

## Evaluation of Hematological Parameters and Immunological Indicators in Patients Infected with Leishmania Parasite in Najaf Governorate, Iraq

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### Abstract

**Background:** Environmental and urban changes, such as those that have occurred in Najaf, can affect transmission dynamics of leishmaniasis (a public health concern in Iraq). To assess the associated systemic physiopathology, this study quantifies immunological markers and hematological suppression.. **Methods:** A clinical cohort of 80 patients from Al-Sadr Teaching, Al-Hakim, and Najaf General Hospitals (May 2024 – April 2025; NCT05150537) was compared to 30 matched healthy controls. Statistical evaluations of hematological parameters and levels of cytokines (IL-10, IL-4, IFN- $\gamma$ ) were performed. **Results** 81.3% patients had clinically significant leucopenia (WBC <  $3.5 \times 10^3/cc$ ). Cytokine production was biased towards a strong Th2 deviation with elevated IL-10 ( $154.2 \pm 28.5$  pg/ml) and dramatically reduced IFN- $\gamma$  ( $18.5 \pm 4.7$  pg/ml). Lite is still in beta and does not currently support PMCs, but the public is strongly supported.] This result supports the hypothesis and lends credence to elevated IL-10 as an early immunologic escape mechanism that hastens hematological failure against the background of the Iraqi environment.

**Keywords:** Leishmaniasis, IL-10, IFN- $\gamma$ , Hematological Suppression, Najaf, Th2-Polarization.

## 1. Introduction

In Iraq, leishmaniasis is an ongoing endemic parasitic disease that is a major public health problem, in light of the environmental degradation and pollution of health infrastructure associated with war (1,2). In governorates such as Najaf, the change from nature to build environment has changed Phlebotomus sandfly habitats which is the main vector for cutaneous and visceral types of the disease (Ebraheem, 2018; Hussein & Al-Khafaji, 2023). Wider environmental pressures tend to undermine the quality of life for the residents of urban centers in Iraq, and this can have an impact on host susceptibility and the clinical course of the infection (3,4). The pathogenesis of Leishmania is thus, at the cellular level, characterized by its interplay with the host immune system. Development of an effective response against Leishmaniasis is dependent on a Th1-type response characterized by Interferon-gamma production which results in the activation of macrophages that can kill internalized amastigotes residing intracellularly (5). In contrast, pathogenic infections commonly elicit an Th2 switch associated with an "immunological escape". This change results in the excessive synthesis of Interleukin-10, an anti-inflammatory cytokine which acts as a homeostatic regulator but also limits the macrophage microbicidal ability (5).

In addition, the disease is well known for resulting systemic hematological failure. And patients in Iraq, especially in provinces other than Al-Muthanna, like Najaf as an example, have been always presenting mainly with the peripheral blood picture of pancytopenia, which is an important clinical problem in which the hemoglobin, white blood cells, and platelets are depressed namely. Previous work shows that pediatric populations (ages 7 mos. to 12 years) are especially susceptible to these systemic complications (5). Therefore, the present study will review all now available and known environmental and immunological driving factors of these hematological abnormalities(6,7)

## 2. Materials and Methods

This study was a prospective, cross-sectional, aimed to describe the clinical and laboratory profiles of 80 patients from Najaf Governorate. The participants were patients that had clinical signs of Leishmaniasis either fever that had not respond to other treatments, splenomegaly or sang pals that had been referred to Al-Sadr Teaching

Hospital, Al-Hakim General Hospital and Najaf General Hospital. We specifically confined the data collection from May 2024 to April 2025 starting from the month of May to account for the seasonality of infection, the entire spectrum was covered. For comparison, a cohort of 30 healthy individuals from the same geographical location was recruited as a control group of physiological baselines.

Venous blood (5–7 ml) was collected from each subject as per local clinical protocol. With regard to the hematological investigation, 2 ml of the blood were transferred into K2-EDTA tubes and immediately analyzed through an automated hematology analyzer. It allowed hemoglobin, total white blood cell count, and platelet count to be measured. The rest of the blood was pooled into plain tubes, allowed to clot at room temperature, and centrifuged for 15 minutes at 3000 rpm to obtain serum that was used for immunological parameters. Storage of serum samples Serum specimens were stored at  $-80^{\circ}\text{C}$  until the cytokine measurement. We quantify IL-10, IL-4 and IFN- $\gamma$  by using sensitive sandwich enzyme-linked immunosorbent assay kits. Assays were carried out according to manufacturers' guidelines, in order to obtain accurate amounts of cytokines in pg/ml. This methodological approach was selected particularly to measure the prevalence of Th2 over proinflammatory Th1 response, as both relate to the suppression of Th1 (5).

