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Frequency of Intestinal Parasites in Patients with Diabetes

Maksut Şahin[1*], Zeynep Taş Cengiz[1], Ahmed Galip Halidi[2], Selahattin Aydemir[1], Abdurrahman Ekici[1]

[1]Van Yüzüncü Yıl University, Faculty Of Medicine, Department Of Parasitology, Van
[2]Mus Alparslan University, Bulanık Vocational School, Mus

ABSTRACT

This study aims to determine whether patients with diabetes are at risk for intestinal parasites.
The study included 200 diabetics and 150 healthy individuals without any chronic disease. Data such as age, gender, diagnosis, and patient laboratory test results were obtained from the hospital automation system. Stool samples were examined by native-Lugol, concentration, modified acid-fast, and trichrome staining methods.
Intestinal parasites were found in 27.5% of the diabetic patients and 14% in the control group; the difference between the two groups was statistically significant (p=0.001). Out of 200 patients, *Blastocystis hominis* was found in 44 (22%), *Cryptosporidium* spp. in one (0.5%), *B. hominis* and *Entamoeba coli* in five (2.5%), *Giardia intestinalis* in two (1%), *B. hominis* and *G. intestinalis* in one (0.5%), *Taenia* spp. in one (0.5%) and *Chilomastix mesnili* in one (0.5%). In the diabetic patient group, statistically significant differences were found between age (p=0.043), duration of diabetes (p=0.006), fasting blood glucose (p=0.028) and HbA1c value (p=0.01) and presence of intestinal parasites. No statistically significant differences were found between place of residence, biochemical values such as cholesterol and LDL, use of insulin and antidiabetic agents, presence of another disease such as hypertension, COPD, neuropathy, nephropathy and intestinal parasite positivity.
Patients with diabetes can be considered a risk group in terms of intestinal parasite infections. Furthermore, individuals with a history of diabetes of more than 10 years and poor glycemic control are at a higher risk for intestinal parasites.

Keywords: Diabetes, intestinal parasites, HbA1c, native-Lugol

Introduction

Intestinal parasitoses are very common in developing countries and cause high rates of morbidity and mortality in endemic areas (1).

According to WHO data, approximately 3.5 billion people, mainly children, are affected by intestinal parasitoses. It is estimated that 450 million people currently suffer from such infections (2, 3).

Extensive research has shown that the risk of disease-causing parasitic infection is increased in HIV (human immunodeficiency virus) positive patients, transplant patients and immunosuppressive drug users (4). The increase in immunocompromised or impaired individuals is recognised as a significant health burden worldwide. People with diabetes, an immunocompromised patient group, have a disease picture characterized by hyperglycemia caused by insufficient insulin secretion, impaired insulin action, or both. Both innate and adaptive immune responses are impaired in diabetic patients and recent studies have shown that these patients are more predisposed to certain infections (4, 5).

There is no comprehensive study on the prevalence of intestinal parasitic infections among DM patients in Türkiye. In this context, this study was conducted to reveal the presence of intestinal parasites in diabetic patients and the risk factors that may cause the emergence of these parasite infections in DM patients, such as diabetes type, age, duration of diabetes, and biochemical parameters.

Materials and Methods

Approval for this study was obtained from the Van Yüzüncü Yıl University Non-Interventional Clinical Research Ethics Committee (Decision No: 2022/10-10; 14.10.2022). Face-to-face interviews with patients and hospital records were examined, and it was questioned whether they had an additional disease or whether they had used

***Corresponding Author:** Dr. Maksut Şahin, Van Yüzüncü Yıl University, Faculty of Medicine, Department of Parasitology, Van
E-mail: maksutsahin@gmail.com, Phone: 0 (533) 269 25 12
ORCID ID: Maksut Şahin: 0000-0002-0875-4654, **Zeynep Taş Cengiz:** 0000-0002-5247-5644, **Ahmed Galip Halidi:** 0000-0002-1780-6671, **Selahattin Aydemir:** 0000-0002-0941-2779, **Abdurrahman Ekici:** 0000-0001-6034-513X
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immunosuppressive or antiparasitic drugs in the last one years. An informed consent form was obtained from individuals included in the study; those who did not consent were excluded.

Those who did not volunteer, those who had an immunosuppressive disease other than diabetes (such as chronic kidney failure, patients undergoing chemotherapy, cancer, leukemia, rheumatoid arthritis, organ transplantation, HIV/AIDS), those who used immunosuppressants or anti-parasitic drugs in the last one years, and those who did not have diabetes were not included in the study.

This study was conducted on a total of 200 diabetic patients examined or hospitalized in the Internal Medicine Polyclinics between November 2022 and April 2023; 150 individuals without any chronic disease served as the control group.

