



# Intestinal Protozoan Infections in Cancer Patients Undergoing Chemotherapy in Shahrekord the Central Southwest of Iran in 2018

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## Abstract

**Background and aims:** Parasitic infections, especially opportunistic parasites, can cause serious problems for immunocompromised patients. In this regard, this study aimed to evaluate parasitic infections in cancer patients undergoing chemotherapy after referring to Kashani hospital of Shahrekord from January to May 2018.

**Methods:** In this cross-sectional descriptive study, 750 stool samples were collected from 250 cancer patients undergoing chemotherapy (three fecal samples from each patient) and sent to the Parasitology Laboratory of Shahrekord University of Medical Sciences. All samples were examined macroscopically and microscopically through the direct method and a specific acid-fast staining method. To confirm the microscopic results, DNA was extracted from isolated parasites and amplification was performed by the polymerase chain reaction with the specific primers on positive samples.

**Results:** In this study, 106 (42.4%) and 144 (57.6%) patients were males and females, respectively. In addition, 187 (74.8%) and 63 (25.2%) of them were living in urban and rural areas, respectively. In terms of the education level, most patients had a low level of literacy. Further, 12 (4.8%) patients were infected with intestinal parasites that including, 7 (2.8%), and 5 (2%) samples were infected with *Blastocystis hominis* and *Giardia lamblia*, respectively.

**Conclusion:** The results of this study showed that *B. hominis* and *G. lamblia* are the most prevalent parasites in cancer patients undergoing chemotherapy. Although the prevalence of intestinal parasites in cancer patients was low, these findings should not be contrary to the need for healthcare regarding the prevention of parasitic infections in cancer patients.

**Keywords:** Intestinal protozoa, Cancer patient, Chemotherapy, Shahrekord

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## Introduction

One of the most important general public health indicators of any community is the status of its infectious diseases including parasitic (i.e., protozoan and helminth) infections.<sup>1,2</sup> Protozoan infections cause global problems such as barriers to social and economic development, particularly in developing countries.<sup>3</sup> Therefore, the efficient control of parasitic infections can be an advantage of the health and economy of countries.<sup>4</sup> Despite persistent efforts and the extensive planning of the World Health Organization (WHO), they are still one of the major health problems in developing countries.<sup>3</sup> According to the WHO report, the number of people who are chronically infected with different types of parasites in worldwide is estimated to be approximately 3 billion persons.<sup>5,6</sup>

In addition, it seems that the probability of infection is high by various types of microorganisms (e.g., parasites, fungi, bacteria, and viruses) in immunocompromised persons such as cancer patients due to the involvement of lymphocytes and the degradation of the immune system of the body. The infection of immunocompromised patients with these microorganisms can be a major cause of morbidity and mortality in these groups.<sup>7</sup> The diagnosis of infection in these patients is often difficult according to the typical symptoms of infection (e.g., fever) and remains hidden or absent due to a decrease in white blood cells, thus it is highly necessary to identify infections in this group of patients.<sup>8,9</sup> On the other hand, the number of immunocompromised patients is increasing gradually in the world due to an increase in the number

of cancer patients and the progress of cancer diagnosis methods, followed by treatment with chemical drugs, radiation and organ transplants in different wards of the hospital.<sup>10</sup> Regarding the limited number of studies on the parasitic infection in immunocompromised patients in Chaharmahal va Bakhtiari province, it is necessary to investigate the frequency of protozoan parasitic infections in cancer patients undergoing chemotherapy. Nonetheless, given the importance of the timely diagnosis of parasitic infections in these patients and their critical need for treatment against these infections, limited studies are available in this regard. It is noteworthy that using diagnostic methods for the parasitic disease is attractive and effective because of features such as the lack of a need for expensive and complicated equipment, high sensitivity and specificity, and the speed in obtaining the results. Accordingly, this study aimed to evaluate the frequency of intestinal protozoa in cancer patients in Shahrekord in 2018.

