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Microbiological Study of Turkey Meat Marketed in Kenitra (North-oust of Morocco)

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Abstract

Turkey meat is considered among meat of poultry most consumed by the Moroccan population, and this due to its richness in vitamins, proteins and essential elements for growth, which gives him an excellent nutritional value. In addition, it is characterized by a low content of fatty acids. But this highly perishable commodity could constitute a real danger of food-borne illness. The aim of present study is to assess the hygienic quality of 168 samples collected from twenty four points of sales located in six neighborhoods in Kenitra. The analysis of the microbiological quality of the samples shows that the percentage of non-compliance for the parameter Total Aerobic Mesophilic Flora is 35%, with the mean concentration 7.21 (log_{10} cfu/g). The respective percentages of non-compliance of the total and fecal coliforms are 68 and 75% with bacterial concentrations 6.69 (log_{10} cfu/g) and 6.42 (log_{10} cfu/g). This non-compliance is due to the presence of the germs of fecal contamination among the identified isolates. The four identified species showed following prevalence: *Escherichia coli* (67.9%), *Klebsiella pneumonia* (17, 9%), *Pseudomonas* sp. (14.1%) and *Salmonella* (0%).

Keywords: Turkey meat; Kenitra; Non-compliance; Bacterial concentration; Contamination

Introduction

In Morocco, the production of turkey meat has experienced a pace of significant development, it is increased from 70,000 tones in 2010 to 80,000 tones in 2015 and its consumption is becoming more and more important given its price suitable for the citizen [1].

Turkey meat is considered among the most consumed poultry meats in the city of Kenitra knowing that the most requested body part by the citizen is the muscle. It is regarded as a potential source of food-borne illness caused by lack of hygiene [2,3]. Turkey meat is a highly perishable food whose hygienic quality depends on the quality of the different operations including the slaughter, cutting, storage and distribution [4]. In Morocco, between 2000 and 2004, 7,118 cases of food-borne infections have been reported in which more than 86% are of bacterial origin [5]. The contamination of foods of animal origin and mainly of meat products is responsible for 28% of cases of food-borne diseases [6]. Many factors have contributed to the increase of foodborne diseases. Mass production, environmental factors, and inadequate knowledge on food handling have contributed to the increase of contaminated foodstuffs [5].

This problem of contamination, therefore, proves the importance of the food hygiene. This work constitutes a contribution to the evaluation of the hygienic quality of turkey meat marketed in Kenitra city.

Materials and Methods

Sample collection

A total of 168 samples of turkey meat have been collected, in aseptic conditions, from 24 points of sale in the city of Kenitra to reason of seven samples by the point of wind, during the period of June-September 2015. Each sample was placed in sterile stomacher bag and transported in a cooler to the laboratory. In the arrival, the samples are analyzed immediately in three repetitions.

Bacteriological analysis

A portion of 25 g of each sample is cut aseptically and then crushed carefully using a chopper ultra-turax and then, added in an Erlenmeyer flask containing 225 ml of sterile physiological water. From this last, we have prepared a series of tenfold dilutions ranging up to 10^{-6} [7]. The bacteriological analysis has focused on the following germs: the Total Aerobic Mesophilic Flora (TAMF), the Total and Fecal coliforms, *Escherichia coli, Klebsiella* sp., *Pseudomonas* sp. *and Salmonella* sp.

Enumeration of the Total Aerobic Mesophilic Flora (TAMF): The enumeration of the TAMF was carried out by using the method of seeding in depth on the Plate Count Agar (PCA, Biokar). The series of tenfold dilution were deposited aseptically in different boxes of sterile Petri dishes containing 10-15 ml of the medium PCA maintained at 45 ± 1°C. The incubation was driving at 30°C for 72 h. Only the boxes of Petri dishes with a number of colonies between 30 and 300 have been retained for the enumeration [8].

