



PREVALENCE OF FECAL COLIFORMS AND ESCHERICHIA COLI O157: H7 ISOLATED IN TURKEY MEAT SOLD IN RABAT SUPERMARKETS (MOROCCO)

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ABSTRACT

Background: The Escherichia coli O157:H7 is responsible of foodborne infection from food origins and causes a serious syndromes of human health as diarrhea disease. The O157:H7 serotype is responsible of the epidemics in all the world which causes thousands of diseases and leaves dozens of deaths. **Objectif:** This work aims to evaluate the presence of pathogenic bacteria: Fecal coliforms and Escherichia coli O157:H7 in products made from turkey meat using a microbiological method. **Methods:** In the framework of this study 91 samples of four preparations of turkey meat: spicy minced meat, not spicy minced meat, spicy sausages and not spicy sausages were collected from four supermarkets in Rabat to evaluate the prevalence of Escherichia Coli O157:H7. Moreover, fecal coliforms were counted using VRBL agar (Lactose Agar Billy crystal violet and neutral red) in order to evaluate the hygienic quality of these products. **Results:** as result the bacteria's identified as pathogens and responsible of food poisoning, while all food have an animal origin. The fecal coliform count shows that 34% of the samples have a no satisfactory hygienic quality and ,58% have acceptable hygienic quality and 8% have a satisfactory hygienic quality. However, the prevalence of Escherichia Coli O157:H7 is 2%. These results spotlight the bad microbiological quality of the raw materials used to prepare the meat products, as well as the contamination in the abattoirs and workshops. **Conclusion:** The existence of strains Escherichia Coli O157:H7 potentially pathogens has been confirmed in turkey meat, which triggered a serious public health problem.

Keywords: *Escherichia Coli O157:H7; prevalence; turkey meat; fecal coliform; microbiological quality; food hygiene.*

1. INTRODUCTION

The microbiological safety of food has become the primary concern of consumers. The increasing incidence of diseases caused by microorganisms transmitted mainly by food, such as Salmonella, E.Coli O157: H7 and Listeria were reported in many countries and are a real health problem.

Meat and meat products are still considered as an essential component of the healthy diet due to its rich nutritional composition [1]. However, this rich in nutrients also causes the growth of many microorganisms including food pathogenic bacteria [2]. This process is highly favored in the summer, and associated with poor hygiene practices which contribute to the prevalence of disease and increases the risk of food poisoning [3].

A Foodborne disease is responsible for the most frequent health crises in the world. The main food vehicle and is suspected or confirmed in 80% of homes is the meat and especially meat of poultry is considered as a germ of foodborne disease [4]. In the world 1.8 million people die each year from diarrheal diseases where most of its could be attributed to food or water contamination [5]. In Morocco over the last two decades (1992-2011), 19625 cases of foodborne disease have identified of which 5688 hospitalizations and 221 deaths [6].

These illnesses are a result of ingesting invasive effect in bacteria, often toxigenic, and then cause lesions both in the gastrointestinal tract at the level of other tissues (sepsis). Eg Salmonella, Shigella, Listeria, Escherichia, Clostridium, Yersinia, Vibrio.

2. MATERIALS AND METHODS

2.1 Sampling

2.1.1 Number and origin of samples: in this study a total of 91 samples from four preparations of turkey meat: not spicy minced meat, spicy minced meat, not spicy sausages and spicy sausages have used which were collected during the period between 02-03-2015 and 03-05-2015 from four supermarkets in Rabat A, B, C and D.

2.1.2 Sampling Protocol: The Sampling was done weekly by taking random samples from each site including four spicy ground meat, not spicy minced meat, spicy sausages and not spicy sausages. The samples were immediately placed in a sterile plastic bag and then in a cooler at 4° C, and then transported to the laboratory of Microbiology and Food Hygiene for microbiological analyzes.

2.2 Microbiological analyzes

2.2.1 Sample Preparation: It's carried out for each sample weighing 25 g in a sterile bag. The weighing is then diluted in 100 ml of buffered peptone water (BPW). Suspensions of each sachet were then homogenized using a paddle mill (Stomacher) to obtain two parent suspensions title 1/5.

The method used is based on the fact that bacteria, placed on a favorable solid medium VRBL on agar (Lactose Agar Billy crystal violet and neutral red), will give birth to a macroscopically visible colony. The development of most bacteria not belonging to the Enterobacteriaceae family is inhibited by crystal violet and bile salts.