Of the diabetic patients, 87 (43.5%) were male, and 113 (56.5%) were female; 74 individuals (49.33%) in the control group were male, and 76 (50.66%) were female. The patients in the patient group were between the ages of 5-83, and those in the control group were between 1-88 years old.

Collection of Samples: Each individual in the diabetic and control groups was given a sterile wide-mouth plastic container. During their stay in the hospital, they were asked to collect 20-30 g of feces in a plastic container with a screw cap and immediately deliver it to the parasitology laboratory. Feces that were placed in an inappropriate container, mixed with foreign matter, kept, frozen or incubated were not evaluated.

Examination of Samples: The samples were examined macroscopically for consistency, colour and adult, larvae and rings of helminths before storage. Then, preparations prepared from faecal samples by native-Lugol method were examined under light microscope at X100 and X400 magnification for trophozoites and cysts/ocysts of protozoa, larvae and eggs of helminths. Immediately afterwards, all samples were prepared by flotation with saturated zinc sulphate solution, formol-ether precipitation and staining methods (6, 7). Three samples were taken from each person in order not to overlook possible parasites. In addition, preparations were prepared from all samples by trichrome staining method.

The preparations prepared by modified acid-fast staining method were examined under light microscope at X1000 magnification for the presence of *Cryptosporidium* spp., *Cyclospora cayetanensis* and *Cystoisospora belli* (6, 7). To detect *Enterobius vermicularis* eggs, samples were taken by cellophane tape method and examined under light microscope at X100 and X400 magnification (7, 8).

The preparations prepared by trichrome staining methods were examined under light microscope at X1000 magnification. The specimens examined under the microscope were evaluated according to the relevant references (7, 9).

Statistical Analysis: The categorical variables among the studied features were expressed as numbers and percentages. Two proportion Z test was used to compare for categorical variable ratios. In addition, the chi-square test was performed to determine the relationship between categorical variables. A statistical significance level of 5% was determined in the calculations, and the SPSS (version 26) and MINITAB (version 14) statistical software packages were used for calculations.

Results

Age of the patient group mean was ± SD: 53.71 ± 13.71; min-max: 5-83 years and the age of the control group mean ± SD: 34.84 ± 19.8; min-max: 1-88 years.

Intestinal parasites were detected in 55 (27.5%) diabetic patients and 21 (14%) individuals in the control group and the difference between the two groups was statistically significant (p=0.001). *Blastocystis hominis* was the most common parasite found in both groups. *B. hominis* was found in 25% of the patient group and 11.33% of the control group; the difference between the two groups was statistically significant (Table 1; p = 0.001). There were no significant differences between the patient and control groups in terms of *G. intestinalis* and *E. coli* positivity.

Of 200 diabetic patients, 44 (22%) had only *B. hominis*, one (0.5%) had only *Cryptosporidium* spp., five (2.5%) had *B. hominis* and *E. coli*, two (1%) had only *G. intestinalis*, one (0.5%) had *B. hominis* and *G. intestinalis*, one (0.5%) had only *Taenia* spp. and one (0.5%) had only *C. mesnili*. Of the 150 individuals in the control group, 13 (8.7%) had only *B. hominis*, three (2%) had *B. hominis* and *E. coli*, two (1.3%) had *G. intestinalis*, two (1.3%) had *E. coli* and one (0.7%) had *G. intestinalis*, *E. coli* and *B. hominis*. *Enterobius vermicularis* was not detected in any of the cellophane tape samples.

When evaluated in terms of risk factors, statistically significant differences were found between intestinal parasite positivity and HbA1c value (p = 0.01; <8 vs. ≥8), duration of diabetes

Table 1: Parasitoses Detected in the Patient Group and the Control Group

Groups and detected parasites	Parasite negative		Parasite positive		p
	n	%	n	%	
Patient group (n: 200)	145	72.5	55	27.5	
B. hominis	150	75	50	25	0.001
Giardia intestinalis	197	98.5	3	1.5	1.000
Entamoeba coli	195	97.5	5	2.5	0.440
Taenia spp.	199	99.5	1	0.5	
Cryptosporidium spp.	199	99.5	1	0.5	
Chilomastix mesnili	199	99.5	1	0.5	
Control group (n: 150)	129	86	21	14	
B. hominis	133	88.66	17	11.33	
G. intestinalis	147	98	3	2	
E. coli	144	96	6	4	

(p = 0.006; <10 vs. ≥10), fasting blood glucose (FBG) (p = 0.028; <200 vs. ≥200) and age (p = 0.043; ≤35 vs. >35) in the diabetic patient group. There was no statistical significance between the presence of parasites and gender, low HDL level (<50) and type of diabetes (Type 1 and Type 2) in the patient group. No statistically significant associations were found between the presence of intestinal parasitosis and place of residence, elevated cholesterol and LDL levels, use of insulin and antidiabetic agents, hypertension, chronic obstructive pulmonary disease (COPD), neuropathy and nephropathy (Table 2).

