Medicina & Laboratorio.

# Medicinay Laboratorio

Online ISSN- 2500-7106 | Print ISSN- 0123-2576
Available online on: https://medicinaylaboratorio.com/

Research Article

# Immunological Aspects of Parvovirus B19 Infection among Women with Threatened Abortion in Kirkuk Province

**Sara Hassan Ahmed[1]***

[1]Department of Medical Microbiology, Kirkuk Health Directorate, Iraq

OPEN ACCESS

**Corresponding Author:**

Dr. Sara Hassan Ahmed

Received: 06 May 2026
Revised: 21 June 2026
Accepted: 17 July 2026
Published: 28 August 2026

doi: 10.61336/ML.20263002.03



ABSTRACT

**Background:** Human parvovirus B19 (B19V) infection during pregnancy may cause fetal anemia, nonimmune hydrops, pregnancy loss and other adverse outcomes. Dysregulated inflammatory responses, particularly involving interleukin-6 (IL-6) and Tumor Necrosis Factor-alpha (TNF-α), may contribute to the pathogenesis of threatened abortion. **Objective:** To investigate B19V seroprevalence and inflammatory-marker concentrations among women with threatened abortion and evaluate their associations with hematological findings and short-term pregnancy outcomes. **Methods:** This case-control study included 90 pregnant women in Kirkuk City, Iraq: 45 women with threatened abortion and 45 healthy pregnant controls. Serum B19V IgM and IgG antibodies, IL-6 and TNF-α were measured using enzyme-linked immunosorbent assays. Complete blood count and Erythrocyte Sedimentation Rate (ESR) were also assessed. Demographic, obstetric, clinical and ultrasonographic data were collected. Group differences were evaluated using appropriate parametric and categorical tests and associations were expressed as Odds Ratios (ORs) with 95% Confidence Intervals (CIs). **Results:** B19V IgM positivity was significantly higher among cases than controls (24.4% versus 6.7%; OR = 4.53, 95% CI: 1.17-17.55; p = 0.039), whereas IgG seropositivity did not differ significantly. Cases had lower hemoglobin concentrations and platelet counts and significantly higher ESR, IL-6 and TNF-α levels. Mean IL-6 concentrations were 15.45±4.22 versus 9.14±2.95 pg/mL, while TNF-α concentrations were 24.78±6.88 versus 14.34±4.32 pg/mL in cases and controls, respectively (both p<0.001). Pregnancy loss (26.7% versus 6.7%; p = 0.021) and subchorionic hematoma (26.7% versus 4.4%; p = 0.007) were more frequent among cases. Among IgM-positive participants, 50.0% experienced pregnancy loss compared with 10.5% of IgM-negative participants. **Conclusion:** Recent B19V infection and increased systemic inflammatory activity were associated with threatened abortion and unfavorable early-pregnancy outcomes. Larger prospective studies incorporating molecular confirmation and follow-up through delivery are warranted.

**Keywords:** Parvovirus B19, Threatened Abortion, Pregnancy Loss, IL-6, TNF-α, Inflammation, Kirkuk

## INTRODUCTION

Threatened abortion, also referred to as threatened miscarriage, is characterized by vaginal bleeding, with or without lower abdominal pain, before 20 completed weeks of gestation in the presence of a closed cervix and a viable intrauterine pregnancy [1]. It is one of the most frequent complications of early pregnancy, affecting approximately 20% of clinically recognized pregnancies. Although many affected pregnancies continue normally, threatened abortion has been associated with an increased risk of miscarriage, preterm birth, preterm premature rupture of membranes, fetal growth restriction, placental abruption and low birth weight [2]. Its etiology is heterogeneous and may involve fetal chromosomal abnormalities, maternal anatomical or endocrine disorders, placental dysfunction, immune dysregulation, coagulation abnormalities, environmental exposures and maternal infection.