Only the colonies which appear red from 0.5 to 1 mm in diameter are taken into consideration and counted. Where the meat dishes contains between 30 and 300 colonies. the results are calculated using colony formula as shown below and expressed in units per gram of product (CFU / g).

$$\text{Fecal coliforms in CFU/g} = \text{number of colonies per plate} * 10 * \text{dilution factor} * 5 \tag{1}$$

Sampling conducted for this work has helped to distinguish 91 samples of four preparations of turkey meat (spicy minced meat, not spicy minced meat, spicy sausages and not spicy sausages) collected from four supermarkets in Rabat. The graph below presents the number of samples for each the categories of meat (Figure 1) and samples sites (Figure 2).

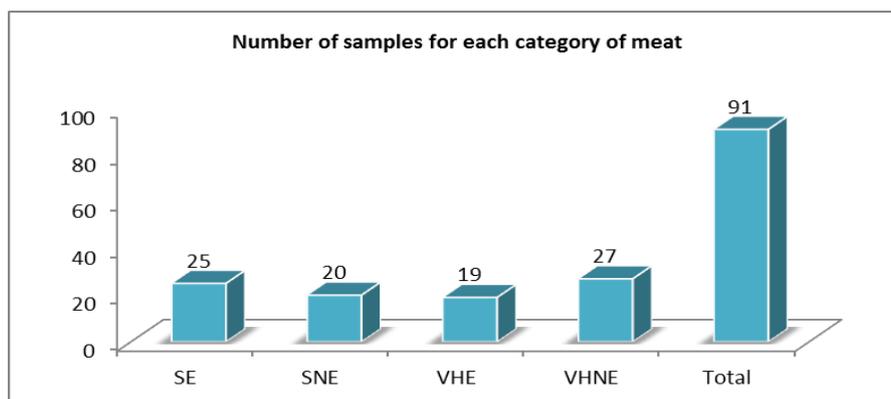


Figure 1: The figure presents distribution of samples according to the categories of meat. **SE:** spicy sausages; **SNE:** not spicy sausages; **VHE:** spicy minced meat; **VHNE:** not spicy minced meat.

25 samples of spicy sausage were collected which consists of 27.47%, 20 samples of non-spicy sausages or 21.98%, 19 samples of spiced minced meat are 20.88% and 27 samples of non-spicy minced meat or a percentage of 29.67%.

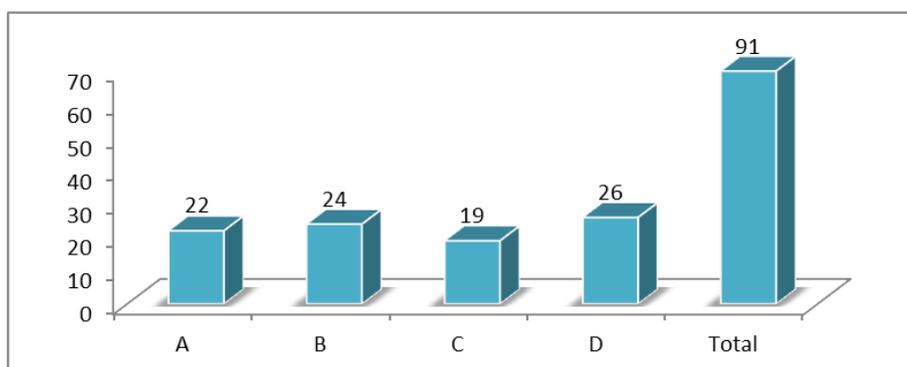


Figure 2: The figure presents distribution of samples based on the studied sites.

22 samples were taken from site A (24%), 24 samples from Site B (26%), 19 samples from Site C (21%) and 26 samples from Site D (29%). In order to get heterogeneous in the representative samples.

3. RESULTS

3.1. Enumeration of fecal coliforms

3.1.1. Distribution of fecal coliform contamination on all samples: The presence of fecal coliform rather thermo tolerant generally a sign of fecal contamination, and may be an indication of the presence of enteric pathogens [7].

Based on the Moroccan regulation on microbiological criteria, samples studied can be classes in 3 groups according to their hygienic quality levels of contamination by fecal coliforms: satisfactory, acceptable quality and unsatisfactory quality.