### Materials and Methods

In this descriptive cross-sectional study, data were collected through observation, interviews and recorded in demographic forms for each patient. The required data such as individual characteristics were extracted from information forms and the results were evaluated and reported accordingly. The study population included cancer patients undergoing chemotherapy who referred to the Kashani hospital of Shahrekord, which is the capital of Chaharmahal va Bakhtiari province located in the central southwest of Iran.

#### *Number and Method of Sampling*

Census and non-probabilistic sampling methods were used in this study. Approximately, 90-100 cancer patients are admitted to the oncology ward of Kashani hospital every month. Therefore, sampling was done on 250 cancer patients who were admitted under the chemotherapy process for 1 month to more than 37 months during January-May 2018 and were willing to participate in the study. Before starting the procedure, permission was obtained from the Ethics Committee and hospital authorities. A questionnaire was prepared, containing information such as age, gender, type of job, and level of education. After questionnaire completion, three stool collection containers labeled with the name or number of the patient were given to each subject. Then, the samples were quickly transported to the Department of Parasitology and Mycology, the School of Medicine, Shahrekord University of Medical Sciences, Shahrekord, Iran.

### *Parasitology Methods*

#### *Direct Method*

The most commonly performed procedure in parasitology

is the observation and identification of trophozoites, cysts, and oocysts of protozoa and ova, along with the larva and the segment of helminths by the wet mount technique. The physiological serum was used to prepare wet slides. Accordingly, the name and number of the patient were written on one side of the slide using a diamond pen, and then a drop of physiological serum was placed on the slide. Using a wooden applicator, a small portion of the sample (about 2 mg) was removed and gently mixed with the physiology serum. Furthermore, the lamellae (22 × 22 mm) were gently placed on the sample and the droplet mixture so that the lamellae edge was in contact with the droplet and prevented bubble formation. For microscopic observations, the entire surface of the slide was regularly examined from one corner upward, downward, backward, and forward at ×100 magnification. In the case of suspected cases, ×400 magnification and appropriate light were utilized for the exact detection of trophozoites and the cysts of protozoa, as well as the eggs and larvae of helminths.<sup>7</sup>

#### *Cold Ziehl Neelsen Method*

The alkaline fuchsin stain includes two strains (A and B) that are prepared as follows.

To prepare solutions A and B, 0.3 g of the fuchsin stain and 5 g of phenol crystals were dissolved in 10 mL of 95% ethanol and 100 mL of distilled water and then heated, respectively. Next, solution B was added to solution A. It should be noted that this stain is stable for 1 year at room temperature. In this study, 1 mL of hydrochloric acid was added to 99 mL of methanol at 95%. To prepare the background stain, 0.3 g of methylene blue was dissolved in 100 mL of distilled water. Moreover, the physiologic serum was used to prepare smear from consistent samples. The smears were prepared in such a way that newspaper inscriptions were readable after drying. The prepared smears were then air-dried, and absolute methanol was applied to stabilize the sample in staining stages.<sup>7</sup> Additionally, the smear prepared from the stool sample was dried. To stabilize the smear, it was fixed for 3 minutes using methanol, and the surface of the smear was covered with carbol fuchsin for 20 minutes. Next, the smeared surface was washed with water. Further, staining was performed 1 to 10 second(s) using 1% chloric acid bleach and then the surface was re-washed with water. In addition, the smeared surface was stained and re-washed using the 0.3% methylene blue solution for 30 seconds. Eventually, the smear was air-dried and then examined by light microscopy at ×100 and ×400 magnifications and using the impression oil at ×1000 magnification.<sup>8</sup>

