

Original Article

Study of *Yersinia enterocolitica* contamination in red meat supplied in Shiraz, Iran

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Summary

Yersinia enterocolitica is one of the intestinal pathogens that are transferred to human through water and food, especially infected meat. Food contamination with this bacterium causes diarrhea, vomiting, stomach pain, appendicitis syndrome, like fever and septicemia. This study aimed study the red meat supplied in Shiraz for *Yersinia enterocolitica* contamination. In this study, 200 samples of meat supplied in Shiraz were prepared, and were classified based on packaging and non-packaging, storage method and retention time of cart and were investigated for *Yersinia enterocolitica* contamination. Samples were transferred to PBS enriched environment containing sorbitol sugar and bile salts. These samples were stored in a refrigerator at 4°C for three weeks and then cultured on selected culture medium of *Yersinia enterocolitica*. Then, microbiological and biochemical subtraction tests were performed on samples. According to study, 13% of the collect meat samples (26 samples) was infected with *Yersinia enterocolitica*. The results showed no significant difference in the frequency of contamination among different groups of the samples. The studies show that a significant amount of red meat can be contaminated with bacteria when meat foods are not heated enough during cooking, and subsequent storage in the refrigerator provides the opportunity to amplification of this bacterium. Moreover, due to the lack of apparent signs of spoilage, this bacterium causes gastroenteritis and other gastrointestinal complications in consumers.

Keywords: *Yersinia enterocolitica*, Red meat, Digestion, Shiraz

Introduction

Microbes cause desirable and undesirable changes in food. Undesirable Changes cause food contamination and eventually food spoilage. *Yersinia enterocolitica* is one of the most important cryogenic pathogenic bacteria. This bacterium grows in the temperature range of 2-4°C by producing enterotoxin on meat stored in the refrigerator. can cause severe diseases without causing any visible signs of spoilage and without changing the smell and taste of meat (Inzana et al., 2001; Razavilar, 2003). *Yersinia* is an important intestinal pathogen that causes gastro-

enterocolitica, mesenteric lymphadenitis and non-digestive complications through food and water (Hamama et al., 1992). The extraordinary economic and nutritional importance of meat has led to extensive studies on microbial contamination and diseases of the region through meat in various countries. Also, healthy eating is one of the main pillars of public health (Farshchian et al., 2010). *Yersinia* is a zoonotic bacterium transmitted to humans through infected animals, especially pigs, cattle, sheep, goats, poultry, and infected vegetables; so this bacterium is known as food-borne. Therefore, microbial control of these

products is essential in terms of *Yersinia* contamination (Chlebicz and Ślizewska, 2018). Due to the importance of this bacterium in the discussion of food health, we decided to study the prevalence and contamination of freshly slaughtered red meat and red meat offered in grocery stores in Shiraz with *Yersinia enterocolitica*.

Materials and methods

In this study, 200 samples of red meat offered in Shiraz were randomly collect. The meats were classified based on the parameters of packing and non-packing, storage and shelf life, and were examined for *Yersinia enterocolitica* contamination. All samples were prepared from livestock slaughtered in the Shiraz industrial slaughterhouse system, which was the same in terms of slaughter. The first group consisted of packaged and unpackaged meats, the second group consisted of non-fresh meats found in butchers with two types of refrigerated and out refrigerated meats, and the third group consisted of freshly slaughtered and non-fresh meats. Samples were collected and transferred to the microbiology laboratory at each time under aseptic conditions and temperature control in the cold box along with the recorded questionnaire of specifications for each sample. Twenty-five g of the red meat sample, which was cut into very thin layers in a completely sterile condition, was transferred to a phosphate-

enriched medium containing sorbitol sugar and bile salts with a pH of 7.2. The samples were refrigerated for 4 weeks at 4°C to enrich the samples and then after 21 days one fold of the enriched suspension was mixed with nine folds of 25% potassium hydroxide (KOH) for 30 seconds. The Culture was performed on selective *Yersinia enterocolitica* CIN (Cefsulodin-Irgasan-Novobiocin) and incubated at 25°C for 24-48 hours. Positive cultures, which were characterized as the growth of convex microbial colonies with a red center and a clear colorless halo, were used for staining, specific and differential cultures, and biochemical experiments (Jabeen et al., 2016). Then, microbiological and biochemical tests were performed on samples (Anderson et al., 2011).

