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Full Length Research Paper

Survey of *Yersinia enterocolitica* contamination in distributed broiler meats in Tabriz City, Iran

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Yersinia enterocolitica **is one of the pathogens of the intestine, which is transmitted to humans through water and food materials, especially contaminated meats and dairy products. The infected meats have no apparent symptoms of putrefaction and eating of such meats in raw or undercooked forms can lead to gastroenteritis, dysentery, mesenteric lymphadenitis, or even death. The aim of this research is to survey the frequency of *Y. enterocolitica* in different types of broiler meats and compare their ratio of infection in Tabriz City. To carry out this study, 120 samples of broiler meats were collected by random sampling. The samples were divided into groups corresponding to the parameters of packaging, no packaging, variety of preservation, and storing time. All of the samples were carried to the laboratory aseptically in cold boxes and the required microbiological tests (culture, staining, and biochemical tests) were performed. 15.8% of all of the studied broiler meats were contaminated with *Y. enterocolitica*. The analysis of results with fisher exact and chi- square tests showed that there was no meaningful diversity among the all the studied samples groups. A considerable quantity of broiler meats was contaminated with this bacterium without any symptoms of infection. If meat products are not cooked well, the preservation of such food materials in refrigerator provides an opportunity for their proliferation; regarding the absence of any symptoms, eating of such foods can cause gastroenteritis and other digestive disorders in consumers.**

Key words: *Yersinia enterocolitica*, broiler meats, contamination, Tabriz.

INTRODUCTION

Yersinia enterocolitica is a Gram negative bacterium and an intestinal pathogen which is transmitted to man through water and foodstuffs. Its epidemics often emerged after consumption of contaminated foodstuffs; particularly contaminated meat (Belgian, 2004). The place of this bacterium in nature is soil, waste water and various types of fresh water. Moreover, the most important contaminated foodstuffs with *Y. enterocolitica* bacteria that have ever been studied are related to different types of packaged and unpackaged or vacuumed red meat, milk, eggs, types of poultry meat, vegetable and edible shell fish (Andersten et al., 1991; Hanna et al., 1985).

In general, human beings were contaminated with *Y. enterocolitica* bacteria through eating contaminated foodstuffs and also a great deal of serological research showed that individuals who concerned with livestock and

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meat are more prone to be contaminated with *Y. enterocolitica* bacteria than others (Nimfa, 1999). In this light, one influential study was carried out in France in order to investigate the seroepidemiology of *Y. enterocolitica*; the result revealed that most individuals are contaminated with bacteria through consumption of the contaminated meat of such bacteria in this country (Gourdon et al., 1999).

In addition, the untouched tissues of healthy animals are free of microbial agents, but when they are slaughtered, the bacteria living in their digestive system, the environment of slaughterhouse, and the process of preparing meat resulted in contamination of meat (Ellen, 2002).

Most bacteria can resist and survive in normal temperature of refrigerators (+4℃), however, the growth and replication of a large number of bacteria in the above-mentioned temperature stopped, more generally, microbial load of fresh meat did not increase as being kept in refrigerators (Ansay et al., 1999). Nevertheless, some psychrophilic bacteria can easily grow and replicate in normal refrigerators. Of these bacteria, we can mainly mention pathogen.

Among several pathogen bacteria which have the power of replication on meat in refrigerator temperature, *Y. enterocolitica* bacterium is of particular importance for two reasons: first, due to the ability of growth, replication and production of Enterotoxin (Nimfa, 1999) on kept meat in refrigerators without changing smell and taste of meat, and second, due to causing serious diseases in consumers of such contaminated meat such as Gastroenteritis, Dysentery, Mesenteric Lymphadenitis, Septicemia, Erythema nodosum, Arthritis, Autoimmune thyroid disease and even death in some cases (Bostan, 2001; Chynoweth et al., 1998; Hasani and Firouzi, 2005; Moore and Madden, 2000; Razavilar, 2003).

Furthermore, meat is a very rich foodstuff that plays an important role in nutritional needs of man. In other words, the nutritional value of meat provides essential proteins of human body, since it contains vital amino acids, mineral salts and remarkable vitamins such as iron, copper, zinc and vitamin B12, thus it has a peculiar place among consumable foodstuffs of man.

From this perspective, a wide range of studies all over the world have focused on microbial contaminations and infectious diseases transmitted through consumption of contaminated meat. One of these microbial contaminations pertains to *Y. enterocolitica*.