#### *Polymerase Chain Reaction*

To confirm the obtained results by microscopy, samples that were positive for intestinal protozoa were tested on positive samples by the polymerase chain

reaction (PCR) and nested PCR for *B. hominis* and *G. lamblia*, respectively, in the parasitology laboratory of Shahrekord University of Medical Sciences. Next, DNA was extracted from stool samples using the Qiagen kit (QIAamp® DNA Stool (50541)) according to the manufacturer's instructions. For purity and quality, DNA was surveyed by a NanoDrop device (Thermo Scientific, Lithuania). Then, microscopic results were confirmed using specific primers, namely, forward primer 3'-ATCTGGTTGATCCTGCCAGT-5'-and reverse primer 3'-GAGCTTTTAACTGCAACAACG-5' of *B. hominis* designated by Pandey et al<sup>9</sup> and the outer forward primer (AL3543) 3'-AAATATGCCTGCTCGTCG-5' and the outer reverse primer (AL3546) 3'-CAAACCTTITCCGCAAACC-5' and the inner forward primer (AL3544) 3'-CCCTTCATCGGIGGTAAGT-5', and the inner reverse primer (AL3545) 3'-GTGGCCACCACICCCGGGGGGGGGGGGGGGGGG of *G. lamblia*. The primers were used for the nested PCR primers described by Sulaiman et al.<sup>11</sup>

The PCR for *B. hominis* was performed and a 600 bp fragment amplified the SSU rDNA gene. Moreover, PCR was performed in a 20 µL mixture containing the template 4 µL of DNA, 0.5 µL of each primer (10 pmol/µL), 5 µL distilled water, and 10 µL of the master mix (Ampliqone, Denmark). The PCR conditions consisted of one cycle denaturing at 95°C for 5 minutes, 35 cycles including denaturing at 95°C for 20 seconds, extending at 58°C for 20 seconds, annealing at 72°C for 20 seconds, and an additional cycle with a 5-minute chain elongation at 72°C.

The nested PCR for *G. lamblia* was done with the production of a 530 bp fragment amplified of the TPI gene for two steps. This work was conducted in a 20 µL mixture containing the template 2 µL of DNA (2.5 µL in the second stage), 0.5 µL of each primer (10 pmol/µL), 7 µL distilled water (6.5 µL in the second stage), and 10 µL of the master mix (Ampliqon, Denmark). After initial denaturation at 94°C for 5 minutes, each of the cycles consisted of denaturation at 95°C for 45 seconds, annealing at 50°C for 50 seconds, and extension at 72°C for 1 minute for 35 cycles, and final extension was done at 72°C for 10 minutes. Finally, the PCR product was stained with power load and electrophoresed on a 1.5% gel, and then was visualized on a UV-Transilluminator by the gel documentation system.

**Data Analysis Method**

The results of each individual's test were recorded in a questionnaire. Eventually, SPSS 16 software and chi-square and Fisher exact tests were used for extracting data and performing statistical tests. In this survey,  $P < 0.05$  was considered statistically significant.

**Results**

In this study, 250 cancer patients within the age range of

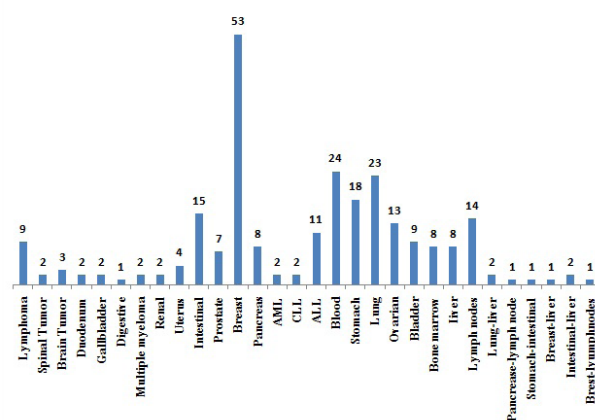
4-85 years with a mean of 49.1 years (SD = 20.09) suffering from different cancer types (Figure 1) were evaluated in terms of intestinal parasitic infections. Table 1 presents demographic data including gender, living place, age, job, education level, and the duration of chemotherapy.