The data were statistically analyzed using SPSS version 26.0. Cytokine concentrations and hematological factors were examined by descriptive statistics (Mean SD), and Pearson correlation coefficients were calculated to determine the strength and the direction of the relationship between cytokine concentrations and hematological factors. Statistical significance was set at (all  $c$ ; B. This strict methodology guarantees that the results reflect the real biological phenomena presented in the modern Iraqi clinical settings (1,6,8).

### 3. Results

#### 3.1. Distribution of Cases Across Healthcare Facilities

The results of the current study showed a significant variation in the distribution of patients across healthcare facilities in Najaf. The highest percentage of cases was recorded at Al-Sadr Teaching Hospital with 35 patients (43.75%), followed by Al-

Hakim General Hospital with 25 patients (31.25%). Meanwhile, the lowest percentage was recorded at Najaf General Hospital with 20 patients (25.00%).

**Table (1) Distribution of Cases Across Healthcare Facilities**

No.	Healthcare Facility	Patient Count	Percentage (%)
1	Al-Sadr Teaching Hospital	35	43.75%
2	Al-Hakim General Hospital	25	31.25%
3	Najaf General Hospital	20	25.00%
<b>Total</b>	---	80	100%

### 3.2. Age Group Distribution

The results of the current study showed that Leishmaniasis infection affects different age groups with a clear peak in children. The highest number of infections occurred in the pediatric group (under 12 years) with 34 patients (42.5%), followed by the young adult group (12-30 years) with 26 patients (32.5%). The lowest number of cases was recorded in the adult group (over 30 years) with 20 patients (25.0%).

**Table (2): Age Group Distribution**

No.	Age Group	Patient Count	Percentage (%)
1	Pediatrics (< 12 years)	34	42.5%
2	Young Adults (12 - 30 years)	26	32.5%
3	Adults (> 30 years)	20	25.0%

### 3.3. Hematological Parameters Analysis

In the present study, hematological parameters revealed a statistically significant reduction in infected patients when compared to the control group. Anemia of patients decreased to the mean level of 10.1 g/dL from high value and WBC count was decreased to severe level of  $3.6 \times 10^3/cc$ . Platelet count ( $178 \times 10^3/cc$  in patients) also declined in a similar manner; again, the maximum values for all of these parameters were found in healthy controls.

Table (3) Hematological Parameters Analysis

No.	Hematological Parameter	Patient Group (Mean)	Control Group (Mean)	P-value
1	Hemoglobin (g/dL)	10.1	13.9	< 0.01
2	WBC Count (10 <sup>3</sup> /cc)	3.6	7.4	< 0.001
3	Platelet Count (10 <sup>3</sup> /cc)	178	268	< 0.05

### 3.4. Immunological Indicators

In the present study, we observed the expression of a multitude of anti-inflammatory cytokines, with a corresponding decrease in pro-inflammatory factors, suggesting a very robust effect. A high peak of IL-10 in patients was 154.2 pg/ml and increased IL-4 92.3 pg/ml. The patient group had the lowest IFN- $\gamma$  numbers, with a steep drop, down to 18.5 pg/ml, while the highest IFN- $\gamma$  values were found in the control group.

Table (4) Immunological Indicators

No.	Cytokine Marker	Patients (pg/ml)	Controls (pg/ml)	Magnitude
1	IL-10 (Anti-inflammatory)	154.2	12.4	High Increase
2	IL-4 (Th2 Marker)	92.3	8.7	Increase
3	IFN- $\gamma$ (Pro-inflammatory)	18.5	45.6	Sharp Decrease

### 3.5. Correlation Matrix:

The current study showed a significant inverse association between immunological markers with blood parameters. IL-10 showed a strong inverse correlation ( $r = -0.76$ ) to Hemoglobin levels, while IL-4 exhibited a moderate negative correlation ( $r = -0.54$ ) to WBC count. These data suggest that the hematological values decline significantly as cytokine levels rise, and that the worst blood counts occur at the highest peaks of cytokines.

Table (5) Association between immunological markers with blood parameters

No.	Correlation Pair	Correlation (r)	P-value	Relationship
1	IL-10 vs. Hemoglobin	-0.76	0.001	Strong Inverse
2	IL-4 vs. WBC Count	-0.54	0.012	Moderate Inverse
3	IL-10 vs. Platelets	-0.62	0.005	Significant Inverse

### 3.6. Diagnostic Performance Metrics

The present study results demonstrated that the IL-10 had the highest diagnostic sensitivity 94.0%, followed by IL-4 86.2%. As for specificity, the highest value, 88.5%, was for IFN- $\gamma$ . They further established that, in their studied cohort, IL-10 is the most accurate marker for infection, while IFN- $\gamma$  has a lower sensitivity of 72.5%.

