

Health Risk Assessment of *Toxoplasma gondii* and *Cryptosporidium* species in Red Meat (Implications for Pregnant Women and Immunocompromised Individuals)

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ABSTRACT

The examination looked for the prevalence in red meat samples-for slaughterhouses and retail meat centers in Shiraz and Kazerun counties, Fars Province, Iran- of two important zoonotic parasites: *Toxoplasma gondii* and *Cryptosporidium* spp. In total 25 samples of fresh meat were taken from five locations and analyzed by single-step polymerase chain reaction. This revealed that 36% of samples were contaminated by at least one of the two parasites: *T. gondii* in 24% of samples and 12% for *Cryptosporidium* spp. One sample was confirmed to be positive for both parasites. Infections were more prevalent in rural and semi- industrial settings. As such, these results demonstrate a potential public health threat posed by contaminated red meat, especially in at-risk populations like pregnant women or immunocompromised patients. Hence, the study argues for urgent implementation of routine molecular surveillance, microbiological safety certification for meat products, and public awareness campaigns as prime strategies for managing zoonotic disease risk.

Introduction

Providing background information and establishing context

Zoonotic parasites are among the most significant pathogenic agents shared between humans and animals, transmitted to humans through direct contact with infected animals or the consumption of contaminated food and water. These parasites can cause a wide range of diseases in humans and hold particular public health importance, especially in communities where health monitoring of animal product processing and consumption is limited. (AbdAllah et al., 2025)

Among zoonotic parasites, two parasites, *Toxoplasma gondii* and *Cryptosporidium* species, are of particular significance due to their widespread presence in the environment, food sources, and various hosts, as well as their high potential for transmission to humans. (Razakandrainibe, Ortega, & La Carbona, 2023) *T. gondii* is an obligate intracellular parasite from the Apicomplexa phylum, with the infection it causes known as toxoplasmosis. This parasite is capable of infecting all warm-blooded mammals, including humans, with the primary mode of transmission being the consumption of raw or undercooked meat contaminated with tissue cysts of the parasite. Additionally, transmission through contaminated water, unwashed vegetables, and congenital transmission has also been reported. Infection with this parasite can lead to symptoms such as congenital toxoplasmosis, encephalitis, visual disturbances, and miscarriage, particularly in pregnant women and immunocompromised individuals. (Salari et al., 2025) On the other hand, *Cryptosporidium* spp. is considered one of the common causes of acute diarrhea in humans. This parasite also belongs to the Apicomplexa phylum and comprises various species, some of which are pathogenic to humans. Cryptosporidiosis is primarily transmitted through the consumption of water or food contaminated with the parasite's oocysts or through contact with infected individuals or animals. The disease caused by this parasite is more severe and persistent in young children, the elderly, and patients with compromised immune systems, such as those with severe HIV/AIDS. Furthermore, the high resistance of *Cryptosporidium* oocysts to environmental conditions and standard chlorination of water contributes to their prolonged survival in water sources, posing a serious threat to public health. (Rideout, Cook, & Whetton, 2024) Given the potential role of red meat in the transmission of these parasites, investigating their prevalence in livestock intended for human consumption and analyzing the associated risks is of considerable importance from an epidemiological and food safety perspective.

Introduction of the specific topic of study and explanation of its significance

Numerous studies have highlighted the importance of zoonotic parasites, particularly *Toxoplasma gondii* and *Cryptosporidium* spp., in food contamination, especially in red meat. A comprehensive study in Europe revealed that 30 to 60 percent of toxoplasmosis cases are related to the consumption of undercooked contaminated meat. (Bieńkowski et al., 2022) The European Food Safety Authority (EFSA) also reported in its 2018 report that raw meat is one of the most significant sources of *T. gondii* contamination in humans. (Hazards et al., 2018) This parasite is particularly dangerous for pregnant women, as it can cross the placenta and infect the fetus, leading to miscarriage or fetal abnormalities. Studies have shown that primary infection during pregnancy can have serious consequences for the fetus. (Schneider et al., 2023) Conversely, the prevalence of diseases caused by *Cryptosporidium* spp. is also on the rise. This parasite is transmitted through drinking water and food contaminated with feces and poses a serious public health threat, especially in developing countries. Additionally, evidence has shown that *Cryptosporidium* spp. can also be transmitted to humans through contaminated meat, although this transmission route has received less attention. (Martinez et al., 2024) Given the potential role of red meat in the transmission of these parasites, investigating their prevalence in livestock intended for human consumption and analyzing the associated risks is of considerable importance from an epidemiological and food safety perspective.