In the present study, 7 (2.8%) and 5 (2%) samples out of 250 cancer patients were infected with *B. hominis* and *G. lamblia*, respectively. These parasites were detected in the group of patients with acute lymphocytic leukemia (n=5, 41.8%), digestive (n=2, 16.7%), chronic lymphocytic leukemia (n=1, 8.3%), lung (n=1, 8.3%), bladder (n=1, 8.3%), stomach (n=1, 8.3%), and breast cancer (n=1, 8.3%). It is noteworthy that all clients were sampled three times, and all samples were examined in duplicate. In addition to the direct method, all samples were examined using modified Ziehl Neelsen staining. The results of this study showed that 100% of the stained samples were negative in terms of the presence of Coccidia oocysts, and the concentration of the extracted DNA was surveyed with a NanoDrop device in terms of quantity, purity, and quantity prepared for the PCR reaction optical density (OD) : 1.75).

Primers AL3543 and AL3546, as well as AL3544 and AL3545, were used to confirm *G. lamblia* infection. All samples of the patients infected by *G. lamblia* in the direct assay were confirmed by the PCR, and the results indicated that they were also positive in the molecular method (Figure 2).

Additionally, the single-step PCR through RD5 and BhrDR primers was used to determine the *B. hominis* genus. In addition, all samples of patients infected with *B. hominis* were examined by PCR methods for final approval of this parasite (Figure 3).

Similarly, all samples of patients infected with *G. lamblia* and *B. hominis* were examined by PCR methods regarding the final approval of these parasites.



**Figure 1.** Evaluation of Cancer Type in Cancer Patients Undergoing Chemotherapy Referring to Ayatollah Kashani Hospital in Shahrekord in 2018.

Note. AML: Acute myeloid leukemia; CLL: Chronic lymphocytic leukemia; ALL: Acute lymphocytic leukemia.

**Table 1.** The Prevalence of Intestinal Parasites Based on Demographic Characteristics in Cancer Patients Referring to Ayatollah Kashani Hospital in Shahrekord in 2018

Variable		No. (%)	Infected No. (%)	Non-infected No. (%)	P Value
Gender	Male	106 (42.4)	7 (6.6)	99 (93.4)	0.198
	Female	144 (57.6)	5 (3.5)	139 (96.5)	
Living place	Urban	187 (74.8)	7 (3.7)	180 (96.3)	0.148
	Rural	63 (25.2)	5 (7.9)	58 (92.1)	
Age groups (year)	<10	17 (6.8)	1 (5.9)	16 (94.1)	0.982
	11-20	12 (4.8)	1 (8.3)	11 (91.7)	
	21-30	14 (5.6)	1 (7.14)	13 (92.86)	
	31-40	27 (10.8)	1 (3.7)	26 (96.3)	
	41-50	49 (19.6)	3 (6.1)	46 (93.9)	
	51-60	55 (22)	2 (3.6)	53 (96.4)	
Job	60 <	76 (30.4)	3 (3.9)	73 (96.1)	0.130
	Farmer	12 (4.8)	2(16.6)	10 (83.4)	
	Ranchman	10 (4)	0	10 (100)	
	Employee	14 (5.6)	0	14 (100)	
	Self-employed	55 (22)	1 (1.8)	54 (98.2)	
	Housewives	116 (46.4)	2 (1.72)	114 (98.3)	
	Unemployed	27 (10.8)	5 (20.8)	22 (79.2)	
Education level	Retired	16 (6.4)	2 (12.5)	14 (87.5)	0.227*
	Illiterate	87 (34.8)	1 (1.15)	86 (98.85)	
	Elementary	60 (24)	7 (11.7)	53 (88.3)	
	Secondary	31 (12.4)	1 (3.2)	30 (96.8)	
	High school	41 (16.4)	1 (2.4)	40 (97.6)	
Duration of chemotherapy (months)	Academic	31 (12.4)	2 (6.5)	29 (93.5)	0.027
	1-6	162 (64.8)	7 (4.3)	155 (95.7)	
	7-12	27 (10.8)	2 (7.4)	25 (92.6)	
	13-24	29 (11.6)	0	29 (100)	
	25-36	17 (6.8)	2 (11.8)	15 (88.2)	
Macroscopic results of stool samples (consistency)	Over 37	15 (6)	1 (6.7)	14 (93.3)	0.946
	Formed	147 (58.8)	8 (5.4)	139 (94.6)	
	Soft	74 (29.6)	3 (6.4)	71 (93.6)	
	Loose	23 (9.2)	1 (4.3)	22 (95.7)	
	Watery	6 (2.4)	0	6 (100)	