Healthy nutrition is a major aspect of health and hygiene of man. However, due to lack of appropriate solutions in order to permanently test consumable meat of man, it seems that we should take necessary measures for characterization of contamination sources and control the infection problems by means of controlling contamination in meat. The purpose of this study was to survey the *Y. enterocolitica* contamination in distributed broiler meats in Tabriz.

MATERIALS AND METHODS

A carcass as a unified sample in sterile boxes in a cold chain temperature (inside cold box) immediately was transferred to Microbiology Laboratory of Hygiene Group of Nutrition and Hygiene Faculty after sampling. In fact, in each case of sampling and in whole sampling, specific characterization of samples was recorded in accordance with questionnaire and then was attached to the sample box.

In this study, a total of 120 samples of chicken meat were bought from Tabriz City (North west of Iran) markets, out of which 30 samples were bought directly from slaughterhouses and packaging centers in order to investigate the resistance of chicken meat which is contaminated with *Y. enterocolitica*.

Our research sampling was taken from various chicken meat slaughterhouses and packaging companies in Tabriz. In order to examine the effect of slaughter environment and packaging on contamination of chicken meat with *Y. enterocolitica*, thus two major companies among many others were randomly selected and then 30 samples were randomly chosen per company among the selected samples and later the results of both groups were compared in detail.

As a matter of fact, there were two types of packaged chicken meat in Tabriz during the conduct of this study: the first one, with extensive consumption, contained full chicken meat packaged in plastic bags and the second one comprises of chicken meat separated into breast or thighs packaged in disposable containers with nylon cover. These two types of packaging were used in sampling so as to investigate and compare the contamination outbreaks.

It is worth noting that unpackaged chicken meat in Tabriz markets was those types of meat that were previously produced in the form of full chicken and packaged by producers. Then, sales agents in retails opened their packaging and separated them into specific parts of breast, thighs, wings and neck and thereby distributed to consumers. We made use of all these parts in our research sampling.

According to prescription of hygienic centers, meat producers should transfer chicken meat to retailing centers inside refrigerators to be distributed to consumers. Thus, in order to compare the frequency of contamination in chicken meat kept and distributed in refrigerators with types of meat kept out of refrigerators, 30 samples out of whole samples were related to those meat kept out of refrigerators.

Notably, in each sampling, complete specification of samples was recorded in related questionnaires and they were immediately transferred to Microbiology Laboratory of Hygiene Group of Nutrition and Hygiene Faculty in suitable sterile boxes under cold box temperature. More specifically, each sample after transferring to laboratory were separated into small parts by means of sterile scalpel and then the entire processes of growth, coloring and biochemical tests were carried out over meat to diagnose *Y. enterocolitica*.

Then, at the end of any test, the results were recorded in checklists and the final results concerning the contamination of samples with *Y. enterocolitica* bacterium were specified based on the recorded results of each process.

Next, the results of tests were recorded in checklists and based on this information, the species and the genera of bacteria were determined in samples. After gathering all research data and applying a statistical method of comparison of proportion of society in fixed numbers of zero, the amount of P value (P) and frequencies of bacterial contamination in different samples were calculated separately.

Finally, to find out whether the differences in the frequency of containment of various types of samples are significant or not, we made use of chi-square and Fisher's exact test.

Table 1. Frequency of these bacteria in packaged and unpackaged chicken meat and their prevalence.

Samples	Sample size	Percentage frequency of positive samples		Percentage frequency of negative samples	
		n	%	n	%
Packaged	60	8	13.3	52	86.6
Unpackaged	60	11	18.3	49	81.6
Total	120	19	15.8	101	84.2

Table 2. Frequency of bacteria in both packaged chicken meat and their prevalence.

Samples	Sample size	Percentage frequency of positive samples		Percentage frequency of negative samples	
		n	%	n	%
Packaged	30	6	20	24	80
Unpackaged	30	2	6.7	28	93.3
Total	60	8	13.3	52	86.7

Table 3. Frequency of bacteria in two major slaughter companies of chicken meat.