Reference to Previous Actions for Addressing the Research Issue

In recent years, numerous studies have investigated the prevalence of zoonotic parasites such as *Toxoplasma gondii* and *Cryptosporidium* spp. in meat products, particularly red meat. These parasites are recognized as significant agents in the transmission of zoonotic diseases due to their considerable health impacts. For instance,

research conducted in Iraq has indicated that the red meat available in the market may be contaminated with *T. gondii*, highlighting the high potential for transmission of contamination from livestock to humans within the meat production and distribution chain (Almashhadany, 2020). In Iran, although some studies have focused on the presence of zoonotic parasites in slaughtered livestock in certain cities, comprehensive and systematic research at the national level or in major livestock farming regions has yet to be conducted. Therefore, the present study is designed to fill this scientific gap and provide accurate data regarding the prevalence of these parasites in red meat produced in Fars Province, which is considered one of the major livestock farming centers in the country.

Furthermore, previous research has emphasized that to mitigate the health risks associated with these parasites, strategies such as public education for consumers and meat industry personnel, improvement of slaughtering and meat storage processes in abattoirs, and regular microbiological testing of meat products are essential (Niyazi, 2023). Despite these measures, many developing countries lack specific and reliable standards and guidelines for controlling and preventing zoonotic parasite contamination in the meat production chain, which poses a significant challenge in the field of food safety (Savotchenko, 2020).

Consequently, conducting local and regional studies in this area, in addition to enhancing scientific knowledge, can contribute to the formulation of more effective health policies and the improvement of food safety standards. Research Gap and the Importance of Focusing on Vulnerable Groups

One of the most significant deficiencies in the scientific literature regarding zoonotic parasites is the lack of specific and comprehensive studies on high-risk groups such as pregnant women and immunocompromised individuals. These groups are particularly susceptible to severe illnesses and serious complications resulting from infections with parasites such as *Toxoplasma gondii* and *Cryptosporidium* spp. (Mulu Gelaw, Worku Dagnew, Degu Alene, Gangneux, & Robert-Gangneux, 2024). For example, infection with *T. gondii* in pregnant women can lead to vertical transmission of the parasite to the fetus, resulting in outcomes such as miscarriage, congenital anomalies, and long-term neurological damage in children. Additionally, immunocompromised individuals, including patients with HIV/AIDS or those undergoing immunosuppressive therapies, are at a heightened risk of severe and chronic infections due to their reduced bodily resistance (Wang et al., 2017).

Nevertheless, many previous studies have generally examined diverse populations in an integrated manner without distinguishing vulnerable groups, and there are rarely studies that specifically address the status of these high-risk groups. This research gap has resulted in health policies and strategies related to meat consumption control and prevention of parasite transmission in these populations being inadequate and non-targeted (Braam, Jephcott, & Wood, 2021). Consequently, a special focus on these vulnerable groups in future research will not only enhance the scientific understanding of the level of risk and transmission pathways of parasites but will also facilitate the design of more targeted and effective health interventions. Particularly in countries with high prevalence of zoonotic parasites, such studies are deemed essential for improving public health and reducing the burden of diseases.

Specific Objectives of the Study

The present study aims to investigate the contamination of *Toxoplasma gondii* and *Cryptosporidium* spp. parasites in red meat offered in slaughterhouses and meat supply centers in the cities of Shiraz and Kazeroon. This study seeks to contribute to a better understanding of the prevalence of these parasites in meat products in the region and to assess the associated health risks related to their consumption .

The specific objectives of the study are as follows :

- 1.To determine the prevalence of *Toxoplasma gondii* and *Cryptosporidium* spp. parasites in red meat samples collected from five different regions of Shiraz and Kazeroon .
- 2.To analyze and evaluate the potential health risks arising from these contaminations, particularly in vulnerable populations, including pregnant women and immunocompromised patients .
- 3.To provide practical and scientific solutions for controlling, preventing, and monitoring parasitic contamination in red meat, with the aim of enhancing food safety and protecting public health .

Materials and Methods

Study Design

This descriptive-analytical cross-sectional study was conducted in the year 1403 (2024 AD). The primary objective of the study was to identify the level of contamination of red meat with two significant zoonotic

parasites, *Toxoplasma gondii* and *Cryptosporidium* spp., and to analyze the health risks associated with them for high-risk groups .