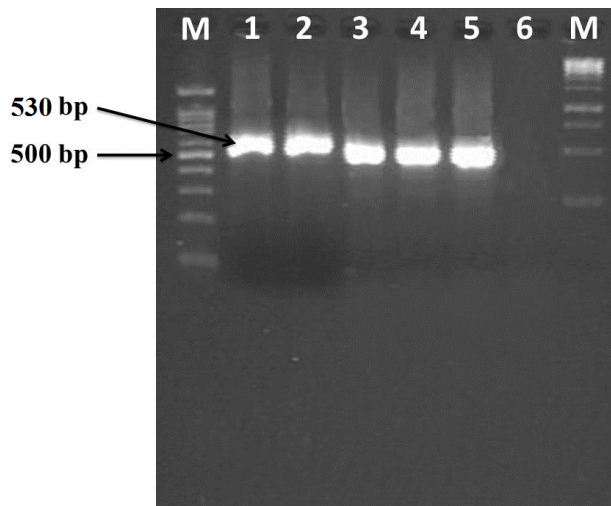
Note. \* $P < 0.05$  represents a statistically significant difference.

The statistical analyses of results indicated that there were no significant relationships between gender, age, living place, job, education level, and intestinal parasites in cancer patients undergoing chemotherapy ( $P > 0.05$ ) although intestinal parasitic infection in these patients was significantly related to the duration of chemotherapy ( $P < 0.05$ ).

## Discussion

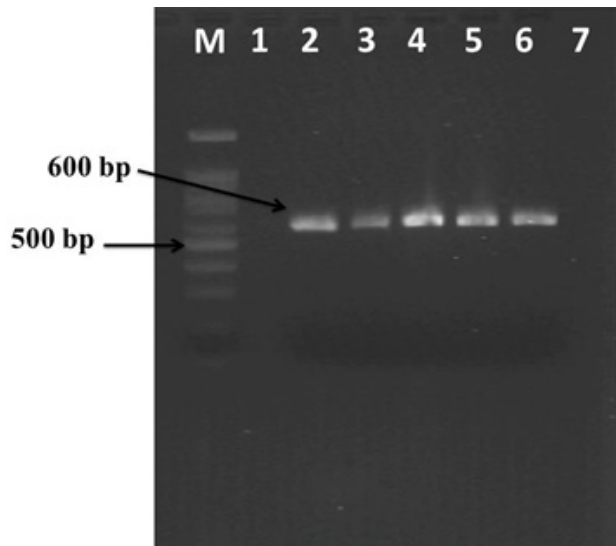
Cancer is one of the highly severe diseases of humans that have a major socioeconomic impact on societies worldwide. More than half of all cancers occur in developing countries such as those located in South America and Asia, and nearly three-quarters of these people live in low- and middle-income countries. Further, the rate of cancer

survivors in developing countries is often one-third of those living in developed countries.<sup>12,13</sup> Annually, 9 million new cancer cases are reported across the world, including 4 and 5 million in developed and developing countries, respectively. Furthermore, more than 1.2 million Americans are affected by cancer each year, and more than 56 000 of them die due to malignancy in the United States. It is predicted that a higher percentage of deaths in the world will be caused by non-communicable diseases in the near future and cancer will account for about 13% of all deaths. It is also predicted that cancer will be the first and the leading cause of human death by 2030.<sup>10,14</sup> The most common cancers in the world are lung, stomach, and breast, respectively. Moreover, the most prevalent types in Iran are skin, breast (in women), gastric and prostate (in



**Figure 2.** Electrophoresis Results of the Specific PCR Products of the Direct Smear Samples of *Giardia lamblia* Positive Samples in a 1.5% Agarose Gel.

