ORIGINAL ARTICLE

Epidemiological and clinical characteristics of infants with visceral leishmaniasis hospitalized in Ardabil city Hospital during 2011–2016

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ABSTRACT

Background: Visceral leishmaniasis (VL) (Kala-Azar) is one of the most important health-related parasitic infectious diseases. VL is an endemic disease in Iran and more than 80 countries worldwide. The aim of this study was to investigate the Epidemiological and Clinical Characteristics of Infants with VL.

Methodology: This cross-sectional study was conducted on 31 patients with diagnosis of VL in Ardabil city hospital, Iran, during 2011–2016. The questionnaire included demographic information, such as age, gender, weight, place of living, level of family education, family history of diseases, and symptoms, to complete the specified goals.

Results: Of all the cases, 18 (58.1%) were girls and the rest were boys. The average age of the patients was 25.8 ± 32.3 months. More than half (61.3%) of the patients were in the Moghan County (Ghermi, Parsabad, Aslandooz). Clinical symptoms began in 10 (32.3%) patients in winter. Fever was the most commonly observed clinical symptom in children with VL (93.5%). Of 27 children with a history of symptom onset, 17 children (63%) were diagnosed at the interval of 2–4 weeks. The result of the direct agglutination test (DAT) was positive in 27 children (87.1%) and the rest were negative. In the clinical presentation of patients, fever, anemia, and enlargement of the spleen and liver were common.

Conclusion: The use of DAT in combination with clinical symptoms seems to be more suitable for initiating treatment than bone marrow aspiration in all the patients.

Keywords: Visceral leishmaniasis, epidemiologic, Kala-azar, Ardabil.

Introduction

Visceral leishmaniasis (VL), also known as kala-azar black fever, is one of the infectious-parasitic diseases that threatens life and in term of health is very important. Attention to VL as a public health problem has been increased. Based on large diversity of clinical forms and epidemiological situations of the disease, it represents that each focal point needs specific principles and procedures for control. VL is an endemic disease in Iran and more than 80 countries worldwide. According to the World Health Organization, 500,000 new cases are annually reported and of them, at least 50,000 are deaths that have occurred in the world. More than 90% of VL cases occur in the Indian subcontinent (India, Bangladesh, Nepal), Sudan, Ethiopia and Brazil [1–3].

Kala-azar in the rural areas is more common than urban areas. Kala-azar in Iran is of Mediterranean type that occurs more in infants and children under 12 years. In children under 5 years with immunodeficiency, the primary factor is *Leishmania infantum*, while in secondary the causing agent is *Leishmania tropica*, which especially

affects immunodeficient individuals. The conveyer of the disease is a special species of phlebotomizing and the parasite tank is dogs. The disease is mostly sporadic in the different regions but in some areas of Iran, especially in the northwest and southwest, including the provinces of Ardabil, East Azerbaijan, Fars, and Bushehr where it has been reported as endemic form and 100–300 new cases are reported annually [4–6]. From 1993 to 2012, about 2,000 cases of Kala-azar were reported in 31 provinces of Iran and 44.6% of them were in the north-west region. The average number of diagnosed cases in Iran was 0.449 per 100,000 and the highest prevalence of disease was in Ardabil with estimation of 57 cases per 100,000 [7–10].

The disease usually occurs without any specific symptoms and it progresses slowly. Its multiple clinical symptoms are an irregular and raging fever to 40°C with restlessness, spleen pain, cough and weight loss, enlargement of the spleen and liver; resulting in enlarged abdominal area, anemia, decrease in the number of blood cells (red blood cells (RBC), white blood cells (WBCs), and platelets), and swelling of the face, arms, and legs [9–11]. The illness of kala-azar because of a change in the color of the patient's skin and due to its high mortality, it is known as the black disease. The cause of death is usually caused by secondary infections and internal bleeding. Individuals with antibody titers against leishmaniasis greater than 1:3,200 is considered as a positive serology [direct agglutination test (DAT) Positive], approximately 75% patients have clinical symptoms and positive physical examination. The most common symptom in the first stage is pale appearance and a long fever, whereas in other stages the symptoms are splenomegaly, hepatomegaly, and lymphadenopathy [10–12]. Kala-azar is an infectious and systemic disease which if not treated, results in mortality and its early diagnosis is the key. On the other hand, Kala-azar is a complex disease because its clinical signs are similar to other common diseases, such as Malaria and typhoid, sometimes these diseases are seen with kala-azar. The most definite method for diagnosis is the observation of amastigotes in biopsy or samples of bone marrow but given the fact that the method is invasive and its sensitivity and specificity are not superior to serology, it seems the serology method (DAT) is good alternative [13–15]. The importance of this disease is due to the death of 100% of untreated patients and significant mortality in patients with immunosuppressive disorder and most childhood infections. In a study on the patients with kala-azar who were hospitalized in northwest of Iran, only 2.8% had mortality which was due to the delayed referral of patients to treatment center followed by secondary infections and constipation [14–16].