Discussion

Intestinal parasites are among the most important pathogens threatening public health; they can cause infections in individuals with suppressed or impaired immune systems in many ways. Diabetic patients, making up the immunocompromised patient group, are considered a risk group for some opportunistic parasites (3-5, 10).

Infections caused by opportunistic parasites are mild or asymptomatic in individuals with a healthy immune system; however, their course is more severe and may even be fatal in individuals with a suppressed immune system (11). In immunocompromised individuals, lymphocytes cannot respond adequately against other infectious agents and opportunistic parasites (12). The most common parasites causing morbidity and mortality in immunocompromised patients are *Cryptosporidium parvum*, *C.cayetanensis*, *Microspora*, *Cystoisospora belli*, *Giardia lamblia*, *Strongyloides stercoralis* and free living amoebas (11, 12). *E. coli*, *I. butschlii* and *C. mesnili*, which live commensally in humans (13), can be transmitted to humans through consumption of contaminated water or food. The high prevalence rate found in the present study indicates poor environmental sanitation and also insufficient compliance with cleaning rules (14, 15). In addition, although there is a general opinion that *E. coli*, *C. mesnili* and *I. butschili* are not pathogenic for humans, some studies have reported that *C. mesnili* can cause diarrhoea. Detection of *E. coli* in humans is important because it shows inadequate environmental cleanliness (16,17).

There is no study in Türkiye showing the prevalence of intestinal parasites in diabetic patients. In studies conducted in other countries, different results were obtained.

In a study conducted on diabetic patients admitted to Arba Minch Hospital in Southern Ethiopia, intestinal parasitosis was found with a rate of 19.5% (5). In a study conducted in Iran, it was reported that intestinal parasitosis was found in 26.3% of diabetic patients and 6.8% of the control group (p<0.05) and the most common parasite species was *B. hominis* (4). In a study conducted in Pakistan, it was reported that intestinal parasites were found in 94.5% and 78.2% of diabetic patients and control group, respectively, and there was a statistically significant difference between the two groups (p=0.02). In a study conducted in India (18), intestinal parasitosis was found in 13.6% of diabetic patients. In another study conducted in Iran, intestinal parasitosis was found in 24.4% of diabetic patients and 23.2% of the control group, and no significant relationships were found between parasite positivity and age and gender (19). In another study conducted in Iran, intestinal parasites were found in 15.6% of

Table 2: Intestinal Parasite Positivity According to Potential Risk Factors in Patients with Diabetes

Patient information and some blood parameters	Patients with diabetes (N: 200)			Patients with parasite (n: 55)		p values
	Risk factors	N	%	n	%	
Gender	Male	87	43.5	30	34.48	0.054
	Female	113	56.5	25	22.12	
Age	≤35	18	9	9	50	0.043
	>35	182	91	46	25.82	
	≤ 20	7	3.5	4	57.14	0.105
	>20	193	96.5	51	26.42	
Place of residence	Rural	45	22.5	12	26.66	0.886
	Urban	155	77.5	43	27.74	
Duration of diabetes	<10	143	71.5	31	21.67	0.006
	≥10	57	28.5	24	42.1	
Diabetes type	Type 1	11	7.33	6	54.54	0.062
	Type 2	189	92.66	49	25.92	
HbA1C	≤6	26	13.06	5	19.23	0.253
	> 6	173	86.93	50	28.9	
	<8	117	58.79	22	17.94	0.01
	≥8	82	41.2	33	41.47	
Cholesterol	>200	77	40.95	21	27.27	0.97
	≤200	111	59.04	30	27.02	
	<300	183	92.42	48	26.22	0.292
	≥300	15	7.57	6	40	
FBS	<200	148	74.74	34	22.97	0.028
	≥200	50	25.25	20	40	
	<126	72	36.36	16	22.22	0.214
	≥126	126	63.63	38	30.15	
LDL	<160	165	88.7	45	27.27	0.901
	≥160	21	11.29	6	28.57	
HDL	<50	101	60.11	33	32.67	0.084
	≥50	67	39.88	14	20.89	