Human parvovirus B19 (B19V) is a small, non-enveloped, single-stranded DNA virus belonging to the family *Parvoviridae*. The virus exhibits a marked tropism for erythroid progenitor cells through its interaction with the erythrocyte P antigen, also known as globoside [3]. Viral replication in erythroid precursor cells can result in cellular destruction and temporary suppression of erythropoiesis [4]. Transmission occurs primarily through respiratory

droplets, although blood-borne transmission and vertical transmission from an infected mother to the fetus may also occur. Maternal infection is frequently asymptomatic or may cause only mild and nonspecific manifestations, including fever, malaise, rash and arthralgia. Consequently, maternal infection may remain unrecognized without appropriate laboratory investigation.

B19V infection during pregnancy is of particular clinical importance because transplacental transmission may produce severe fetal complications. Destruction of fetal erythroid precursor cells can cause profound anemia, tissue hypoxia, high-output cardiac failure, nonimmune hydrops fetalis and intrauterine fetal death. A systematic review and meta-analysis demonstrated that maternal B19V infection is associated with increased risks of fetal hydrops and fetal loss, particularly when infection occurs during early gestation [5]. Prospective evidence has also shown that the gestational timing of maternal infection substantially influences fetal morbidity and mortality, with the greatest vulnerability occurring during the first half of pregnancy, when fetal erythropoietic activity is particularly high [6]. Nevertheless, most maternal infections result in favorable pregnancy outcomes and serological evidence of infection alone does not establish that B19V is the direct cause of vaginal bleeding or subsequent pregnancy loss.

The diagnosis of recent maternal infection is generally based on B19V-specific Immunoglobulin M (IgM) and Immunoglobulin G (IgG) antibodies. Detectable IgM usually supports recent or acute infection, whereas IgG positivity without IgM generally indicates previous exposure and probable immunity. However, interpretation may be complicated by the timing of sample collection, delayed antibody production, prolonged IgM persistence, false-negative results and differences in the diagnostic performance of commercial assays [7]. Polymerase chain reaction detection of B19V DNA may provide additional diagnostic value when serological findings are negative, equivocal, or inconsistent with clinical and ultrasonographic observations [8].

Confirmed recent maternal infection requires careful fetal assessment rather than an assumption of inevitable fetal harm. Serial ultrasonography is used to identify fetal edema, ascites, cardiomegaly, placental abnormalities and other manifestations of hydrops. Doppler measurement of the fetal middle cerebral artery peak systolic velocity provides a valuable non-invasive method for identifying moderate-to-severe fetal anemia [9]. When severe anemia is confirmed, fetal blood sampling and intrauterine transfusion may be considered in specialized fetal-medicine centers. Routine universal screening of all pregnant women is not generally recommended; instead, testing is directed toward women with compatible symptoms, documented exposure, or suggestive fetal findings.

In addition to direct hematological effects, B19V infection may influence pregnancy through activation of maternal inflammatory pathways. Successful pregnancy requires precisely regulated immune adaptation that permits tolerance of the semiallogeneic fetus while maintaining an effective response against infectious agents. Interleukin-6 (IL-6) is a pleiotropic cytokine involved in implantation, placental development, acute-phase signaling, hematopoiesis and antimicrobial defense. Tumor Necrosis Factor alpha (TNF-α) contributes to normal immune regulation; however, excessive TNF-α production may promote endothelial activation, trophoblast apoptosis, coagulation and amplification of inflammatory responses [10].

Disturbance of the balance between pro-inflammatory and regulatory immune mechanisms may contribute to placental dysfunction and adverse pregnancy outcomes. Previous research has demonstrated alterations in maternal cytokine profiles among women with threatened miscarriage and has suggested that increased inflammatory activity may be associated with an unfavorable pregnancy outcome [11]. Nevertheless, circulating IL-6 and TNF-α concentrations are not specific to B19V infection. They may be affected by gestational age, maternal age, body mass index, other infections, chronic inflammatory conditions, medication use, sample handling and laboratory methodology. Maternal serum cytokine concentrations may also differ from immune activity occurring locally at the maternal-fetal interface.