The results expressed in percentage of contamination by fecal coliforms in each sample site and for each category of meat are shown in Table 1:

Analysis of the data in Table 1 allowed us to classify the samples in three classes of hygienic quality (Figure 3).

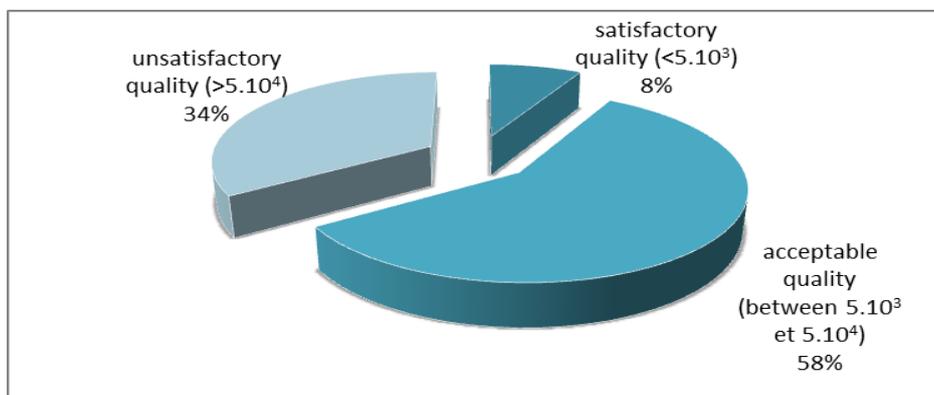


Figure 3: The figure showed hygienic quality classes of samples analyzed according to their Contamination by fecal coliforms expressed in CFU/g.

The contamination levels in the samples vary between a minimum value of 2.103UFC / g and a maximum value of 6.106UFC / g. The average value of the contamination by fecal coliforms on all samples is 6.105UFC / g.

From all the samples examined 34% were contaminated by fecal coliforms and content more than 5.104UFC / g so they have an unsatisfactory hygienic quality, 58% content from 5103 to 5.104UFC / g so they have an acceptable hygienic quality, of the samples that are consistent and do not exceed 5.103UFC / g, so they have a satisfactory hygienic quality.

3.1.2. Repartition of fecal coliform contamination based on meat categories:

Table 1: the table showed the hygienic quality classes of the samples analyzed according to their contamination by fecal coliforms based on the categories of meat CFU/g.

Quality	satisfactory sanitary quality (<math>< 5.10^3</math> UFC/g)	Acceptable hygienic quality: (between 5.10^3 et 5.10^4 UFC/g)	Unsatisfactory hygienic quality (>5.10^4 UFC/g)
SMM	5%	69%	26%
NSMM	8%	48%	44%
SS	12%	56%	32%
NSS	5%	65%	30%

SS: spicy sausages; NSS: not spicy sausages; SMM: spicy minced meat; NSMM: not spicy minced meat.

The data in Table 1 is illustrated in Figure 4:

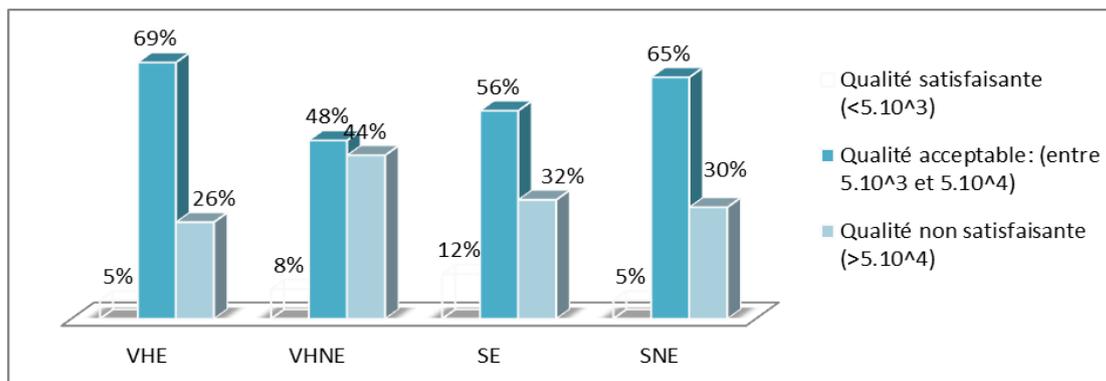


Figure 4: The figure showed the hygienic quality classes of the samples analyzed according to their contamination by fecal coliforms based on the categories of meat CFU/g.