N: total number of patients; n: number of patients with parasitosis; LDL: low-density lipoprotein; HDL: high-density lipoprotein; FBS: fasting blood sugar

diabetic patients and 10% of the control group (20). In a study conducted in Egypt, intestinal parasites were found in 25% of diabetic patients and 7% of the control group and the difference was statistically significant ($p<0.001$). In the study, no statistically significant relationships were found between intestinal parasitosis and place of residence and gender. It was found that the age group with the highest rate of parasite infection in diabetic patients was individuals over 10 years of

age ($p<0.003$) and infection was higher in type 1 diabetic patients than in type 2 diabetic patients and the difference was statistically significant ($p<0.001$) (21). In a study conducted in Kirkuk, Iraq, intestinal parasites were found in 82.81% of gastroenteritis patients and 17.86% of diabetic patients (22). In a study conducted in Nigeria, intestinal parasitosis was found in 18.7% of diabetic patients, whereas parasitosis was not found in the control group ($p=0.022$). In the study, a statistically significant relationship was reported between intestinal parasitoses and anaemia ($p=0.016$) and no statistical significance was observed in terms of type and duration of diabetes (23). In a study conducted by Sisu et al. in Ghana (24), intestinal parasitosis was found in 12.5% of diabetic patients. In the study, statistically significant associations were found between the presence of parasitosis and FBS ($p<0.0001$), duration of diabetes mellitus ($p=0.017$), type of treatment (metformin: $p=0.0101$), nephropathy ($p<0.0001$) and no history of visiting a dietician ($p<0.0001$). In a study conducted by Aourarh et al. (25), intestinal parasitosis was found in 48% of patients with diabetes and 10% of patients without diabetes ($p<0.001$). In another study conducted in Ethiopia on patients with diabetes mellitus, intestinal parasitoses were found in 19.2% (3). In a study conducted in Sudan, intestinal parasites were found in 20.6% of type-2 diabetes patients and 10.6% of the control group ($p = 0.017$) (26). In a study conducted in Cameroon, intestinal parasites were found in 10% of diabetic patients and 23.5% of the control group and the difference was not statistically significant (27).

In a study conducted by Nazlıgül et al. (28), intestinal parasitosis was found in 47% of diabetic patients and 55% of the control group, and no statistically significant relationship was found between parasites and diabetes and gender.

In a study conducted in Brazil, intestinal parasites were found in 64% of diabetic patients and a statistically significant relationship was found between type-2 diabetes and parasite detection (29). In a study conducted in India, intestinal parasites were found in 44.32% of diabetic patients and 63.68% of the control group (10).

In the present study, as in some studies mentioned above (4, 20, 21, 23, 25, 26, 30), higher rates of intestinal parasitosis were found in diabetic patients compared to the control group and a statistically significant relationship was found between parasite positivity and diabetes. In some studies (10, 19, 27, 28), statistically significant relationships were not found between diabetic group and control group.

In five studies (3, 5, 18, 22, 24) conducted on diabetic patients without a control group, the rate of intestinal parasitosis was found to be lower than in the present study. In the study conducted in Brazil (29), it was found to be higher compared to the present study.

In three studies (19, 21, 23) evaluating the relationship between sexes in terms of parasitosis in diabetic patients, no statistically significant relationship was found similar to the present study. As in the present study, two studies (21, 28) found statistically insignificant relationships between gender and the incidence of intestinal parasitosis. In one study (21), no statistically significant relationship was found between place of residence and parasite infection, similar to the findings obtained in the present study.

In a study (21), a higher rate of intestinal parasitosis was found in patients with type-1 diabetes compared to patients with type-2 diabetes and the difference was found to be statistically significant ($p<0.001$), and this result supports the result of the present study. In another study (23), the relationship between type of diabetes and parasite positivity was found to be statistically insignificant, whereas in another study (29), intestinal parasitoses were found at a higher rate in the group of patients with type-2 diabetes and the difference was found to be statistically significant. In one study (19), the relationship between age and the incidence of intestinal parasitosis was found to be statistically insignificant, while in another study (21), the highest rate of intestinal parasitosis was found in the group over 10 years of age ($p<0.003$). In a study conducted by Akinbo et al. (23), similar to the present study, a statistically significant relationship was found between age groups in terms of the frequency of intestinal parasitosis in diabetic patients ($p=0.043$; $p=0.047$).

In a study (24), unlike our study, a statistically significant relationship was found between diabetes duration (<5 years) and the presence of parasites. In the study, a statistically significant relationship was found between FBS levels (>11 mmol/L) and the presence of parasites, similar to the present study. In another study (23) was reported that there was no statistically significant relationship between the duration of diabetes and the presence of parasites.

In conclusion, according to the present study's findings, intestinal parasitoses should be considered a risk factor for patients with diabetes,

especially for those with high HbA1c levels and those who have had diabetes for more than 10 years. Based on these results, it would be appropriate to consider these parasites in diabetic patients. Furthermore, large-scale studies are needed to better understand the importance of intestinal parasitosis in diabetic patients.

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