Note. PCR: Polymerase chain reaction. M: 100 bp marker (Column 6: Negative control; Column 1: positive control; Columns 2-5: Patients' sample 530 bp, 6: Negative control).



**Figure 3.** Electrophoresis Result of the Specific PCR Products of the Direct Smear Samples of *Blastocystis hominis* Positive Samples in a 1.5% Agarose Gel.

Note. PCR: Polymerase chain reaction. M: 100 bp marker (Column 1: negative control; Column 2: Positive control; Columns 3-6: Patients' sample 600 bp).

men), bladder, colorectal and esophageal cancers.<sup>10,13,15</sup>

Chemotherapy, radiotherapy, immunotherapy, and surgery are therapeutic interventions in the cancer treatment that have helped increase the life expectancy of cancer patients and, in some cases, have led to complete remission.<sup>16</sup> One of the main agents in the pathogenesis, progress, and treatment of different cancers is the status of the immunity system of patients.<sup>10</sup> The immune system of cancer patients is suppressed by a variety of factors including cancer itself (especially, the malignancies of B and T lymphocytes), spreading of malignant cells into other tissues such as bone marrow, high-dose

chemotherapeutic agents, and corticosteroids.<sup>12</sup> The suppression immunity of humans by different agents such as chemotherapeutic drugs is placing the person at the risk of infection by a wide variety of opportunistic agents such as parasites that may cause a potentially lethal disease in this population.<sup>13</sup> Opportunistic parasitic infections including worms and protozoa cause mild or asymptomatic diseases in immunocompetent people although they can cause fatal diseases in immunocompromised patients. Additionally, immunosuppression in cancer patients, which is due to chemotherapy, may trigger latent intestinal parasitic infections. Therefore, parasitic infections are one of the common health problems in cancer patients.<sup>14</sup> Various studies have focused on the prevalence of parasitic infections in cancer patients, indicating different results affected by some factors such as the number of patients, the type of malignancy, the immunocompromising effects of the chemotherapy, the geographical climate status of the study area, the socioeconomic and hygiene status of a society, and the use of different diagnostic methods with different sensitivity levels.<sup>17-20</sup> Based on the findings of epidemiological studies on cancer patients for assaying intestinal parasitic infections, the rate of these infections varies from 12.9% to 88.9% in different areas of the world.<sup>13,16,17,19-23</sup> Various reports are available regarding the prevalence of intestinal infections among immunocompromised people in Iran while information about these infections on cancer patients is rare in comparison with other immunocompromised groups such as human immunodeficiency virus/acquired immunodeficiency syndrome (HIV/AIDS), organ transplant, and hemodialysis patients. However, the results of several studies conducted in Iran showed that the prevalence of intestinal parasite infections among cancer patients under chemotherapy varies from 4.16% to 35.9%.<sup>5,18,24-29</sup> On the other hand, the current widespread use of chemotherapy has altered the pattern of some parasitic infections so as they have become one of the most important causes of the global risk of health and probably the cause of an increase in the rate of morbidity and mortality in immunocompromised patients.<sup>30,31</sup> This cross-sectional descriptive study assessed the epidemiological state of intestinal parasitic infections among cancer patients undergoing chemotherapy in Shahrekord, Iran. The results of our study demonstrated that 12 (4.8%) out of 250 cancer patients were infected by two common protozoans, including 7 (2.8%) and 5 (2%) patients who were infected with *B. hominis* and *G. lamblia*, respectively. Similarly, the prevalence of intestinal parasitic infections in cancer patients was reported as 4.16% in Guilan (Northeast Iran),<sup>18</sup> 6.7% in Kashan and Qom (Central Iran),<sup>28</sup> 10% in Ardabil (Northwestern Iran),<sup>5</sup> 24.8% in Isfahan (western Iran),<sup>26</sup> 16.3% in Hamaden (Southwest Iran),<sup>29</sup> 19.8% in Urmia (Northwest Iran),<sup>32</sup> 25.9% in Tehran (Northwestern Iran),<sup>27</sup> and 35.9% in Mashhad (Northeast Iran).<sup>24</sup> The

