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Investigation of the Status of Infection of Minced Beef With *Sarcocystis* Parasite in Samples Collected From Butchers in Tabriz

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Abstract

Introduction: Sarcocystosis is one of the most common protozoan parasites in the domestic livestock and causes severe infection in some hosts such as cattle and sheep. In addition to domestic animals, *Sarcocystis* infects many wild animals, birds, cold-blooded animals, and humans. The purpose of this study was to investigate and estimate the initial status of *Sarcocystis* infection in minced beef in Tabriz.

Methods: For digestion test and diagnosis of *Sarcocystis* contamination from butchers in Tabriz, 300 samples and 100 grams of minced meat samples were prepared and transferred in plastic bags and separately to the parasitology laboratory of the Faculty of Veterinary Medicine in Islamic Azad University of Tabriz, Iran. After preparing the digestive solution according to the standard method, an experiment was performed for each sample.

Results: In this study, out of the total number of 300 collected samples, 189 samples were infected with the *Sarcocystis* parasite (63%).

Conclusion: The results of this study revealed that the prevalence of *Sarcocystis* infection is high and this issue, in addition to inflicting economic losses due to livestock losses and seizure of infected carcasses or organs, as well as reducing livestock production, indicates the existence of health risks for residents. Due to the life cycle and epidemiology of this parasite, the need for further studies and control and health measures is important.

Keywords: Sarcocystis, Minced meat, Beef, Butchers of Tabriz, Iran

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Introduction

Attempting to maintain human and animal health and providing desirable and healthy food for human consumption are among the serious and important tasks of the veterinary community of any country; this is conducted through the prevention, control, and treatment of livestock diseases, especially common diseases between humans and animals, and monitoring of the slaughter and distribution of livestock products. Sarcocystosis is one of the parasitic diseases that is of considerable economic importance based on the experiences gained in Iran and other countries in slaughter animals (1).

This parasite is considered one of the coccidial parasites that causes cysts in animals. Different species of this parasite in its evolution have two hosts, including the final host of the group of carnivores and the intermediate host of the group of herbivores (2).

Sarcocystosis is one of the most common protozoan parasites in domestic livestock and causes severe infection in some hosts such as cattle and sheep. In addition to domestic animals, Sarcocystis infects many wild animals, birds, cold-blooded animals, and humans. Some species of Sarcocystis can cause disease, and thus weight loss, anorexia, fever, anemia, muscle weakness, decreased milk production, abortion, and sometimes death in intermediate hosts such as cattle, sheep, goats, and pigs. Some types of Sarcocystis in humans cause gastrointestinal disorders such as nausea, vomiting, and diarrhea (3). Humans are usually infected by eating undercooked or raw livestock meat. So far, various species of Sarcocystis have been reported from animals around the world, and three species of sarcocysts have been identified in cattle, and their life cycle in dogs, cats, and humans has undergone investigation. The form of the parasite is almost the same in all hosts, but its dimensions are different. Bags known as Meisher tubes are occasionally observed with the naked eye. In sheep, these objects are about 1 cm or smaller in size and are rarely found with the naked eye in cattle. So far, few studies have been performed on the infection rate of ruminants in Iran with



this protozoan, and most studies have been conducted on the macrocyst infection of this parasite, which is far less than the actual rate of infection with this parasite (4). It is also pathogenic to animals that act as final or definitive hosts (e.g., dogs). All organs or cysts formed by the species of the genus *Sarcocystis* are called sarcocysts.

Previously, *Sarcocystis* was thought to be insignificant, but over time, it became clear that the parasite could lead to encephalitis and even abortion (5).

Considering that minced beef is one of the most consumed foods in the life of urban people, the presence of parasitic infections can cause health problems and diseases in humans. Therefore, this study aimed to investigate the status of *Sarcocystis* parasite infection in minced beef; it is an attempt to highlight the importance of the *Sarcocystis* parasite in human health, and the results of this study can pave the way for further studies in this field (6).

Materials and Methods

This study sought to evaluate the status of *Sarcocystis* parasite infection in minced beef in Tabriz in 2021. To perform digestive experiments on beef collected from butchers in Tabriz, 300 samples of minced beef in the amount of each sample of 100 g of minced meat samples were randomly prepared from different butchers in Tabriz in plastic bags and separately; then, they were transferred to the parasitology laboratory of the Faculty of Veterinary Medicine, Islamic Azad University of Tabriz, Iran and tested for *Sarcocystis* infection.