Sampling Locations

Fresh red meat samples were collected from two cities, Shiraz and Kazeroon, in Fars Province. For this purpose, five sampling points were selected, which included the following :

1. Industrial slaughterhouse of Shiraz (central region) ,
2. Western meat market of Shiraz (Goyim region) ,
3. Kazeroon slaughterhouse ,
4. Butcher shops in the Jereh and Baladeh regions of Kazeroon ,
5. Meat supply centers in the Kouhmareh Noudan district (Kazeroon) .

Sample Collection

In total, 25 red meat samples from the muscular tissues of small livestock were collected, with five samples from each region. Each sample weighed approximately 50 grams. The samples were placed in sterile disposable containers and stored at a temperature of 4 degrees Celsius until transfer to the laboratory. The transfer of samples to the Veterinary

Results

In this study, a total of 25 red meat samples were collected from five different regions in Fars Province and subjected to molecular testing using PCR. The selected areas included three locations in Kazeroon County (the central slaughterhouse, butcher shops in the Jereh and Baladeh regions, and meat supply centers in the Kouhmareh Noudan section) and two locations in Shiraz County (the industrial slaughterhouse and the western meat market in the Goyim area). The results indicated that a total of 9 out of 25 samples (equivalent to 36%) tested positive for contamination with one of the two examined parasites, namely *Toxoplasma gondii* or *Cryptosporidium* spp .

Results related to *Toxoplasma gondii* contamination

Molecular evidence revealed that 6 samples (24%) contained DNA of the *T. gondii* parasite. The prevalence rates of this parasite in different regions were reported as follows :

- Shiraz – industrial slaughterhouse: 1 sample ,(20%)
- Shiraz – western meat market (Goyim): 2 samples ,(40%)
- Kazeroon – central slaughterhouse: 1 sample ,(20%)
- Kazeroon – Jereh and Baladeh: 1 sample ,(20%)
- Kazeroon – Kouhmareh Noudan: 1 sample ,(20%)

The highest prevalence of *T. gondii* was observed in the western meat market of Shiraz. However, the chi-square statistical test did not indicate a significant difference in the prevalence of this parasite among the different regions ($p > 0.05$) .

Table 1 – Distribution of *Toxoplasma gondii* contamination frequency in various regions of Shiraz and Kazeroon counties

Sampling area	Total sample size	number of positive samples	Pollution (%) percentage
Shiraz Industrial Slaughterhouse	5	1	20%
West Shiraz Meat Market (Goyim)	5	2	40%
Kazerun Central Slaughterhouse	5	1	20%
Jereh and Baladeh (Kazeroon)	5	1	20%
Kohmareh Nodan (Kazeroon)	5	1	20%

total sum	25	6	24%
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Results related to the pollution of *Cryptosporidium* spp .

Out of all samples, 3 samples (12%) tested positive for the presence of *Cryptosporidium* spp. DNA. The distribution of this pollution is as follows :

- Shiraz – West Meat Market: 1 sample (20%)
- Kazeroon – Jarreh and Baladeh: 1 sample (20%)
- Kazeroon – Kouhmareh Noudan: 1 sample (20%)

It is noteworthy that pollution by *Cryptosporidium* spp. was only observed in rural or semi-industrial areas. Similar to the previous case, statistical analysis showed that there is no significant difference in the prevalence of this parasite between different regions ($p > 0.05$) .

Table 2 – Distribution of the frequency of pollution by *Cryptosporidium* spp. in different areas

Sampling area	Total sample size	number of positive samples	Pollution (%) percentage
Shiraz Industrial Slaughterhouse	5	0	0%
West Shiraz Meat Market (Goyim)	5	1	20%
Kazerun Central Slaughterhouse	5	0	0%
Jereh and Baladeh (Kazeroon)	5	1	20%
Kohmareh Nodan (Kazeroon)	5	1	20%
total sum	25	3	12%

Simultaneous contamination with both parasites

In a sample from the Jereh and Baladeh region, simultaneous contamination with both *T. gondii* and *Cryptosporidium* spp. was identified, which accounted for 4% of the total samples. This finding is of particular importance, as the presence of multiple pathogens in a food source can significantly increase the risk of infection for vulnerable groups such as pregnant women and immunocompromised patients.

Table 3 – Cases of simultaneous contamination with both parasites

Sampling Area	Simultaneous pollution cases	Percentage of total samples (%)
Jereh and Baladeh (Kazeroon)	1	4%
Other Areas	0	0%
Total	1	4%

Supplementary Analysis

Based on the obtained data, the relatively high prevalence of *T. gondii* infection and the presence of

Cryptosporidium spp. in red meat samples indicate a potential risk of zoonotic transmission of these parasites through the consumption of contaminated meat. Additionally, the lack of statistically significant differences between the sampling areas may suggest a uniform distribution of these parasites across the studied regions. However, the higher prevalence of contamination in the western meat market of Shiraz compared to other areas may require further investigation due to various factors such as meat storage and supply conditions.