findings of some of these investigations are in line with our results while those of some other studies contradict our findings. In this regard, studies conducted in different areas of the world demonstrated variable results. For instance, Menon et al. showed that 42% of children with different types of cancer in Malaysia were infected with intestinal parasites.<sup>21</sup> In addition, Jeske et al reported that 61.6% of cancer patients undergoing chemotherapy in the south of Brazil were infected with intestinal parasites.<sup>23</sup> Another study conducted in India revealed that 16.5% of 1029 cancer patients were infected with intestinal parasites.<sup>20</sup> Likewise, Botero et al concluded that 32.4% of immunocompromised patients with and without gastrointestinal manifestations in Colombia, including individuals with acute lymphoid leukaemia, chronic myeloid leukaemia, HIV, and other immunocompromised conditions (principally haematological disorders), were infected with these parasites.<sup>33</sup> Concerning the relationship between parasitic infections and cancer or diseases with immune system deficiency, it is hypothesized that parasitic infections are highly prevalent in cancer patients and those undergoing chemotherapy. Several studies have shown that the prevalence of intestinal parasites in cancer patients taking immunosuppressive drugs is higher compared to healthy control groups.<sup>34,35</sup> In a study conducted by Monsef et al, the prevalence of intestinal parasites in patients with malignancy was lower than that of the general population, which may be due to the effect of the applied drugs in chemotherapy.<sup>29</sup> On the other hand, the results of some studies indicated that the prevalence of parasitic infections in these patients was not significantly different compared to other healthy people. For instance, Salehi et al. found that despite the reducing effect of chemotherapy-related drugs on the immune system, they may have a destructive effect on parasitic cells and reduce the parasites in these individuals as well. Furthermore, risk factors for the acquisition of parasitic infections are the same in both immunocompetent and immunosuppressed individuals although cancer patients may be less exposed to parasitic agents due to intensive care. Additionally, those undergoing chemotherapy may be less exposed to parasitic infections due to decreased activity and increased health care.<sup>25</sup> Environmental, exogenous, and endogenous factors, as well as individual factors, including genetic predisposition contribute to the creation and development of cancer. Therefore, people's occupation is one of the external environmental factors that can cause cancer.<sup>36</sup> The results of the present study showed that the rate of intestinal parasitic infections was higher in housewives in comparison with other jobs. This is mainly because most housewives are involved in cleaning, cooking, washing vegetables and fruits, shopping, and the like. Hence, exposure can be an important factor in parasitic infection. Moreover, Azizi et al investigated the relationship between parasitic infections and cancer and found the highest rate

