikanli, 2014). These analyzes are indicative of whether the points of food sales obey the hygiene and sanitation rules. The average value of TMAB in MCK samples was determined as 5.46 log cfu/g, 6.04 log cfu/g, 5.81 log cfu/g, 5.15 log cfu/g in Trabzon, Giresun, Ordu and Samsun provinces, respectively (Table 4).

The mean value of TMAB in MCK samples was determined as follows: Trabzon; 5.46 log cfu/g, Giresun; 6.04 log cfu/g, Ordu; 5.81 log cfu/g, Samsun; 5.14 log cfu/g in Trabzon, Giresun, Ordu and Samsun provinces respectively (Table 4).

The average TMAB values of the study were found out to be lower when compared to the study conducted by Kardeş (2017) (average 7.86 log cfu/g). It can be stated that the average TMAB values of samples obtained from Trabzon, Giresun and Ordu provinces are high when compared to the findings (1.6x10<sup>5</sup> cfu/g) of the study by Cerit et al. (2014), while the average values of the samples obtained from Samsun province are low.

The average TPAB values of the study were found out to be lower when compared to the study conducted by Aslan et al. (2012) (average 4.54 log cfu/g).

The average number of yeast and mold in MCK samples from Trabzon, Giresun, Ordu and Samsun provinces are as follows respectfully; 4.57 log cfu/g, 2.54 log cfu/g, 4.98 log cfu/g, 5.03 log cfu/g. The results of this study (on average) were found to be lower when compared to the results (7.44 log cfu/g) of the study of MCK obtained from Siirt province by Kardeş (2017). The yeast mold average values of MCK samples collected from all provinces studied in this research were found to be low compared to the values (7.44 log cfu/g) of the study done by Kardeş (2017) on MCK samples obtained from Siirt province. When the findings of this study are compared with the average yeast-mold results (3.32 log cfu/g) of the study on MCK samples by Cerit et al. (2014), the average values of the samples collected from Giresun province are lower compared to their research findings and the average values of the samples collected from Trabzon, Ordu and Samsun provinces are higher. When the findings of this study are compared in terms of the number of yeast-mold with the research on MCK offered for the consumption in the center of Bursa the results (2.9x10<sup>4</sup> cfu/g) found by Delikanli et al. (2014) are lower than the average value obtained from the

samples collected from Trabzon, Ordu and Samsun provinces, while they are higher than the samples collected from Giresun province.

When compared to the results (2.9x10<sup>4</sup> cfu/g (4.46 log cfu/g) of the study on MCK samples obtained from the center of Bursa by Delikanli et al. (2014), in terms of the number of yeast-mold, the average value of samples collected from Trabzon, Ordu and Samsun provinces is high and the average value of samples collected from Giresun province is low. The most important group in Food Microbiology is coliform microorganisms and they are evaluated as the indicators of hygiene in foods. For some foods, they are intended to 'disappear' in a certain amount of samples, while in many foods, plant and soil origin coliforms are allowed to be present in certain numbers. For this purpose, counting or presence-absence tests are applied in coliform group analyses (Akpınar et al., 2019).

Average coliform microorganisms in MCK samples collected from Trabzon, Giresun, Ordu and Samsun provinces are 2.09 MPN/g, 25.63 MPN/g, 40.66 MPN/g, 26.23 MPN/g respectively. When compared to the results of this present study, the average coliform microorganism value (3.7x10<sup>1</sup> MPN/g) of the MCK samples obtained from Sakarya province by Cerit et al. (2014) is higher than Trabzon, Giresun and Samsun provinces and lower than Ordu province.

Coliform group bacteria are considered as a hygiene indicator in foods (Akpınar et al., 2019). Non-pathogenic strains of *E. coli*, one of the coliform group members, are found in the normal intestinal flora of all humans and generally all those that are warm-blooded (Erol, 2007). The presence of *E. coli* in samples of food, environment etc. indicates direct fecal contamination. *E. coli* can often be found in many foods due to poor hygienic conditions (Akpınar et al., 2019).

The findings of this study on the determination of average *E. coli* count are as follows: 1.25 MPN/g, 24.40 MPN/g, 38.90 MPN/g, 25.11 MPN/g respectively for Trabzon, Giresun, Ordu and Samsun provinces. When compared with the average values of this study, the average *E. coli* value (1.6x10<sup>1</sup> MPN/g) of the study by Cerit et al. (2014) is lower than Giresun, Ordu and Samsun provinces and higher than Trabzon provinces.

In the literature search, microbiological, serological and histological studies were mostly used on the meatballs (including raw meat), and different parameters were used.

### Conclusion

As a result, MCK, as a food product ready for consumption is traditionally consumed in Turkey. Since the materials of the MCK are not heat treated and the preparation stage is manual, they can create food safety problems for the consumer under unhygienic production conditions. In addition, depending on the production and storage conditions, the first hygienic quality of raw materials is important for microbial development. In MCK which is sold by retail, production can be made under conditions that do not comply with hygiene and sanitation rules, and the product may be spoiled because of storing it under inappropriate temperature conditions. Many indicator and pathogen microorganisms can be found and develop in the composition of MCK, which is dangerous for human health. In this study, microbial quality of MCK ready for consumption has been examined in four pilot provinces in Black Sea Region. According to the research findings, it is stated that production and sales conditions of MCK affect the microbial flora of personnel hygiene and sanitation.

**Ethics Committee Approval:** Ethics committee approval is not required in this study.

**Peer-review:** Externally peer-reviewed.

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