Results

Out of 200 red meat samples, 100 samples were packaged meat, and 100 samples were unpackaged meat. Out of 100 samples from unpackaged meats, 11 samples were infected with *Yersinia enterocolitica*. Out of 100 samples from packaged meats of, 15 samples were infected with *Yersinia enterocolitica*. The prevalence of *Yersinia enterocolitica* in packaged and unpackaged meats is described in Table 1. According to the statistical analysis, no significant difference between packaged and unpackaged groups in terms of *Yersinia enterocolitica* contamination was observed ($p > 0.05$) (Figure 1).

Table 1. Percentage of *Yersinia enterocolitica* in packaged and unpackaged red meats

Sample type	number of samples	Positive sample		Negative Sample	
		Number	Percentage	Number	Percentage
packaged	100	15	%15	85	%85
No packaging	100	11	%11	89	%89
Total	200	26	%13	174	%87

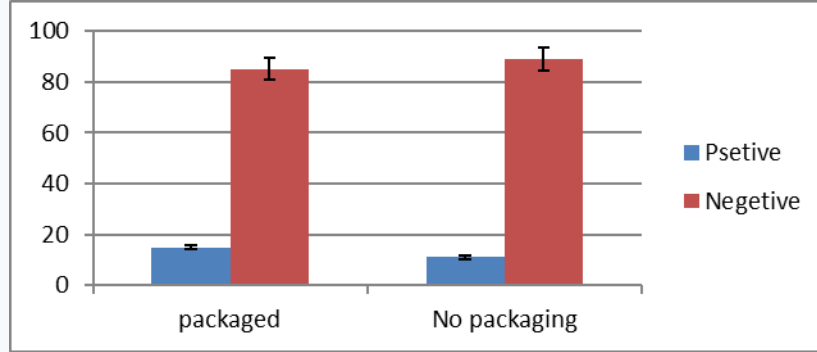


Fig. 1. Comparison chart between packed and unpacked groups

Unpackaged red meats are offered to consumers in two types, stored in the refrigerator and stored outdoors inside the butcher shop. Therefore, to compare the level of *Yersinia enterocolitica* contamination in these two types of unpackaged red meat, 25 butchers were sampled equally from 25 butcher shops in Shiraz. The results are

presented in Table 2. After performing the experiments, the statistical analysis of the data using the Fisher test showed that there was no significant difference in the level of contamination between meats stored inside the refrigerator and those stored outside the refrigerator ($p \geq 0.05$) (Figure 2).

Table 2. Percentage of *Yersinia enterocolitica* in red meats without packaging and stored either in the refrigerator or out of the refrigerator

Sample type	number of samples	Positive sample		Negative Sample	
		Number	Percentage	Number	Percentage
Inside the refrigerator	50	6	% 12	44	%88
Out of the fridge	50	5	% 10	45	%90
Total	100	11	% 11	89	%89

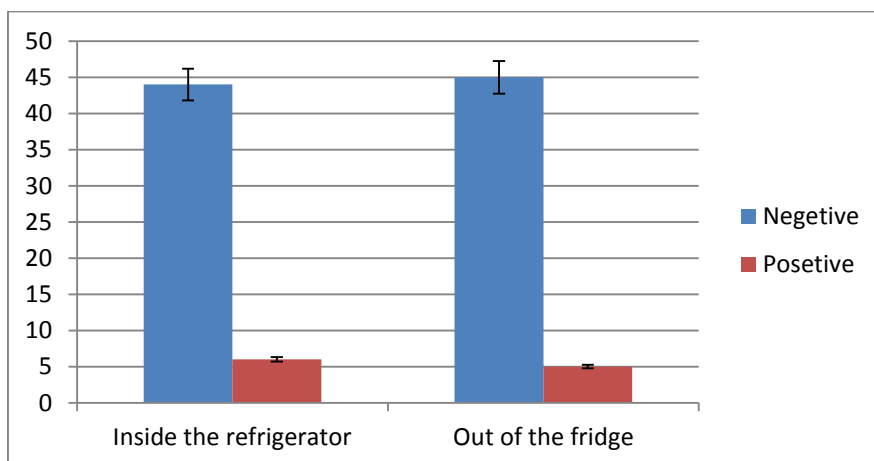


Fig. 2. Comparison chart between inside the refrigerator and outside the refrigerator groups

In the study, the level of contamination of meat carcasses kept in the open-air of the shop was

compared with the meat stored in the refrigerator, which had not been in the open air of the shop for

a long time. 50 samples of freshly slaughtered red meat was prepared from Shiraz Industrial Slaughterhouse with the results of 50 samples randomly selected from all packaged and non-packaged red meats that reach the consumer after

the transfer process. Fisher test showed that there was no significant difference in the rate of *Yersinia* infection ($p \geq 0.05$) (Figure 3). The results are present in Table 3.