Samples	Sample size	Percentage frequency of positive samples		Percentage frequency of negative samples	
		n	%	n	%
Packaged	30	6	20	24	80
Unpackaged	30	5	16.6	25	83.4
Total	60	11	13.3	52	86.7

RESULTS

In this study, a total of 120 samples of meat, including 60 packaged and 60 unpackaged chicken meats were randomly bought from distribution centers, slaughter-houses and packaging centers in Tabriz. After testing and recording the results in checklists and conducting statistical tests, we obtained the following findings: First, out of 120 tested samples of chicken meat, 19 samples were contaminated with *Y. enterocolitica*, while 101 samples were not contaminated and accordingly, we found that 15.8% of samples were contaminated with the aforementioned bacteria. The results of chi-square test on research data showed that there was no significant difference between packaged chicken meat and unpack-aged one in terms of the frequency of contamination with *Y. enterocolitica* (*P>0˙05*). The frequency of these bacteria in packaged and unpackaged chicken meat and their prevalence were presented in Table 1.

With regard to two types of chicken packaging, that is, packaging of full chicken in plastic bags and packaging of separated chicken into breasts or thighs in disposable containers with nylon cover, we compared the frequency of bacteria based on Fisher's exact test and found that there was no significant difference between the two aforementioned types of packaging (that is, 60 packaged and 60 unpackaged chicken meats) in their contamination outbreaks (*P>0˙05*). Table 2 depicts the frequency of bacteria in both packaged chicken meat and their Prevalence in detail.

In order to find out the effect of the slaughter environment of poultry on contaminated chicken meat, that is, packaged and unpackaged types of chicken meat with *Y. enterocolitica*, we randomly selected our samples equally among packaged and unpackaged meat of two major slaughter companies among different companies and compared their results. The Fisher's test indicates that there was no significant difference between two companies in terms of the frequency of contamination of meat (*P>0.05*). These findings are displayed in Table 3.

To gain insights into the keeping manner of chicken meat, the comparison was made between the frequency of contamination in meat kept in refrigerators and meat kept out of refrigerators, the conducted tests on 30 samples which were randomly selected from each group and the chi-square test proved that there was no signi-ficant difference in the frequency of contamination in two groups of meat (*P>0˙05*). To better illustrate these findings, the results are presented in tangible way (Table 4).

In a total of 120 examined samples in our study, 30 samples were bought from freshly slaughtered and packaged chicken meat from slaughter houses and packaging companies in order to be compared with those 30 samples which were randomly selected from whole

Table 4. Frequency of bacteria in two types of chicken meat kept in refrigerators and out of refrigerators.

Samples	Sample size	Percentage frequency of positive samples		Percentage frequency of negative samples	
		n	%	n	%
Non refrigerated	30	7	23.3	23	76.7
Refrigerated	30	4	13.3	26	86.7
Total	60	11	18.3	49	81.7

Table 5. Frequency of bacteria in two types of freshly slaughtered and old chicken meat.

Samples	Sample size	Percentage frequency of positive samples		Percentage frequency of negative samples	
		n	%	n	%
Freshly slaughtered	30	2	6.6	28	95
Non freshly slaughtered	30	7	23.3	23	82
Total	60	11	15	51	84.2

sample of the study bought from Tabriz markets. The results of chi-square test showed that there was no significant difference between two groups ($P>0.05$). Table 5 shows more detailed information about the frequency and prevalence of the meat.

DISCUSSION

Taking into account the contamination of various types of chicken meat samples in Tabriz, the results estimated that 15.8% out of whole sample were contaminated with *Y. enterocolitica*. These results are in consistent with similar studies, for instance, Soltan et al. (2004) estimated the frequency of contaminated meat (14.5 %) in Tehran.

In the current research, the frequency of bacteria in packaged and unpackaged meat was estimated to be 13.3 and 18.3%, respectively, though, statistically through applying chi-square test, significant differences were not observed in the frequency of bacteria in these two groups of samples. However, we should keep in mind that since all produced meat in industrial slaughter houses was initially distributed to markets in packaging form and then meat was unpackaged in retails in order to be sold separately in the form of breasts and thighs of chicken. Thus, the increase of contamination in unpackaged meat can be derived from secondary source of contamination due to contamination in working place, workers' hands, cutting knife and so on. However, a distinct study should be conducted to thoroughly specify the effect of any aforementioned cases.

In comparison of two types of packaging meat in Tabriz markets: Full chicken packaged in nylon bags and separated parts of the chicken such as breasts and thighs packaged in disposable containers with nylon cover, the results of Fisher's exact test realized no significant difference in the frequency of contamination in two groups. However, 13.3% increases in contamination of meat samples in the first type of packaging was observed. The difference can be attributed to the washing quality and packaging of two types of aforementioned sampling in industrial slaughter houses which required to be explored in other studies.