Legend: SE: spicy sausage; SNE: not spicy sausage; VHE: spicy minced meat; VHNE: not spicy minced meat.

Of all the samples examined, 44% of samples of not spicy minced meat, 32% of samples of spicy sausages, 30% of samples of not spicy sausages and 26% of samples of spiced minced meat have a coliform contamination fecal greater than 5104 CFU / g and therefore an unsatisfactory hygienic quality.

3.1.3. Repartition of fecal coliform contamination based on sampling sites:

Table 2: The hygienic quality classes of the samples analyzed according to their Contamination by fecal coliforms based on sampling sites in CFU/g.

Quality sampling site	satisfactory sanitary quality (<5.103 UFC/g)	acceptable hygienic quality: (between 5.103 et 5.104 UFC/g)	unsatisfactory hygienic quality (>5.104 UFC/g)
A	9%	59%	32%
B	0%	62,5%	37,5%
C	11%	63%	26%
D	12%	50%	38%

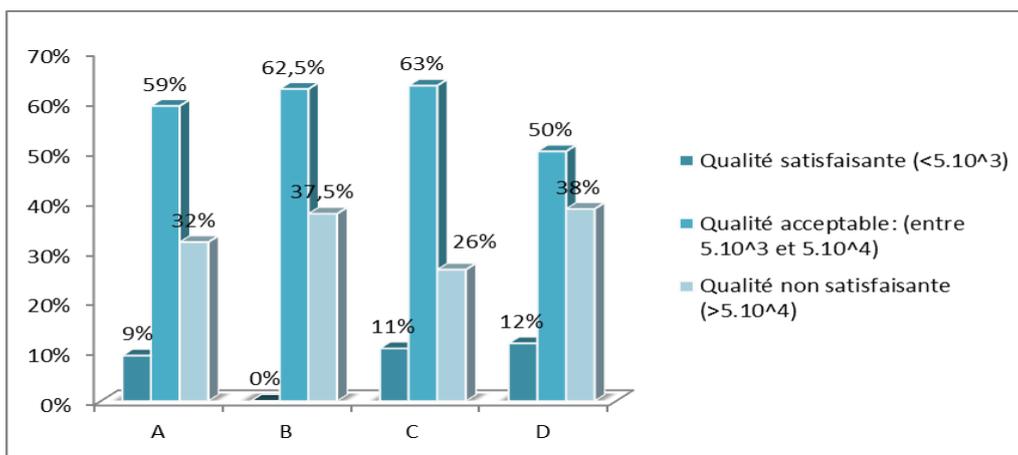


Figure 5: the figure presents classes of the hygienic quality of the samples analyzed according to their fecal coliform contamination based on sampling sites in CFU/g.

The sampling collected from the site D represents most of the products that have an hygienic quality not satisfactory with a rate of 38%, followed by the Site B with a rate of 37.5%, the site A has a rate of 32% and the Site C with 26% of non-compliant products. The four sites represent similar percentages, regards the products of hygienic quality acceptable, the sites C, B, A and D have respectively 63%, 62.5%, 59% and 50%.

We have found a low dominance on the four sites, products of hygienic quality satisfactory, the sites C and A have respectively 12%, 11%, and 9% of compliant products while the site B presents no product Conformat.

In general, we have found that there are a dominance of products with acceptable hygienic quality in studied four sites.

3.1.4. Detection of Escherichia coli O157: H7/:

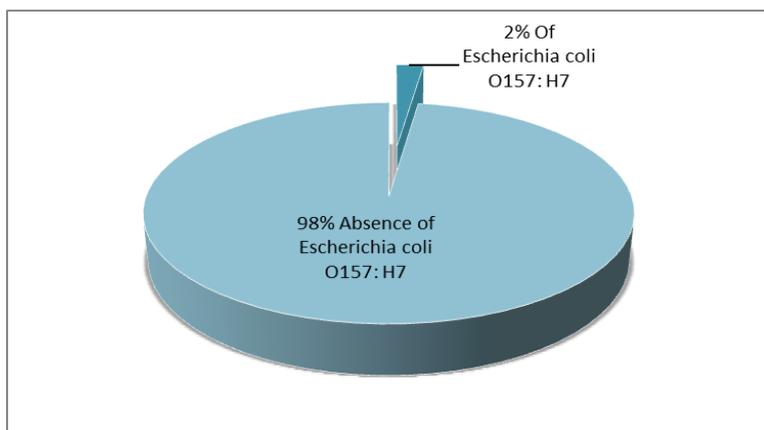


Figure 6: The figure presents prevalence of Escherichia coli O157: H7 on all samples.