Enumeration of total and fecal coliforms: The Enumeration of Total and Fecal coliforms was carried out on Violet Red Bile Lactose Agar (VRBL). Total coliforms are counted after 24 h of incubation at 37°C. For Fecal coliforms, the boxes of Petri dishes are incubated at 44°C for 24 h [9]. Only colonies with a yellow aspect orange and a diameter of more than 0.5 mm were counted.

Escherichia coli: Colonies of Fecal coliforms were isolated in the Middle VRBL and subcultured on another selective medium Eosin Methylene Blue (EMB). Then, the boxes of Petri dishes have incubated at 37°C for 24 h. The presence of *Escherichia coli (E. coli)* is indicated by the metallic reflection and the green color of the Colonies [10].

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***Klebsiella* sp.:** Similarly, colonies of Fecal coliforms were isolated from the mid-VRBL and sown on the mid-Eosin Methylene Blue (EMB), after their incubation at 37°C, for 48 h [10]. The *Klebsiella* colonies have shown large, rounded and very mucous membranes [11].

***Pseudomonas* sp.:** The presence of colonies of *Pseudomonas* has been carried out on the mid-Pseudomonas Agar Base, after incubation boxes of Petri dishes to 44°C for 48 h [12]. The *Pseudomonas* is characterized by the yellow color with a clear halo.

***Salmonella*:** The research of *Salmonella* requires the achievement of several of the systematic steps. The pre-enrichment was carried out in 225 ml of buffered peptone water (Difco) incubated at 37°C for 24 h, followed by an enrichment in the selective liquid medium: Rappaport Vassiliadis (RV). The high concentration of magnesium chloride and the presence of the green malachite are slowing down the growth of microorganisms except salmonella. A volume of 0.1 ml of the medium pre-enriched was added to 10 ml of the medium RV and incubated at + 41°C ± 2°C, for 24 h. The isolation was carried out on the mid-Hektoen (Difco). The selective enriched liquid medium RV was stocked on the selective agar medium Hecktoen, previously sunk in boxes of Petri dishes, and were incubated at +36°C ± 2°C, for 24 h [13,14].

Biochemical identification

All above-cited bacteria have been tested by Gram staining and by oxidase activity. The identification of the isolates was carried out by the determination of the biochemical profile using the API gallery 20 (Bio Mérieux, Marcy l'Etoile, France).

Results and Discussion

The non-conformity of samples was assessed according to the joint order of the Ministry of Agriculture and Rural Development, the Ministry of Health and the Ministry of Industry, Trade and Telecommunications No. 624-04 Safar 1425 (8 April 2004) published in the Official Bulletin n: 5214-30 Rabbi 1-1425 (20-5-2004) relating to microbiological standards and which must meet the animal foodstuffs. The acceptable upper limit of the Total Aerobic Mesophilic Flora (TAMF) in the meat of poultry is of the order of 6.7 ($\log_{10}$ cfu/g), 2 ($\log_{10}$ cfu/g) for Total coliforms and 4 ($\log_{10}$ cfu/g) for Fecal coliforms. However, the Moroccan standards are devoid of security criteria concerning *Escherichia coli, Klebsiella pneumonia* and *Pseudomonas* sp. Therefore, the comparison of our results was made in reference to other studies. For *Salmonella* and according to the Moroccan standard, it must be absent in 25 g of the sample of the meat [13].

Percentage of the non-compliance of the studied samples

Total aerobic mesophilic flora: The results of the microbiological analysis of turkey meat samples marketed in the city of Kenitra (Table 1) show that the average value of the Total Aerobic mesophilic flora (FMAT) 6.44 ($\log_{10}$ cfu/g) is less than the value declared by the Moroccan standards of the microbiological safety criteria of foods (6.7 ($\log_{10}$ cfu/g)). These results are lower than those of Amara et al. [14] and Cohen et al. [15], the found values are respectively in the order of 7.15 and 7.4 ($\log_{10}$ cfu/g). The values of our results are lower also to those found in the meat of poultry by Chaiba and Rhazi Filali [16] (7.45 ($\log_{10}$ cfu/g)). The non-compliance of the studied samples has presented a rate of 35% (Figure 1).