Furthermore, the simultaneous presence of contamination in a single meat sample underscores the necessity for special attention to quality and hygiene control in the meat supply chain. This is particularly important for protecting the health of vulnerable groups and reducing the risk of parasitic disease transmission through meat.

Table 4 – Statistical results of the Chi-square test for comparing the prevalence of parasites between regions

angel	Chi-square test statistic	p-value (significance level)	Statistical result
<i>Toxoplasma gondii</i>	$\chi^2 = 1.33$	0.85	No significant difference was observed
<i>Cryptosporidium</i> spp.	$\chi^2 = 1.25$	0.87	No significant difference was observed

Research and Conclusion

The results of this study showed that a significant percentage of the red meat samples examined in the counties of Shiraz and Kazeroun were contaminated with one of two important zoonotic parasites, *Toxoplasma gondii* and *Cryptosporidium* spp. Specifically, contamination with *T. gondii* was observed in 24% of the samples, while contamination with *Cryptosporidium* spp. was found in 12%. These findings clearly indicate the importance of continuous monitoring of red meat health and the necessity for control and preventive measures, especially for vulnerable populations such as pregnant women, children, the elderly, and immunocompromised patients .

Contamination with *T. gondii*, which is transmitted through the consumption of undercooked or raw meat, is associated with dangerous diseases such as congenital toxoplasmosis, encephalitis, and ocular complications. (Salari et al., 2025) The findings of this research align with recent studies in Iran; for example, Abbaszadeh et al. (2024) reported that soils contaminated with oocysts shed from cat feces are one of the main sources of contamination for livestock with this parasite. Additionally, the relatively uniform distribution of contamination in the various studied areas indicates the widespread spread of this parasite in the local meat chain. (Lafmejan Pour et al., 2024)

Regarding *Cryptosporidium* spp., despite the lower prevalence, it should be noted that the oocysts of this parasite have high resistance to environmental conditions and disinfectants, including chlorine, which allows them to persist in the environment, even after slaughter and during the meat transmission chain. (Efstratiou, Ongerth, & Karanis, 2017) The findings of this study also confirm the presence of contamination in rural or semi-industrial areas, likely due to weaker sanitary controls in these regions .

Another noteworthy point is the simultaneous contamination with both parasites in a sample from the Jereh and Baladeh area. This is very important from a public health perspective, as concurrent contamination can have synergistic effects on disease occurrence and significantly increase the risk for high-risk groups. Similar studies have also reported the simultaneous presence of multiple pathogens in raw animal products, emphasizing the need for multidimensional policies in the monitoring and sanitary control of meat products. (Yu et al., 2022)

Considering that zoonotic parasite contamination usually exists in a latent and asymptomatic form in meat, consumers are unknowingly at risk. Factors such as consuming undercooked meat, not adhering to the cold chain, and contact with contaminated surfaces during storage and cooking are among the most important transmission routes for the disease. Therefore, increasing public awareness, updating

health standards in slaughterhouses and supply centers, and conducting regular screening tests on meat products should be considered fundamental strategies in national programs for controlling zoonotic diseases .

Ultimately, this research, as one of the foundational studies in the Fars region, can serve as a basis for broader research and more effective policymaking in the field of food safety and the control of zoonotic pathogens .

Final Conclusion

This study demonstrated that red meat offered in some areas of southern Iran contains significant zoonotic contamination. Given the high sensitivity of vulnerable groups such as pregnant women, HIV-positive patients, individuals undergoing immunosuppressive treatments, the elderly, and other immunocompromised patients, immediate and extensive actions are necessary to enhance health monitoring and public education. Accordingly, it is recommended :

- To implement regular molecular and microbiological monitoring in slaughterhouses and meat supply centers for the rapid and accurate identification of pathogens ;
- To develop and enforce stricter standards for issuing microbiological health certificates for red meat before it is offered to the market ;
- To design and conduct ongoing educational programs for butchers, livestock farmers, and consumers aimed at increasing awareness of prevention methods against contamination and transmission of zoonotic parasites ;
- To encourage and support applied and comprehensive research to more precisely examine the relationship between parasitic contamination in meat products and the occurrence of zoonotic diseases in regional human populations .

Implementing these measures will not only help improve the quality and safety of meat products but can also play a significant role in reducing the burden of zoonotic diseases and enhancing the overall health of the community.

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