Table 3: The table showed characteristics of Escherichia coli O157: H7 isolated.

Sampling site	Sample category	Fecal coliform CFU / g	Escherichia coli O157: H7
A	SS	2E+	Presence
A	NSMM	3E+06	Presence

Legend: **SS**: spicy sausages; **NSMM**: not spicy minced meat.

A total of 70 isolated Escherichia coli strains on EMB medium and showed a metallic green reflection, only 2 strains showed agglutination with O157 and H7 sera.

The prevalence of Escherichia coli O157: H7 in this study was 2% (2/91) (Figure 6), while 98% (89/91) of the samples showed no agglutination with O157 and H7 sera. Both stem from the same site A and two turkey meat categories: spicy sausages and non-spicy minced meat.

4. DISCUSSION

The summary of our results showed overall not very high safety, given that 90% of samples analyzed are considered unfit for human consumption, according to Moroccan law.

The count of fecal coliform gives an idea about the quantitative contamination of the products analyzed. In this, study 34% of products analyzed were unsatisfactory hygienic quality, this percentage is lower compared to the study of Cohen, (2007) in Morocco on chicken meat which showed that 48.4% of samples were unacceptable hygienic quality [8].

Our results are also lower compared with those of Tran, et THI, (2004) in Hanoi who found that 68.75% of different products of animal origin have an unacceptable hygienic quality, and also of those obtained by EL Allaoui, (2013) [9] [10].

In addition, it was found that 58% of products analyzed were acceptable hygienic quality; this rate is high compared to the results optened by EL Allaoui, (2013) in the Meknes supermarkets [10].

This high rate of the fecal coliform contamination can be explained by a high rate of initial contamination in slaughterhouses, or by additional contamination during the preparation of these products. These infections are often due to either the use of a raw material of poor quality, the unhygienic conditions of carriage, or failure to comply with hygiene rules when preparing or contamination by staff, tools and work surfaces and breaking the refrigeration of prepared products.

Regarding the category of the most contaminated meat fecal coliform, corresponds to the not spicy minced meat with a percentage of 44%, this may be due to the action of spices that play a very important role in decreasing the water activity and inhibiting the growth of microorganisms, especially they contain salt. While spicy ground meat is the category representing less contamination by fecal coliforms with a percentage of 23%.

In comparing the sampling sites, we find that the site D has more unsatisfactory product quality (38%) which can be explained by the following non-compliance with hygiene by staff, the contamination by improperly cleaned equipment or the poor quality of the raw material.

In addition, the detection of Escherichia coli O157: H7 was 2%. Moreover, Aouni, (2006) reported a prevalence of 4% and 14% in sausages and minced meat respectively [11]. In same optic Tebani, (2009) revealed a prevalence of 16.7% and 33.3% in minced meat and large intestine respectively [12].

The two strains isolated in this study come from the site A, turkey meat category: spicy sausages and not spicy minced meat. This may be due to cross-contamination among samples since they were taken from the same site.

5. CONCLUSION

The consumption of contaminated meat products with coliform and Escherichia coli O157: H7 is a significant health risk faced by consumers, and should be treated as a serious public health problem.

In Morocco, the incidence of disease caused by these pathogens is underestimated hence the importance of studies to quantify its presence in food of animal origin and clarify the extent of the problem caused by these germs and its impact on the public health risk in the country.

In this study, 91 samples have taken from turkey meat products, and results showed that 90% are unsuitable for microbiology point of view of consumption.

The results for the enumeration of fecal coliforms showed that 34% of the samples were non-compliant. However, the prevalence of *Escherichia. Coli* O157: H7 is 2%.

Based on these results, we can say that the products examined along this study have poor hygienic quality and may be a potential risk to consumers.

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Conflict of interest

The authors declare no conflicts of interest.

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