



Some epidemiological feature of human visceral leishmaniasis in North Khorasan Province during 2010-2018

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Abstract

Background & Aims: Visceral leishmaniasis, a potentially fatal disease caused by *Leishmania infantum*, is prevalent in the Mediterranean region and the Middle East. In Iran, it is endemic in the Northwest, Southwest, Northeast, Southeast, and central regions, with sporadic occurrences elsewhere. This study aimed to ascertain the period prevalence, epidemiological characteristics, and the most common clinical symptoms and laboratory signs of visceral leishmaniasis in North Khorasan Province.

Materials & Methods: Over a nine-year period from 2010 to 2018, clinical records of visceral leishmaniasis cases were scrutinized to gather demographic and clinical data. Diagnosis was confirmed paraclinically through positive smears for Leishman bodies in bone marrow aspirates. Each patient completed a questionnaire.

Results: A retrospective analysis was conducted on 24 diagnosed patients. Reported case numbers varied annually, with prevalence ranging from 0.0 to 1.1 per 100,000, peaking in 2017 and reaching its nadir in 2011. The majority of patients were hailed from rural areas (83.3%), with age distribution spanning from 8 months to 62 years. Most patients (95.8%) were under 10 years old, and 66.7% were male. Predominant clinical symptoms and laboratory signs included fever (91.7%), splenomegaly (79.2%), anemia (83.3%), thrombocytopenia (58.3%), and leukopenia (54.2%).

Conclusion: The incidence of visceral leishmaniasis in North Khorasan Province surpasses the national average by threefold, indicating its significance as an endemic hotspot for the disease in Iran.

Keywords: Iran, North Khorasan, Pediatric infection, Vector-borne diseases, Visceral leishmaniasis, Zoonotic diseases

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Introduction

The leishmaniasis are a group of diseases caused by more than 20 species of the genus *Leishmania*, transmitted by phlebotomine sandflies. There are three

main forms of the disease: cutaneous leishmaniasis (CL), visceral leishmaniasis (VL), also known as kala-azar, and mucocutaneous leishmaniasis (MCL)(1). In 2018, CL and VL were endemic in 92 and 83 countries or territories, respectively (2).

VL is the most serious form of leishmaniasis with a 95% fatality rate among untreated cases. Globally, an estimated 50,000 to 90,000 new cases of VL occur annually and is characterized by irregular prolonged fever, weight loss, enlargement in the liver and spleen, in addition to anemia. The disease is exclusively transmitted by species of the genus *Phlebotomus* in the Old World and is caused by *Leishmania infantum* in the Mediterranean area and the Middle East (2).

Domestic dogs (*Canis familiaris*) are the most important reservoir of the disease in the Mediterranean region, and wild canines seem to have a potential role in the sylvatic transmission cycle of the disease causative agent in this region (3).

In Iran, VL has been reported in all 31 provinces up to 2010. The disease is endemic in the Northwest (4), Southwest (5), Northeast (6, 7), Southeast (8), and center (9) of the country and is considered as sporadic in other parts of the country (10). According to recent studies, the northwest and northeast of Iran had the highest VL seroprevalence in humans (10).

Based on studies conducted in the last three decades in VL endemic regions of Iran, *Ph. kandelakii* in northeastern regions, *Ph. tobbi* in the northern region, *Ph. perfiliewi transcucasicus*, *Ph. kandelakii*, and *Ph. tobbi* in northwestern region, and *Ph. keshishiani*, *Ph. major s.l.*, and *Ph. alexandri* in southern regions of Iran are acknowledged as probable or confirmed VL vectors (10).

Researchers have studied various aspects of VL in North Khorasan Province. Natural leptomonal infections have been reported in *Phlebotomus* (*Lar.*) *kandelakii* in Shirvan district (11). Some researchers have investigated *Leishmania* infection in the reservoir in this region and detected *L. infantum* and *L. tropica* in asymptomatic dogs as domestic reservoirs of the disease (12). In an investigation, anti-*Leishmania infantum* antibodies were detected in foxes and jackals. Furthermore, one fox and one jackal were parasitologically positive, and *L. infantum* was confirmed by sequence analysis (13).