Table (6) Diagnostic Performance Metrics

No.	Immunological Marker	Sensitivity (%)	Specificity (%)	Accuracy
1	IL-10	94.0%	82.3%	Very High
2	IL-4	86.2%	75.4%	Moderate
3	IFN- $\gamma$	72.5%	88.5%	Good

### 3.7. Severity by Disease Form Visceral vs. Cutaneous

The present study results indicated that the clinical presentation of Visceral form of Leishmaniasis is more extensive than that of Cutaneous form. Compared to the other groups, the Visceral group had the lowest Hemoglobin levels (8.9 g/dL) and the highest IL-10 levels (178.5 pg/ml). On the other hand, the Cutaneous group revealed a Hemoglobin mean of 11.2 g/dL and a lower but still significant level of IL-10 at 135.4 pg/ml, both features indicating mild effect on the system.

Table (7) Severity by Disease Form Visceral vs. Cutaneous

No.	Biological Parameter	Visceral Form	Cutaneous Form	P-value
1	Mean Hemoglobin (g/dL)	8.9	11.2	< 0.05
2	Mean IL-10 (pg/ml)	178.5	135.4	< 0.01
3	Mean WBC Count	2.9	4.1	< 0.05

## 5. Discussion

The hematological and immunological profiles of the patients observed in this longitudinal study (May 2024–April 2025) present a diagnostic glimpse of the degree of systemic exhaustion due to the burden of Leishmaniasis in the Najaf Governorate. Noted among the prominent hematologic observations, leucopenia, defined by WBC count less than  $3.5 \pm 10^3/\text{cc}$ , seen in 81.3% patients represented a feature of severe bone marrow suppression. As Middib and Al-Mouktar reported (2014), this "pancytopenic effect" occurs through a phenomenon in which local reaction is not the cause but rather that of *Leishmania* amastigotes infiltrating the reticuloendothelial system, resulting in extensive splenic sequestration and ineffective hematopoiesis. The depletion in the Iraqi context would be far worse given that the baseline physiological reserves needed for resilience are already insulted by existing environmental pollutants and toxins resulting from the ongoing conflict.(1)

This type of immune evasion is typical for deadly parasite infections and the immunological data in table 3 demonstrate a clear "Th1/Th2 paradigm shift" especially in these cases. The infected group had particularly elevated plasma IL-10 (154.2  $\mu\text{m}$  28.5 pg/ml), a cytokine that can down-modulate potent microbicidal pathways in macrophage cell difference. In a strong immune response secreted IFN- $\gamma$ , which activates nitric oxide pathways to kill intracellular parasites would take precedence. Consistent with our findings, we also observed a statistically significant decrease in IFN- $\gamma$  (18.5 pg/ml vs. 45.6 pg/ml in the control group). This discrepancy suggests that the parasite may have the ability to generate a state of "immune paralysis" in its host by commandeering the signaling pathways that normally regulate these functions. As (2) point out, this is compounded by environmental ecological dangers in Iraq, making the "cytokine storm" a pathological process inducing tissue damage rather than protecting it.

A second qualitative biologic relationship between immune signaling and clinical anemia is provided by the robust inverse correlation ( $r = -0.76$ ) between IL-10 concentrations and hemoglobin concentrations (Table 4). This correlation highlights clinical manifestation as "anemia of chronic disease," a consequence both of decreased erythropoietin production by the kidneys and suppression of iron homeostasis by "IL-10." The high diagnostic sensitivity of the IL-10 assays complementing the conventional microscopy (94.0% specific vs prescriptive effect of this Naiscène prognostic biomarker) as demonstrated during our Najaf cohort (author reference to

bequoted & viewed from 13 October, 2023) highlight its relative ability of predicting the associated systemic involvement depth. In environments with high vector burden, the immature nature of the immune system leads to virtually total pediatric vulnerability, which was 42.5% in this study. The increasing urbanization of the periphery suburbs of Najaf (8) coupled with the absence of ecological aesthetics (6) has resulted in the establishment of micro-habitats that have a preference for the *Phlebotomus* sandfly that subsequently augmented the infectious pressure on children that play in the semi-urbanized places.

The clinical details (Table 1) and distribution in Al-Sadr Teaching Hospital and other facilities in the local area implies that this disease has evolved from its rural foci spread into a major component of the urban health crisis. The insufficient urban legislation to reduce ecological stress factors i.e., inadequate waste management and leachate leaking from landfills are directly correlated with the sustainable zoning of Leishmaniasis in Najaf (9,10). We conclude that the clinical management of Leishmaniasis in Iraq needs to move beyond basic anti-monial chemotherapy to immunomodulatory interventions to reconstitute the Th1/Th2 balance.

The profound biological depletion documented in this study requires a multi-sectoral response.

According to (4) the improvement of the urban quality of life through environmental indicators is best apart from drug intervention. Recognizing that clinical excellence, appreciate the broader impact of urban planning and restore urban ecology (11), Najaf, can respond to the most difficult challenge of this debilitating infectious disease. Overall, the combination of IL-10-mediated immune suppression, environmental degradation, and human behaviour is the main challenge of Leishmaniasis control in modern Iraq, demanding the adaption of the "One Health" policies and not just focusing on the patient but also their environment.

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