In an attempt to make a comparison between the effect of the place of slaughter houses and different types of packaging on contamination of chicken meat, among different samples of companies, two major ones were randomly selected and then among the samples of two companies, 30 samples were randomly chosen per company. Their results were compared by chi-square test indicating that there was no significant difference in the frequency of bacteria among production of these two companies.

Unlike formal provisions in distribution of chicken meat, some salesmen (particularly in central markets of the city) distribute their meat out of refrigerators; it was observed that meat kept out of refrigerators had 10% high frequency of bacterial contamination than meat kept in refrigerators. Although, Fisher's exact test did not provide any statistical evidence on significant difference, it can be due to some manipulations such as opening packaging of chicken, accumulating a large number of chicken carcasses over each other and lack of hygiene in such distribution places.

Comparing the frequency of contamination in freshly slaughtered chicken meat and old one, it was found that the contamination frequency in samples of first group, though, not statistically remarkable compared to second group by applying chi-square test. The high frequency of contamination in old meat was indicative of enhancing the chance of meat contamination in packaging processes. To a large extent, this can be interpreted in terms of gathering healthy and contaminated chicken

over each other, contaminated working tables, and contaminated gloves of workers and also touching contaminated samples and ability of bacteria in growth and replication on kept meat in refrigerator temperature.

In sum, with reference to the findings of this study, *Y. enterocolitica* contamination of chicken meat in markets can lead to dangerous diseases like Food poisoning, Gastroenteritis and serious problems in Digestive system for consumers of such meat, more importantly, due to lack of sufficient internal temperature of cooking of meat especially in barbecue and also keeping these food stuffs in refrigerators which accelerates the growth and replication of bacteria. Therefore, it is suggested that in line with more comprehensive studies of diagnosing accurate ways of meat contamination and preventing them, we should inform people to cook meat appropriately to destroy bacteria and accordingly reduce diseases caused by consumption of contaminated meat.

REFERENCES

Andersten JK, Sorensen R, Glensbjerg M (1991). Aspects of the epidemiology of *Yersinia entrocolitica*, Intl. J. Food Microbiol., 13: 231-237.

Ansay SE, Dorrling KA, Kasper CW (1999). Survival of *Escherichia coli* O157 in ground-beef patties during Storage at 2, -2, 15 and then -2℃. J. Food Prot., 62: 1243-1247.

Belgian S (2004). The presence of *Yersinia entrocolitica* and other Yersinia species in ground beef in Aydin Turkey, Turkish J. Vet. Anim. Sci., 28: 489-495.

Bostan K (2001). Effects of cooking and cold storage on the survival of Campylobacter jejune in meat, Arch. Lebensmittelhyg, 52: 28-30.

Chynoweth RW, Handson JA, Thom K (1998). Aerobic growth and survival of *Campylobacter jejune* in food and stream water, Lett. Appl. Microbiol., 27: 341-344.

Ellen DM (2002). Survival and growth of bacterial pathogens on raw meat during chilling, Food Res. Inst. Univ. Wisconsin-Madison, pp. 1-4.

Gourdon F, Beytout J, Reynaud A (1999). Human and animal epidemic of *Yersinia entrocolitica* O: 9, Emerg. Infect. Dis., (5)5: 719-721.

Hanna MO, Zink ZL, Carpenter ZL, Vanderzant C (1985). *Yersinia entrocolitica* like organisms isolated from vacuum-packaged beef and cuts, J. Appl. Microbiol., 46:493-499.

Hasani TAM, Firouzi R (2005). Animal disease due to bacteria. Tehran University press. 2nd edition. pp. 268.

Moore JE, Madden RH (2000). Survival of *campylobacter coli* in porcine liver. Food Microbiol., 18:1-10.

Nimfa MS (1999). Seroepidemiologic study on the occurrence of antibodies against *Yersinia entrocolitica* and *Yersinia pseudotuberculosis* in urban and rural population of the Lubin region (Eastern Poland),. Ann. Agric. Environ. Med., 6:57-61.

Razavilar V (2003). Pathogenic microbes in food and epidemiology of food poisoning. Tehran university press. 2nd edition, pp. 96-98.

Soltan DM, Taromi M, Golkar F (2004). Survey of *Yersinia enterocolitica* contamination in distributed broiler and red meats in Tehran. Abstract articles of 13th congress of Iran infectious diseases, pp. 27.