To perform the test, a digestive solution is required, which was prepared using 100 cc phosphate buffer, 10 cc hydrochloric acid, and Pepsin powder 2.5 g.

An experiment was performed for each sample after preparing the digestive solution in several steps. First, 50 g of minced meat was poured into 100 cc of the digestion solution and placed in a Ben Murray at 37°C for 30 minutes. For the greater effect of the digestive solution on the sample, the sample was shaken inside the snake bin every few minutes. After 30 minutes, the mixture was poured on a cleaning cloth of three to four layers, and the liquid obtained by squeezing the mixture in a cleaning cloth was transferred to a test tube. The filtered liquid was centrifuged at 1500 rpm for 10 minutes. After centrifugation, the supernatant was then discarded, and the necessary expansions on the slide were prepared from the resulting sediment. After drying the slide, the expansion was fixed with methyl alcohol and stained with Giemsa paint. Finally, the stained slides were examined under a light microscope with a lens of forty hundred (Figures 1 and 2).

Results

In this study, out of 300 samples of tested minced beef, 189 samples were positive and infected with the *Sarcocystis*

parasite (63%), (Table 1). No *Sarcocystis* infection was observed in 111 cases of minced meat samples. The results of this study revealed that the prevalence of *Sarcocystis* infection in Tabriz is relatively high. This issue, in addition to imposing economic losses due to livestock losses and confiscation of infected carcasses or organs, as well as reducing livestock production, represents the existence of health risks to human populations. Due to the life cycle and transmission of this parasite, it requires more comprehensive health and control measures.

Discussion

Considering that protein is one of the basic needs of human nutrition, in proportion to the growing population and the growing need of society for livestock products, related diseases and health issues of meat and food should be given more attention. Sarcocystosis is one of the most common protozoan parasites in domestic livestock and leads to severe infection in some hosts such as cattle and sheep. Abattoir examination typically detects only



Figure 1. Sarcocystis Bradyzoite Cysts in Beef Striated Muscle. H & E staining ($\times\,100)$



Figure 2. Sarcocystis Bradyzoites in Digested Samples After Staining With Giemsa ($\times 1000$)

Table 1. Infection Rate of Sarcocystis Parasite in the Minced Beef of Tabriz

Number of Tested _ Samples	Positive Cases		Negative Cases	
	Number	Percent	Number	Percent
300	189	63	111	37

macroscopic cysts. Therefore, the statistics provided by slaughterhouse inspection are normally lower than the actual statistics of *Sarcocystis* parasite infection (7).

The economic value of billions of rials of the country's livestock capital and attention to various diseases, especially parasitic diseases, threatens human sanitation and health in addition to having negative effects on the amount and quality of livestock production and reveals the need to pay attention to the issue of animal health. According to the statistics of the Ministry of Agriculture, East Azerbaijan province in general and the central city of Tabriz in particular are important centers of red meat production in the country.

Some species of *Sarcocystis* in humans cause gastrointestinal disorders such as nausea, vomiting, and diarrhea. Therefore, considering the importance of this parasite, the present study was performed in this regard (8,9).

Dogs and cats are usually definitive hosts for a number of known species of ovine *Sarcocystis*. One of the causes of severe infection in intermediate hosts is attributed to the fact that farm animals are in close contact with herding guard dogs and that dogs infect pastures with *Sarcocystis* sporocysts. A study in Baghdad demonstrated that infected dogs excrete about 200 million sporocysts (about 4 million sporocysts daily) during the infestation period, causing the ruminant to become infected with *Sarcocystis* (9,10).

In Iran, the first research dates back to 1974, when Afshar et al examined the prevalence of *Sarcocystis* in sheep (8). Further, Diezbasnis and Fayer investigated *Sarcocystis* in cattle in 1980.

Using the digestion method in a study on buffalo meat in the Ghaem Shahriyar slaughterhouse around Tehran, Claveria reported the presence of 100% contamination (10,11).

The results of some studies indicated the superiority of the digestive method over the pathological method in some aspects, including the fact that the digestive method is faster and the equipment and materials used are less expensive and less. Moreover, it was found that staining of samples is less expensive, and significant amounts of muscle tissues are digested in the digestive method, increasing the chances of obtaining positive results, while in the pathological method, this is impossible (12-14).