Total coliforms: The degree of contamination of the samples by the total germs has been assessed by the calculation of the average of the loads found in the analyzed samples, the load of Total coliforms is 3.87 ($\log_{10}$ cfu/g). This value is lower than that found in the carcasses of chickens of the traditional slaughter of Meknes city; the abundance of Total coliforms was 4.4 ($\log_{10}$ cfu/g) [16]. In our study, it was found that 68% of the analyzed samples are non-complaint (Figure 2).

Fecal coliforms: The average rate of contamination of turkey meat samples is 5.98 ($\log_{10}$ cfu/g). In this study, the load is less than these mentioned by El-Allaoui et al. [13] (8.15 ($\log_{10}$ cfu/g)), and higher than these reported by Cohen in 2006 (2.5 ± 1.2 ($\log_{10}$ cfu/g)) [5]. Our results revealed a rate of non-compliance of 75% of the totality of the analyzed samples (Figure 3). This contamination would seem to be obvious during the evisceration although the majority of these germs can be caused the gastro-enteritis disorders for consumer.

Origin of the non-compliance

On 168 samples, 118 bacterial strains were isolated (Table 2). The results of the microscopic study and the biochemical identification of bacteria responsible for the non-compliance of turkey meat show that coliform bacteria are responsible for the non-compliance of the majority of the analyzed samples.

***Escherichia coli*:** The prevalence of *Escherichia coli* is 67.8%. This value is compatible with that found by Patyal et al. (68%) [17], it is lower than that found by El-Allaoui et al. (83%) [18], and largely superior to that found by Gupta (22.5%) [19] and Iroha (2%) [20]. *E. coli* is a hygienic indicator, its presence in turkey meat is a sign of enteric pathogenicity and which may cause a risk of contamination to the consumer. It was found in abundance. This can be explained by the

Points of sale	TAMF ($\log_{10}$ cfu/g)	CT ($\log_{10}$ cfu/g)	CF ($\log_{10}$ cfu/g)
1	6.25	2.65	6.33
2	6.48	3.57	6.33
3	6.46	3.14	6.01
4	6.52	3.15	5.92
5	6.45	3.20	5.62
6	6.33	3.02	5.40
7	6.28	3.29	5.34
8	6.28	3.65	5.38
9	6.40	3.85	5.34
10	6.41	4.02	5.53
11	6.51	3.98	5.71
12	6.41	4.11	5.78
13	6.51	4.53	6.13
14	6.53	4.69	6.19
15	6.47	5.30	6.40
16	6.57	5.40	6.49
17	6.46	4.97	6.52
18	6.48	4.92	6.49
19	6.50	4.21	6.36
20	6.40	3.57	6.03
21	6.48	3.45	5.98
22	6.49	3.22	5.89
23	6.45	3.40	5.97
24	6.50	3.67	6.27
Average	6.44	3.87	5.98
Standard*	6.7	2	4

C.T: Total Coliforms; C.F: Fecal Coliforms; TAMF: Total Aerobic Mesophilic Flora. *According to the joint order of the Ministry of Agriculture and Rural Development, the Ministry of Health and the Ministry of Industry, Trade and Télécommunications No. 624-04 Safar 1425 (8 April 2004) published in the Official Bulletin n: 5214-30 Rabbi 1-1425 (20-5-2004) relating to microbiological criteria of use for food.

Table 1: Result of microbiological analyzes of samples collected from 24 points of sale.