Leishmaniasis is among the top 10 neglected tropical diseases (14). Therefore, this study was

conducted to determine the period prevalence of VL in North Khorasan Province, one of the disease endemic areas. Moreover, some epidemiological aspects of the disease and the most common clinical and laboratory manifestations of the disease in North Khorasan Province were investigated.

Materials & Methods

Study area:

This epidemiological study was conducted in North Khorasan Province, northeast of Iran, between 36°37'-38°17' N latitudes and 55°53'-58°20' E longitudes, covering an area of more than 28,434 km². The province is bordered by Turkmenistan in the north, Khorasan Razavi Province in the east and southeast, Semnan Province in the southwest and Golestan Province in the west. It includes 10 counties with a population of about one million inhabitants (according to the Civil Registration Organization of North Khorasan for 2023). The province has desert and mountainous areas and receives about 250 mm of rainfall annually. The city of Bojnurd is the center of the province (15).

Data collection and analysis:

Over a period of 9 years, from 2010 to 2018, the clinical files of VL cases were reviewed to gather statistics on demographic and clinical details. This study was conducted using data from hospitals and the health service registry of the Vice-Chancellor for Health, North Khorasan University of Medical Sciences, Bojnurd, Iran. Patients were diagnosed paraclinically through a positive smear for Leishman bodies in a bone marrow aspiration (BMA). All diagnoses were confirmed by a specialist physician. A questionnaire was completed for each patient, extracted from their medical records.

Results

We retrospectively evaluated 24 infants, children, and adults with VL in the North Khorasan Province during 2010 to 2018. The number of reported cases varied each year. The prevalence of the disease ranged from 0.0 to 1.1 per 100, 000 with the highest

prevalence in 2017 and the lowest in 2011 (Figure 1). Additionally, the best model that shows the trend of prevalence is a fifth-order equation, and the R-square

for this model is equal to 0.97, which indicates that it explains the observed trend well.

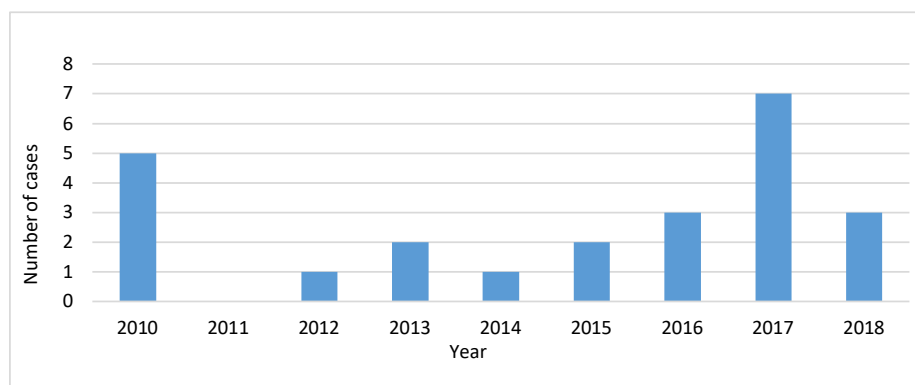


Fig. 1. Number of outpatient and hospitalized cases of visceral leishmaniasis during 2010-2018 in North Khorasan Province

The population of North Khorasan Province was 867,727 in 2011 (Census Report of Statistical Centre of Iran 2011) and 863,092 in 2016 (Census Report of Statistical Centre of Iran 2016). Therefore, we estimated the population of the province at 865,000 for the study period. The incidence rate of VL for this period was estimated at 0.31 per 100,000 population.

Demographic information and some of the clinical symptoms and laboratory signs are shown in Table 1. Up to 83.3% of patients were from rural areas. The patients were aged from 8 months to 62 years old. The majority of sufferers (95.8%) were under 10 years old. The overall mean age of the study population was 5.06 years (SD = 6.5 years) and 66.7% of them were male.