Despite the potential relationship among B19V infection, inflammatory activation and adverse pregnancy outcomes, evidence integrating viral serology, hematological parameters, circulating cytokines, ultrasonographic findings and pregnancy outcomes remains limited in northern Iraq. Regional investigation is important because the prevalence of B19V exposure and its clinical consequences may vary according to demographic characteristics, socioeconomic conditions, population immunity, seasonal viral circulation and access to antenatal and fetal-medicine services.

Therefore, this case-control study aimed primarily to compare the prevalence of B19V IgM seropositivity between women with threatened abortion and apparently healthy pregnant controls in Kirkuk City, Iraq.

## MATERIALS AND METHODS

A hospital-based case-control study was conducted in Kirkuk City, Iraq, between 1 January and 30 June 2026. The study included 90 pregnant women divided equally into two groups. The case group comprised 45 women presenting to obstetric outpatient or emergency services with threatened abortion, defined as vaginal bleeding, with or without lower abdominal pain, before 20 completed weeks of gestation, in the presence of a closed cervix and a sonographically confirmed viable intrauterine pregnancy. The control group comprised 45 apparently healthy pregnant women

attending routine antenatal-care services without vaginal bleeding, abdominal pain, or other clinical evidence of threatened abortion. Controls were selected to achieve comparability with cases regarding gestational age.

**Study Population**

Pregnant women aged 18-42 years with a singleton pregnancy between 6 and 20 completed weeks of gestation were eligible for inclusion. The diagnosis of threatened abortion was established by an obstetrician based on clinical examination and ultrasonographic confirmation of fetal viability.

Women were excluded if they had a multiple pregnancy, ectopic or molar pregnancy, cervical dilatation, known fetal chromosomal or major structural abnormalities, autoimmune disease, chronic inflammatory disease, current immunosuppressive or corticosteroid therapy, chronic renal or hepatic disease, hematological disorders associated with chronic anemia, or a documented acute infection other than suspected human parvovirus B19 infection. Women who declined to provide written informed consent were also excluded.

**Data Collection**

Demographic, obstetric, clinical, laboratory and ultrasonographic information was collected using a structured questionnaire and data-recording form. The recorded demographic variables included age, residence, educational level, socioeconomic status, occupation, body mass index, ABO blood group and Rh status.

Obstetric information included gestational age, gravidity, parity, history and number of previous miscarriages, previous threatened abortion, antenatal-care attendance, previous stillbirth, previous preterm delivery and history of congenital fetal abnormalities. Clinical variables included vaginal bleeding, abdominal pain, fever during pregnancy, recent viral illness, contact with individuals with suspected or confirmed viral infection, history of blood transfusion, comorbid diseases and current medication use.

Laboratory measurements included complete blood count, erythrocyte sedimentation rate, serum anti-B19V IgM and IgG antibodies, Interleukin-6 (IL-6) and Tumor Necrosis Factor-alpha (TNF-α). Ultrasonographic findings included fetal cardiac activity, fetal viability, subchorionic hematoma, placental abnormalities and fetal hydrops. Pregnancy outcome was classified as continuation of a viable pregnancy or spontaneous pregnancy loss during the specified follow-up period.

**Specimen Collection and Processing**

Approximately 5 mL of peripheral venous blood was collected aseptically from each participant using sterile disposable equipment. Two millilitres were transferred into an ethylenediaminetetraacetic acid tube for hematological analysis. The remaining 3 mL were transferred into a serum-separator tube and allowed to clot at room temperature.

Serum was separated by centrifugation according to the validated laboratory procedure, divided into appropriately labeled aliquots and stored at −20°C until serological and cytokine analyses. Repeated freeze-thaw cycles were avoided to minimize possible degradation of antibodies and cytokines. All specimens were assigned coded identification numbers and laboratory personnel were blinded to participants' clinical groups whenever feasible.