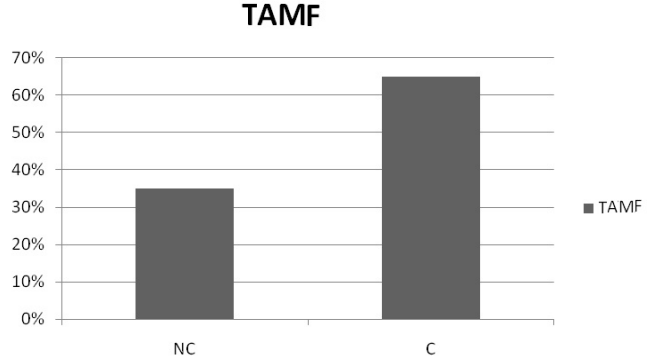

Figure 1: Percentage of non-compliance of the analyzed samples for the FMAT. NC: Non-Compliant; C: Compliant.

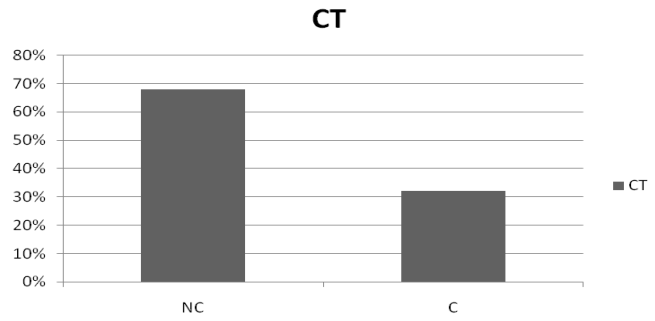

Figure 2: Percentage of non-compliance of the analyzed samples for total coliforms. NC: Non-Compliant; C: Compliant.

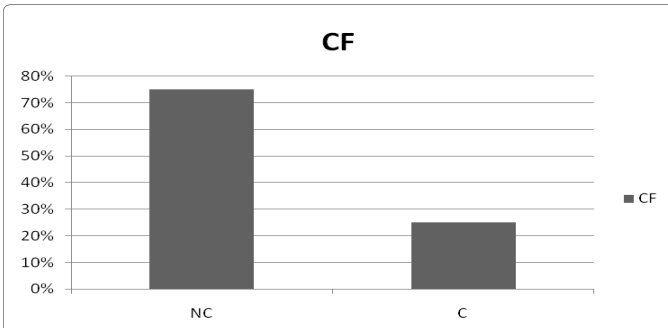

Figure 3: Percentage of non-compliance of the analyzed samples for Fecal coliforms. NC: Non-Compliant; C: Compliant.

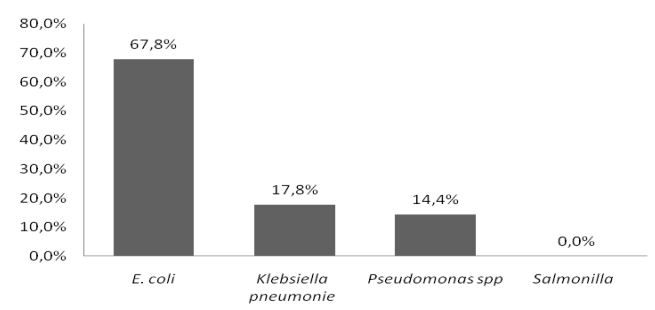

Figure 4: Percentages of germs responsible for the non-compliance of the samples of turkey meat.

lack of good hygienic practices among the personnel responsible for the sale or in their premises during the process of slaughter and cutting of turkey meat (Figure 4).

***Klebsiella pneumonia*:** The prevalence of *Klebsiella pneumonia* was 17.8%. This rate is far lower than that found in the turkey meat by Davis (38%) [21], and by Neslihan in the chicken meat (47%) [22]. The presence of *Klebsiella pneumonia* in meat is an indicator of faecal contamination and a sign of poor conditions of hygiene [23,24].

***Pseudomonas* sp.:** For *Pseudomonas* sp. the prevalence was 14.4%. This value is very lower than that found by Arnaut-Rollier in chicken meat (19.2%) [25]. The presence of this bacterium is probably due to its prototrophic properties and to these needs in mineral nitrogen, carbohydrates, mineral salts but it does not require the presence of the growth factors as well as its psychrotroph characteristic which the growth temperature varies between 0 and 37°C [26,27]. Therefore, its presence in the analyzed samples denotes a secondary contamination which could be associated to a bad conservation in the time [28] and the modification of the organoleptic characteristics of the meat.