Table 1. Characteristics of 24 patients with visceral leishmaniasis during 2010-2018 in North Khorasan Province

Variables		Number (Percent)
Age	Less than 1 year	2 (8.33)
	1-4 years	18 (75)
	5-9 years	3 (12.5)
	10 and more	1 (4.17)
Gender	Male	16 (66.7)
	Female	8 (33.3)
Seasons	Spring	6 (25)
	Summer	10 (42)
	Autumn	3 (12)
	Winter	5 (21)
Residence	Urban	4 (16.7)

Variables		Number (Percent)
Districts	Rural	20 (83.3)
	Bojnurd	8 (33.3)
	Raz va Jargalan	5 (20.8)
	Manch	4 (16.7)
	Samalqan	4 (16.7)
	Shirvan	2 (8.3)
	Jajarm	1 (4.2)
Clinical symptoms	Fever	22 (91.7)
	Anemia	20 (83.3)
	Splenomegaly	19 (79.2)
	Hepatosplenomegaly	9 (37.5)
LAB signs	Thrombocytopenia	14 (58.3)
	Leukopenia	13 (54.2)
	CRP +++	6 (25)
	High ESR	6 (25)

Moreover, the results of hematological examination and clinical symptoms of some patients that were available are shown in detail in Table 2.

Fever was observed in 13 out of 14 patients (92.9%) and was the most common symptom in this

group of patients. Hepatosplenomegaly and/or splenomegaly (85.7%) were another common symptom of the disease. The most common laboratory signs were anemia (92.9%), leukopenia, and thrombocytopenia (78.6%).

Table 2. The results of laboratory blood test and clinical symptoms of patients with Visceral leishmaniasis during 2010-2018 in North Khorasan Province

Case number	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Gender	M	M	M	M	F	F	F	F	M	M	F	M	M	F
Age (month)	23	16	48	57	13	51	19	42	19	9	12	24	12	60
Fever	+	+	+	+	+	+	+	+	+	+		+	+	+
Hepatosplenomegaly			+	+			+		+					
Splenomegaly	+		+	+		+	+	+	+	+	+	+	+	+
Anemia	+		+	+	+	+	+	+	+	+	+	+	+	+
Thrombocytopenia	+	+	+			+	+	+	+	+	+	+	+	
Leukopenia	+	+		+		+	+	+	+	+		+	+	+
Erythrocyte														
Sedimentation Rate (ESR)				18	14	100					75	89	63	86
White blood cells	2.91	2.18	5.1	4.9	16.4	2.8	1.4	3.4	2.2	2.8	5.8	3.8	4.2	4.9

Case number	1	2	3	4	5	6	7	8	9	10	11	12	13	14
(WBC)														
Platelets	27000	38000	112000	178000	449000	73000	16000	119000	65000	41000	75000	95000	71000	271000
Hemoglobin	9	13	9.4	8.6	11.9	4.4	8.2	6.1	6.9	7.2	7.3	8.4	10	8.2
Aspartate														
aminotransferase	33	47	16	43		22		33			56	22	73	18
(AST)														

Discussion and Conclusion

VL is a fatal disease if not treated in over 95% of cases and is one of the diseases that is endemic in Mediterranean region and Iran.

Our finding indicated that North Khorasan Province is one of the important foci of VL in Iran. This has been shown by the incidence of the disease in this focus and the incidence proportion of the country during the last decade. The disease is more prevalent in Bojnurd and Raz va Jargalan, which are located in the northern part of the province. Since North Khorasan Province has more than 300 kilometers of border with Turkmenistan, the importance of these findings increases.

Based on the finding of this study, 24 people were infected with VL in 9 years, and the incidence rate of VL for this period was estimated at 0.3 per 100,000 populations in North Khorasan Province. Whereas, almost 100 new symptomatic cases of VL reported annually, and the incidence rate of VL during this period was to be estimated about 0.1 per 100,000 for Iran (10).

According to a previous study in this area, 164 patients with VL were reported from 1990 to 2010, for 20 years of investigation, and the prevalence of the disease from 1998 to 2008 ranged from 0.99 to 2.53 per 100, 000 which shows a significant decrease in disease incidence in this focus (6). This decrease in prevalence could be mainly attributable to a number of factors, including urbanization and reduction of rural depopulation, improvement in living conditions and infrastructure facilities in many villages and cities, and increased awareness and health knowledge among the population.

In the current study, only patients who were hospitalized at provincial hospitals were investigated and this is one of the limitations of the study, while in the previous study (6), all patients from inside and outside hospitals of the province were investigated and more than 30% of patients were diagnosed in hospitals outside the province.