In the present study, which was performed on beef in Tabriz, Iran, 63% of the samples were infected with the *Sarcocystis* parasite, representing a relatively high level of infection in slaughtered cows in Tabriz, which requires further studies in this field.

In a study conducted in 2001 using the tissue expansion method in Isfahan, Iran, 94.8% of the studied cows were infected, the results of which are consistent with those of this study (15).

In another study on beef mince in Bukan, Iran, in which

minced meat samples were digested, 74 out of 102 beef minced samples (72.54%) were positive for *Sarcocystis* infection, and the density of bradyzoites was observed in abundance in all tested samples (3), which is in line with the results of a study conducted in Tabriz (4,5).

In a study conducted by Dubey et al. on the infectious species of sarcocyst in cattle, it has been shown that their final hosts are dogs and carnivores, and microscopic sarcocyst cysts are produced in intermediate hosts such as cattle (14).

It is also stated that the species that are the final host of the cat are in limited areas due to less contact between the cow and the cat or sporocysts excrete less from cats than from dogs, indicating why microscopic cysts are more common than macroscopic ones (15,16).

Based on the results of this study, the high prevalence of *Sarcocystis* represented that microscopic cysts are present in meat carcasses consumed by humans, which requires further attention. Additionally, from a health point of view, the recommendations for observing the complete cooking of meat, and the like should be emphasized more.

In a 1974 study in Australia, more than 90% of cows were diagnosed as infected by *Sarcocystis* (17).

In a 1986 study by Heckeroth et al over 1273 cows slaughtered in 9 areas in South Moravia, the contamination rate of 75-93% was reported by the digestion method. In another study by Pena and Ogassawara, the rate of contamination by the digestion method in cattle in the Brno region of Czechoslovakia was 85.4% (18). These results conform to the results of a survey conducted in Tabriz (63%)(27).

Poulsen and Stensvold estimated that the world's cows were infected with the parasite by more than 90%. In the study by Rahdar, the heart (98%) and tongue (78%) had the highest infection rates, respectively (19-22).

The results of various studies demonstrated that the world's cows are severely infected with this parasite (23,24).

Some of the main reasons for the high prevalence of microscopic contamination with this parasite in cattle are the frequency of the final hosts of microscopic species of parasites, namely, humans and dogs, the consumption of raw beef or semi-cooked beef by humans and dogs, and the release of livestock in the environment. The other reasons include the slaughter of livestock outside the slaughterhouse, the presence of stray dogs and their access to slaughterhouse waste and meat waste, the contamination of water and forage of animals with sewage, and dog excrement (25-27).

Conclusion

The results of this study show that the prevalence of Sarcocystis infection is high and this issue, in addition to inflicting economic losses due to livestock losses and seizure of infected carcasses or organs, as well as reducing livestock production, indicates the existence of health risks for residents Is also Due to the life cycle and epidemiology of this parasite, the need for further studies and control and health measures is important.

Suggestions

- 1. Performing research on anti-Sarcocystis vaccines
- 2. Considering that domestic animals are infected by carnivorous fecal sporocysts and often keep cysts in their muscles in adult animals such as cattle and buffalo, the final hosts (dogs and cats) should not be allowed to feed on raw meat and carcasses of dead animals, and dogs and cats should not be allowed to enter the farm
- 3. Using anticoccidial drugs (amprolium and salinomycin) for prevention in pets

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Authors' Contribution

Conceptualization: Yagoob Garedaghi. Data curation: Agustin Olmedo-Juárez. Formal analysis: Yagoob Garedaghi. Funding acquisition: Yagoob Garedaghi. Investigation: Agustin Olmedo-Juárez. Methodology: Yagoob Garedaghi. Project administration: Agustin Olmedo-Juárez. Resources: Yagoob Garedaghi. Supervision: Yagoob Garedaghi. Validation: Yagoob Garedaghi. Visualization: Agustin Olmedo-Juárez. Writing–original draft: Yagoob Garedaghi. Writing–review & editing: Agustin Olmedo-Juárez.

Competing Interests

The listed authors declare no conflict of interests in any capacity, including competing or financial.

Ethical Approval

All ethical principles are fully observed in this study.

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