of infection in housewives.<sup>26</sup> In the present study, the majority of infected patients were either illiterate or had an elementary level of education. This is in conformity with the results of other studies and is scientifically justified because people with low levels of education are more prone to various infectious and parasitic diseases due to the lack of knowledge about the methods of transmission and the lack of receiving necessary health educations. In another study, Niazi et al evaluated the prevalence of giardiasis among the staff of food processing and distribution centers in the economic zone of Pars-e Jonubi (South pars). According to their results, the highest parasitic infection was observed in people with a lower level of education compared to other groups.<sup>37</sup> Although cancer patients could be infected with different parasites, epidemiological studies on cancer patients indicated that most prevalent parasites were *B. hominis* and *G. lamblia* among these patients, which is consistent with the results of our study. In this regard, assessing the frequency of the intestinal parasites between children with lymphohematopoietic malignancy in Mashhad, Zabolinejad et al reported *G. lamblia* (18%), *Entamoeba coli* (6.7%), and *B. hominis* (5.6%) as the most prevalent parasites.<sup>24</sup> Additionally, Esteghamati et al detected intestinal parasites in cancer, organ transplant, and primary immunodeficiency patients in Tehran. According to their results, *B. hominis* (22.3%) was the predominant intestinal parasitic infection in cancer patients, followed by *G. lamblia* (2.3%) and *Dientamoeba fragilis* (1.1%).<sup>27</sup> *B. hominis* is a highly common infection that is not usually diagnosable by conventional laboratory methods. In addition, the reported prevalence of this parasite is less than its real rate. In a study conducted by Rahimi-Esboei et al, *B. hominis* was the most common parasitic infection in Mazandaran province (8.1%).<sup>38</sup> Moreover, the infection by this parasite is widespread in other provinces of Iran.<sup>39</sup> In a similar study by Amin et al in Mazandaran province, 44.68% of patients attending gastrointestinal clinics were infected with *G. lamblia* parasite, which was significantly higher than other infections.<sup>40</sup> However, other studies reported that the *G. lamblia* was the most prevalent protozoan compared to other parasitic infections. In a systematic review and meta-analysis study by Abasian et al, the prevalence of *G. lamblia* was estimated to be 14.7% (1.48-39.54%) in Iran, including 15.1%, 19.2%, and 6.7% among children under 10 years old, adolescents, and 20-30 year old adults,<sup>41</sup> which is consistent with the findings of studies conducted in western countries. The prevalence of *G. lamblia* in the study by Monsef et al was reported to be 5.8% in cancer patients,<sup>29</sup> which is in line with the reported rate in this study. *Cryptosporidium* is one of the important opportunistic parasites with cosmopolitan distribution that can infect wide hosts. Although this protozoan in immunocompetent individuals causes acute self-limiting disease, in immunocompromised patients, it

can be a potentially life-threatening disease with severe and chronic diarrhea, and therefore, many epidemiological studies investigated *cryptosporidium* in immunocompromised persons.<sup>42</sup> In this study, the samples of cancer patients were evaluated by the cold Ziehl Neelsen staining method but coccidian parasites were detected in any of the samples. By investigating the slides of stool samples by the hot acid-fast staining method, Salehi et al. reported that cancer patients in Tehran were not infected with *Cryptosporidium* spp. or *Isospora belli*. Contrarily, Berenji et al<sup>43</sup> and Zabolinejad et al<sup>24</sup> indicated that children with cancer undergoing chemotherapy in Tabriz and Mashhad were infected with *Cryptosporidium* spp.

### Conclusion

The cancer patients, because of the immunosuppressive effect of chemotherapeutic agents, are potentially exposed to infection with different microorganisms such as opportunistic parasites. In this study, the prevalence of intestinal parasites in cancer patients undergoing chemotherapy was low and the detected parasites were *B. hominis* and *G. lamblia*. However, these findings should not be contrary to the need for healthcare to prevent opportunistic parasitic infections in cancer patients. In addition, the occurrence of opportunistic parasites such as *Cryptosporidium* spp. in immunosuppressed patients could be a potentially fatal risk. However, physicians should consider intestinal parasites as an important causative risk in cancer patients. Hence, it is advised to perform periodically screening for cancer patients in order to detect intestinal parasites by specific methods with high sensitivity such as PCR.

### Conflict of Interest Disclosures

The authors declare that there is no conflict of interests regarding the publication of this article.

### Ethical Approval

This project was approved by Shahrekord University of Medical Sciences under the ethics code of IR.SKUMS.REC.1396.273.

### Authors' Contributions

BK and RA conceived and designed the study. SMB helped to collect the samples. RA and SMB performed laboratory works. BK, ES, and MMH analyzed the data, drafted the paper, and revised the manuscript for important intellectual content.

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