***Salmonella* sp.:** The *Salmonella* presents an importance in the veterinarian areas and on the medical plan, by the economic losses caused by animal diseases and by the high incidence in the consumer. Therefore, the typhoid fever and food-borne diseases due to *Salmonella* [29] provoke most often the gastro-enteritis. *Salmonella* is involved in 76.2% of food-borne illness. Given the broad spectrum of animals that can be carriers of *Salmonella*, a great variety of food products may be at the origin of a human contamination: meat and particularly poultry, meat products, eggs and dairy products. Salmonellosis can give rise to important spots which can reach a national scale if a food marketed to wide dissemination is contaminated.

In this study, the rate contamination of turkey meat by *Salmonella* is zero (0%), although the contamination of faecal origin has been relatively important. Indeed, a high load in fecal coliforms can predict the presence of salmonella.

The rate of contamination by *Salmonella* is variable according to studies. In Morocco, Bennani et al. [30] have reported a rate of 13% of the positive samples to the presence of Salmonella sp on samples of poultry meat in Fez [30]. In Casablanca, Karraouan et al. have found a contamination rate of 20.3% in Turkey meat [31]. Chaiba [32] reported a rate of 20.83% in chicken meat in Meknes. However, Beli et al. have revealed a low prevalence of *Salmonella* in turkey meat in Albania (8.2%) [33]. In Ireland, Jordan and al have found a rate of 3.1% [34] and in the United Kingdom, Zhao et al. have found a rate of 5.6% [35].

The rate of Isolation of Salmonella is higher after evisceration, which confirms that the operations of slaughter and preparation in the slaughterhouse and cutting plants can be considered as a major source of contamination of the meat by *Salmonella*.

The presence of the pathogenic germs in the analyzed samples shows that it is interesting to determine the origin of contamination. This can be achieved only by the adoption and implementation of the quality approach in applying the guidelines of the Hazard Analysis Critical Control Point (HACCP) plan, as well as the mastery of the guide to good hygiene practices.

Conclusion

The results of analyses of 168 turkey meat samples, collected from 24 sale points in the city of Kenitra, show the presence of four bacterial strains: *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas* sp. *and*

Species	F	G	Ox	Cat	Glu	Lac	Cit	Ur	TDA	ONPG	Vp	H_2S	Ind	N.S.I
Escherichia coli	B	-	-	+	+	+	-	-	-	+	-	-	+	80
Klebsiella pneumonia	B	-	-	+	+	+	+	+	-	+	+	-	-	21
Pseudomonas sp.	B	-	+	+	-	-	+	-	-	-	-	-	-	17
Salmonella sp.	B	-	-	+	+	-	+	-	-	-	-	+	-	0
Total														118

F: Form, G: Gram, Ox: Oxidase; Cat: Catalase; Glu: Fermentation of Glucose; Lac: Fermentation Lactose; Cit: Citrate, Reduction of Nitrates to Nitrites; Ur: Urease; TDA: Tryptophan Deaminase; ONPG: Ortho-Nitro-Phényl-Galactopyranoside; Vp: Voges-Proskauer; H_2S: Production of the Gas; Ind: Training of Indole; N.S.I: Number of Isolated Strains.

Table 2: Biochemical characteristics of the Strains *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas* sp., and *Salmonella* sp. isolated of turkey meat costs.

Salmonella. The *E. coli* strain was found in all points of sale with a total dominance (67.8%), followed by *K. pneumonia* with a prevalence of 17.8%, then *Pseudomonas* sp. with a rate of 14.4%. While the *Salmonella* has been absent.

These results suggest that there may be a lack of strategies of adequate control of turkey meat. Such inadequate controls can include the absence of good hygiene practices during the slaughter, transport, storage and distribution.

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