In this study, all patients were diagnosed paraclinically by BMA, while in most investigations, serological, molecular, and BMA have been used. Direct microscopic detection of the parasite is the gold standard method for diagnosis of *leishmania*. Although the specificity of this method is high, sensitivity is not perfect. The sensitivity of BMA was evaluated to be 53%–86%, with the best performance (16). Moreover, the specificity of DAT decreases when its use is restricted to symptomatic patients (17). In research in Ardabil Province, out of 1,200 cases, only 277 cases (23%) were positive by BMA, and 77% were negative (18). These results show that we can expect an increase in disease cases in this region if other diagnostic methods were used.

In this study, 83% of patients infected with visceral leishmaniasis were children less than 5 years old, with an overall mean age of 5 years, while in the previous study, 72.5% of patients were under 5 years old with an overall mean age of 4.7 (6). In the southwest of Iran, 91.5% of the children involved were under 5 years old (5). In the northwest of Iran, 78.1% of children were under 5 years old (18). This finding is consistent with the result of our study.

Based on previous investigations, the disease has affected more men than women. Shirzadi reported 57% of visceral leishmaniasis cases in men (7). In our study,

67% of patients were male, and this finding is consistent with the results of other studies.

Sarkari showed that most cases of infection occurred in winter in the south of Iran (5). And in northeastern Iran, the most cases of infection were in winter and spring (19). In the north of Iran, the most cases of the disease were seen in spring and winter (20). In this study, the most cases of the disease were seen in summer and then in spring. This could be attributable to a delay in the diagnosis and treatment of the disease. The development of the direct agglutination test (DAT) as a simple and dependable diagnostic method would be a very useful method in the detection of the early stage of the disease.

According to an investigation in Tehran hospitals, 97.1% of patients had fever, 97.1% had splenomegaly, 97.1% had pallor and weakness, and 88.2% had hepatomegaly. Moreover, anemia (97.1%), thrombocytopenia (91.2%), and leukopenia (67.6%) were the most frequent laboratory abnormalities (21). A study in Ardabil reported 93.5% fever and 83.9% anemia in patients (22). According to research in Yemen, 100% of patients had fever. Moreover, 97% had splenomegaly, 92% had pallor, and 42% had hepatomegaly (23).

The most common signs and symptoms at an investigation in Türkiye were fever (88.9%), splenomegaly (100%), hepatomegaly (100%), anemia (92.4%), leukopenia (78.7%), and thrombocytopenia (76.2%) (24).

In a study in northwestern Iran, thrombocytopenia, anemia, and high ESR were observed as the most common signs and symptoms in patients (25). In a study in the southwest of Iran on 367 cases of visceral leishmaniasis, chronic fever, pallor, weight loss, abdominal distention, and hepatosplenomegaly were predominant clinical features (26). In a 15-year study on 380 patients, 64% had thrombocytopenia and 87.3% had anemia. In this study, visceral leishmaniasis patients had high ESR and positive CRP, as well as increased liver enzymes SGOT and SGPT (5). In a 23-year study in northwest Iran on 2824 patients, they also showed high ESR and positive CRP, increased liver

enzymes SGOT and SGPT, and decreased hemoglobin and platelets (18).

According to these investigations, the most common signs and symptoms are fever, anemia, splenomegaly, hepatomegaly, an increase in liver enzymes, a decrease in hemoglobin and platelets, high ESR and positive CRP. Moreover, VL can cause a variety of hematological disorders such as leukopenia, thrombocytopenia, and anemia which is consistent with the results of other studies.

In conclusion, the incidence of VL in North Khorasan Province is higher than the average incidence of the disease in Iran, and this area is an important endemic focus of visceral leishmaniasis in the country. The variation in the incidence of VL indicates neglect of this disease and highlights the necessity for more attention and additional studies in this area. The most common signs and symptoms of the disease in this area are in accordance with the findings of other studies in Iran and other countries in the Mediterranean region.

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Ethical statement

The study has been approved by the Ethics Committee of North Khorasan University of Medical Sciences (IR.NKUMS.REC.1398.015).

Conflict of interest

The authors declare that there is no conflict of interest.

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Data availability

The raw data supporting the conclusions of this article are available from the authors upon reasonable request.

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