**Hematological Investigations**

Complete blood count parameters, including hemoglobin concentration, total white blood cell count and platelet count, were measured using an automated hematology analyzer. The erythrocyte sedimentation rate was measured using the laboratory's standardized method. Internal quality-control materials were analyzed according to the manufacturer's recommendations before processing study specimens.

**Detection of Anti-Parvovirus B19 Antibodies**

Serum anti-human parvovirus B19 IgM and IgG antibodies were detected using validated commercial enzyme-linked immunosorbent assay kits in accordance with the manufacturer's instructions. All reagents and serum specimens were allowed to reach room temperature before analysis. Standards, controls and participant samples were processed under identical assay conditions.

Optical density was measured using a calibrated microplate reader at the wavelength specified by the kit manufacturer. Results were classified as positive, negative, or equivocal according to the manufacturer-provided cutoff values. Equivocal samples were retested using the same procedure.

IgM positivity was considered evidence suggestive of recent or acute B19V infection, whereas isolated IgG positivity with negative IgM was interpreted as previous exposure and probable immunity. Concurrent IgM and IgG positivity was interpreted as a recent infection or early convalescent immune response. Seronegative participants were defined as negative for both IgM and IgG.

The following information must be added from the kits used:

- Manufacturer and country
- Kit and catalogue numbers
- Lot numbers
- Analytical sensitivity and specificity

- Positivity and equivocal cutoffs
- Intra-assay and inter-assay coefficients of variation

**Measurement of IL-6 and TNF-α**

Serum IL-6 and TNF-α concentrations were measured using quantitative sandwich ELISA kits according to the manufacturer's protocols. Standards were prepared at the specified concentrations and a standard calibration curve was constructed for each analytical run. Serum samples, standards, negative controls and positive controls were tested in duplicate whenever sufficient sample volume was available.

Following the sequential addition of samples, detection antibodies, enzyme conjugates, chromogenic substrates and stopping solution, absorbance was measured using a calibrated microplate reader. Cytokine concentrations were calculated from the corresponding standard curves and expressed as pg/mL. Samples with concentrations above the upper analytical limit were diluted and retested using the dilution factor recommended by the manufacturer. Assay results were accepted only when the standard curve and quality-control measurements satisfied the manufacturer's validity criteria.

**Ultrasonographic Assessment and Follow-Up**

Transabdominal or transvaginal ultrasonography was performed by a qualified obstetrician or radiologist, depending on gestational age and clinical indication. Ultrasonographic evaluation documented gestational age, fetal cardiac activity, fetal viability, subchorionic hematoma, placental abnormalities and evidence of fetal hydrops.

Women with serological evidence of recent B19V infection were referred for specialist obstetric assessment and serial ultrasonographic surveillance. When clinically and gestationally appropriate, middle cerebral artery peak systolic velocity Doppler was used to assess the possibility of fetal anemia. Participants were followed until at least 20 completed weeks of gestation or until spontaneous pregnancy loss. Whenever available, subsequent pregnancy and fetal outcomes were obtained from medical records.

**Ethical Considerations**

The study protocol was reviewed and approved by the Kirkuk Health Directorate. Written informed consent was obtained from every participant before enrollment and specimen collection. Participant information was coded and treated confidentially. The study was conducted in accordance with the ethical principles of the Declaration of Helsinki.

**Statistical analysis**

Analyses were performed on the reproducible dataset. Continuous variables are summarized as mean±standard deviation and categorical variables as number (percentage). Welch's independent-samples t test compared continuous variables; Fisher's exact test compared categorical variables when expected counts were small. Associations are reported as Odds Ratios (ORs) with 95% Confidence Intervals (CIs). Spearman correlation assessed relationships among IL-6, TNF-α and ESR. Exploratory binary logistic regression evaluated selected predictors; model complexity was constrained by the number of events. Two-sided $p<0.05$ was considered statistically significant. No imputation was required because the dataset was complete.