Climate and other environmental changes are potential factors for expanding the geographical range of carriers and the transfer of leishmaniasis in the future. Various weather and humidity conditions affect serology that is due to mosquito activity in the climate. It seems that the number of infected dogs in each region is the biggest risk factor in the transfer of kala-azar [15–18]. Due to

the importance of early and timely diagnosis of kala-azar and consequently, the treatment of it due to severe fecundity as well as climatic and environmental changes in different areas, and its effect on the spread of disease, the changes in the epidemiological pattern and clinical features of the disease were examined in the study.

Subjects and Methods

This descriptive cross-sectional study has been done on 31 patients with diagnosis of kala-azar, who were hospitalized in Ardabil city hospital, Iran, during 2011–2016. A questionnaire containing demographic information, such as age, gender, height, weight, residence place, family education, family history, and disease's symptoms, was completed for the patients and the collected data was analyzed by descriptive and analytical statistical methods in SPSS version 21.

Results

Of all children, 18 (58.1%) were girls and the rest were boys, 25 (80.6%) were under the age of 2 years, of which, 14 (56%) were girls and 11 (44%) boys. The average age of patients was 25.8 ± 32.3 months. Most of the patients (67.7%) lived in the Moghan area (Ghermi, Parsabad, Bilesavar) (Table 1). Of all children, 21 (67.7%) were in the rural and the rest were in urban. Clinical symptoms began in 10 (32.3%) patients in winter. The onset of clinical symptoms was in October and February, each with 5% (16.1%) which was more than other months (Figure 1). Fever was the most common observed clinical symptom in children with kala-azar (93.5%) (Figure 2). Of all children, 17 (63%) were diagnosed between 2 and 4 weeks of onset of clinical signs of VL. Among children with specific laboratory findings; 26 children (83.9%) had anemia, 16 (51.6%) leukopenia, and 15 (48.6%) thrombocytopenia (Table 2). The result of the DAT test was positive in 27 children (87.1%) and was negative in the rest. Out of 24 cases of bone marrow aspiration (BMA), in 14 cases (45.2%) there was evidence for confirmation of VL. False negative values in the BMA was 32.3% and in DAT was 12.9% (Table 3). In all 31 patients with kala-azar in this study, Glucantime was the first line drug for all the cases. Of these patients, 23

Table 1. Frequency of patients by city and sex.

City	Girl		Boy	
	n	%	*n*	%
Ardabil	4	22.2	2	15.4
Meshkin shahr	1	5.55	1	7.7
Moghan area	11	61.1	10	76.9
Astara	1	5.55	0	0
Khalkhal	1	5.55	0	0
Total	18	58.1	13	41.9

(74.1%) had appropriate response to the treatment and 8 (25.9%) had inappropriate response; one patient died before the beginning of secondary drug and remaining cases showed the appropriate response to the secondary drug. It should be noted that the secondary drug in seven patients that did not respond to Glucantime was Amphotericin B. Patients treated with Glucantime did not show significant complications. Of the seven patients who received amphotericin during their course of treatment, 6 (85.7%) developed hypokalemia and 1 (14.3%) hepatotoxicity.