Table 3. Percentage of *Yersinia enterocolitica* in freshly slaughtered and leftover red meat

Sample type	number of samples	Positive sample		Negative Sample	
		Number	Percentage	Number	Percentage
Remaining	50	6	% 12	44	% 88
Fresh	50	5	% 10	45	% 90
Total	100	11	% 11	89	% 89

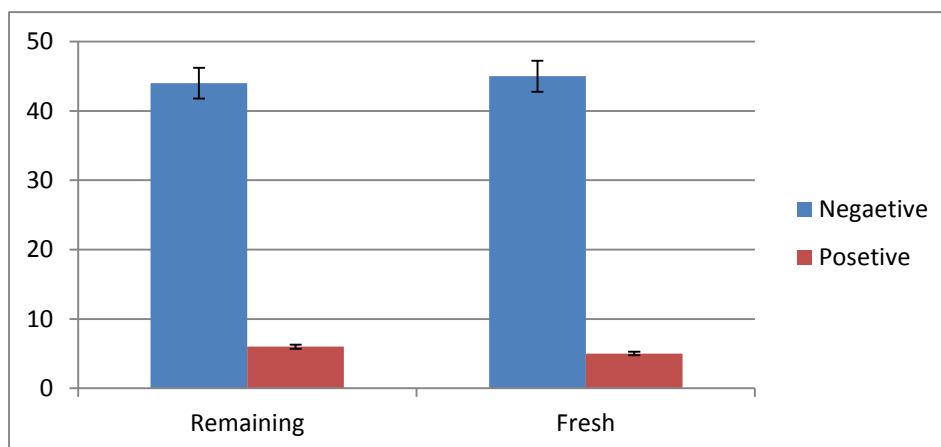


Fig. 3. Comparison chart between inside the fresh and outside the Remaining groups

Discussion

Yersinia enterocolitica is an important bacterium in food hygiene, and this bacterium can cause various digestive disorders humans in all different age groups and even in developed countries (Ghasemzadeh and Namazi, 2015). Enterocolitica yersiniosis is a common disease between humans and animals (zoonosis) and endangers the health of both (Kim et al., 2017). Therefore, recognizing the potential sources of this infection in the epidemiology of the disease seems necessary. Abundance information are available concerning the prevalence of *Yersinia enterocolitica* in other types of meat in different countries. The prevalence

of bacteria in various studies has been reported from 9% to 99.2% (Butler, 2004). In a survey of chicken and red meat in Tehran, the infection rate was reported to be 8.2% (Esfandiari et al., 2021). Another study reported 16% infection in chicken and red meat with *Yersinia* bacteria in Shemiran city (Ashrafi, 2009). In another study, the rate of contamination of chicken and red meat with the above bacteria was 44.4% in the south of Tehran (Sultan et al., 2006). In a study, 13.3% contamination of *Yersinia enterocolitica* was reported in the meat of Tabriz (Farshchian et al., 2010). In another study conducted in Shahrekord city, the contamination rate of chicken and red

meat with *Yersinia enterocolitica* was reported to be 14% (Shakerian et al., 2011). In another study conducted by (Sirken et al. 2004) in Aydin, Turkey, the incidence of red meat infection with *Yersinia enterocolitica* was reported to be 27.9% (Siriken, 2004). In the study of (Estrada et al. 2011) Argentina, the bacterium was found to be present in a variety of livestock and poultry meats, play a vital role in transmitting the disease (Estrada et al., 2011). In this study, the rate of *Yersinia enterocolitica* infection in red meat of Shiraz was 13%.

In comparison, there was no significant difference in the rate of infection with the above bacteria in the selected samples. However, the number of contaminants in packaged meats was higher than non-packaged meats, which may be due to the mixing of different carcasses during the packing process. Also, due to the lack of a significant difference in the statistical comparison of the level of contamination between fresh meat and leftover meat, it was observed that the amount of contamination of leftover meat was more than fresh meat. Residues of secondary contaminants during meat transfer to shops and contact of healthy and contaminated carcasses may increase contamination.

Statistical comparison of the frequency of contamination in refrigerated and non-refrigerated meats was not significant; but the amount of contamination in refrigerated meats was slightly higher than meats stored outside the refrigerator, which can be justified due to the cold-loving nature of this bacterium.

Conclusion

The abundance of this bacterium in red meat has a similar pattern with other regions. According to the present results, to prevent human infection with this bacterium, hygienic measures and training of people involved in the production, processing and preparation of livestock meat from slaughterhouses to supply centers are highly recommended.

Conflict of interest statement

The authors declare no conflict of interest.

Ethical approval

Not applicable

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