Discussion

In the present study, 58.1% of the patients with VL were girls and the sex ratio was 1.4:1. In a study done by Molaie et al. [19], in Ardabil province during the years 1986–2009, the prevalence of VL was higher in males than females (1.3–1). Barati et al. [16] did not find any significant difference between the two sexes in term of anti-leishmaniasis antibodies. In a study by Mohebali [20], the prevalence of VL was 58% in males. In the present study, 35% of patients were under 1-year old, 80.6% were under 2-year old, and 87% were under 5 years of age. In a study by Molaie et al. [19], 93% of patients were less than 5 years of age and 17% were less than 1 year. It seems that failure to complete the body's immune system, the primary immune system, such as thin skin, defective production of antibodies, and defects in the quality of performance of B cell and T cells, can be the cause of high prevalence of Kala-azar in children under 2 years of age which requires separate examination. In this study, 67.7% of patients were from Moghan areas. Also, 67.7% of patients lived in the rural areas and the rest were from the Urban areas. Based on studies by Mccall [14] and Mohebali [20], Ardabil province is one of the endemic areas in terms of prevalence of VL. In a study by Molaie et al. [19], 66.1% of VL cases were children of Meshgin-shar city and 31% of them were in Moghan area, the climatic change in these is due to increase in the population of transporter and mosquitoes in the soil or reservoir due to the growth of agricultural projects, this requires ecological and environmental studies of reservoirs and carriers that necessitates the induction of endemic studies for province's health and control in Moghan region. In the present study, the most frequent time for the onset of clinical signs was winter (32.3%). In a study by Molaie et al. [19], the most frequent incidence of VL was in the end of winter and beginning of spring (67.7%). It seems that altering the endemic region of the disease and climate changes are effective for the disease occurrence. By investigating the studies by Molaie [19] and Mohebali [20], fever, anemia, and size of spleen and liver are common signs that were seen in patients with VL. In line with previous studies, fever, anemia, splenomegaly, and hepatomegaly were the most common clinical signs in the present study. Regarding this finding, doing a systemic examination and Complete blood count (CBC) screening and antibiotic arbitrary use in these patients are very effective in early detection. Laboratory evaluation of patients in the present study showed that decrease in hemoglobin, white blood cell count and platelet count were seen in 83.9%, 51.6%, and 50% of patients, respectively. N87.1% of the patients with VL had a positive DAT (27 cases of 31 patients). Out of 24 cases of BMA, 14 cases (58.3%) had confirmed information for the diagnosis of disease; in which four patients had positive DAT and two patients had positive BMA. According to the results, all the patients do not need BMA, it is only required in cases with suspected negative DAT. Due to the fact that DAT has false negative less than aspiration and since BMA is an invasive method and may be many of patients do not consent to doing this test and on the other hand, according to the early diagnosis of disease, it seems that DAT in combination with clinical symptoms to be more appropriate than BMA for starting treatment. In the present study, it has been emphasized that Glucantime is probably safer than amphotericin B, in terms of side effects but the secondary drug has more efficacy. However, the sample size in this study to investigate this claim is not enough and the confirmation requires more extensive clinical trials in the future.
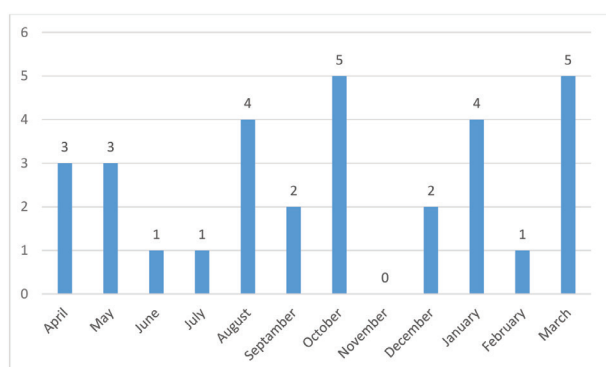
Figure 1. *Frequency of onset time of clinical symptoms by season and month.*
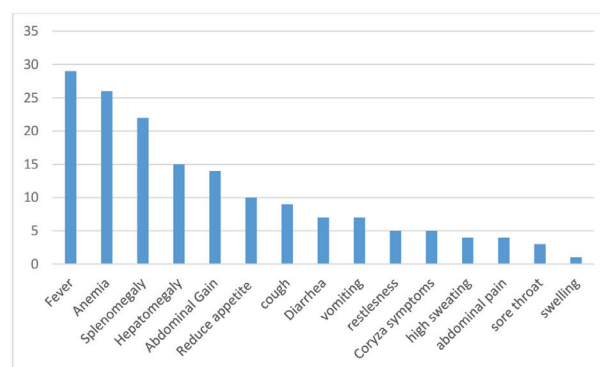
Figure 2. *Frequency of observed clinical symptoms in patients*

Table 2. Frequency of changes in laboratory finding.

Variables	Normal		Decreased		Increased	
	n	%	n	%	n	%
Hb	5	16.1	26	83.9	0	0
WBC	11	35.5	16	51.6	4	12.9
Platelet	15	48.4	15	48.4	1	3.2
Alanine aminotransferase (ALT)	12	38.7	0	0	19	61.3
Aspartate aminotransferase (AST)	8	25.8	0	0	23	74.1
Alkaline phosphatase (ALK.P)	8	25.8	0	0	23	74.1
erythrocyte sedimentation rate (ESR)	2	6.5	26	83.9	0	0

Table 3. DAT and BMA test results in patients.

BMA / DAT	Negative		Positive		None		Total	
	n	%	n	%	n	%	n	%
Negative	2	50	2	50	0	0	4	100
Positive	8	29.6	12	44.4	7	25.9	27	100
Total	10	32.3	14	45.2	7	22.5	31	100

Conclusion

Most of the patients with VL were females. Most cases of VL occurred in Ardabil province under the age of 2 years. The incidence of VL in the winter was higher than other seasons. VL is endemic disease in Ardabil and its prevalence in the Moghan area and the rural areas are higher than other areas. In the clinical presentation of patients studied in this study, fever, anemia, and the size of spleen and liver were common. The false negative cases in the DAT test were lesser than BMA, and in a significant percentage of patients performing BMA was not satisfactory. The use of DAT in combination with clinical symptoms was more appropriate to initiate treatment than BMA in all the patients. The results showed that glucantime was safer than amphotericin B for complications but amphotericin B was more effective.

List of Abbreviations
BMA   Bone marrow aspiration
DAT   Direct agglutination test
VL   Visceral leishmaniasis

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

Consent for publication
Informed consent was obtained from the patients.

Ethical approval
The study was ethically approved by Ardabil University of Medical Sciences.

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