## RESULTS

A total of 90 pregnant women were included, comprising 45 women with threatened abortion and 45 healthy pregnant controls. The groups were comparable regarding age, BMI, gestational age, gravidity, parity, residence, antenatal-care attendance and history of contact with an infected individual ($p>0.05$). A previous miscarriage was significantly more frequent among cases than controls (48.9% versus 15.6%, $p = 0.001$). Fever during pregnancy was also more frequent among cases, although the difference narrowly failed to reach statistical significance ($p = 0.051$), as presented in Table 1.

The serological profiles of the two study groups are presented in Table 2. B19V IgM positivity was significantly more frequent among women with threatened abortion than controls (24.4% versus 6.7%, $p = 0.039$). IgM-positive women had approximately 4.5-fold greater odds of belonging to the threatened-abortion group (OR = 4.53; 95% CI: 1.17-17.55). In contrast, B19V IgG seropositivity did not differ significantly between cases and controls (62.2% versus 53.3%, $p = 0.522$). The combined IgM-positive/IgG-positive profile was more frequent among cases, but this difference was not statistically significant.

Table 1: Baseline Demographic and Obstetric Characteristics

| Characteristic | Cases (n = 45) | Controls (n = 45) | p value |
|---|---|---|---|
| Age (years) | 29.52±5.07 | 28.19±4.62 | 0.197 |
| BMI (kg/m$^2$) | 26.05±3.45 | 25.54±3.28 | 0.470 |
| Gestational age (weeks) | 11.72±2.64 | 12.08±2.71 | 0.524 |
| Gravidity | 2.96±1.38 | 2.91±1.58 | 0.887 |
| Parity | 1.02±1.01 | 0.76±0.93 | 0.197 |
| Rural residence | 23 (51.1) | 15 (33.3) | 0.135 |
| Previous miscarriage | 22 (48.9) | 7 (15.6) | 0.001 |

| | | | |
|---|---|---|---|
| Regular antenatal care | 24 (53.3) | 33 (73.3) | 0.079 |
| Fever during pregnancy | 12 (26.7) | 4 (8.9) | 0.051 |
| Known contact with infected person | 15 (33.3) | 9 (20.0) | 0.233 |

Table 2: Parvovirus B19 Serological Profiles

| Serological profile | Cases n (%) | Controls n (%) | OR (95% CI) | p value |
|---|---|---|---|---|
| B19V IgM positive | 11 (24.4) | 3 (6.7) | 4.53 (1.17-17.55) | 0.039 |
| B19V IgG positive | 28 (62.2) | 24 (53.3) | 1.44 (0.62-3.34) | 0.522 |
| IgM+/IgG+ | 6 (13.3) | 1 (2.2) | 6.77 (0.78-58.73) | 0.110 |
| IgM−/IgG− | 12 (26.7) | 19 (42.2) | 0.50 (0.20-1.21) | 0.183 |

Table 3: Hematological and Inflammatory Parameters by Study Group

| Parameter | Cases (n = 45) | Controls (n = 45) | p value |
|---|---|---|---|
| Hemoglobin (g/dL) | 11.49±0.81 | 11.86±0.73 | 0.023 |
| White blood cells ($\times 10^3/\mu L$) | 8.13±1.30 | 7.72±1.54 | 0.174 |
| Platelets ($\times 10^3/\mu L$) | 240.35±34.96 | 262.43±36.72 | 0.004 |
| ESR (mm/h) | 26.27±7.57 | 16.98±6.27 | <0.001 |
| IL-6 (pg/mL) | 15.45±4.22 | 9.14±2.95 | <0.001 |
| TNF-α (pg/mL) | 24.78±6.88 | 14.34±4.32 | <0.001 |

Table 4: Biomarkers and Pregnancy loss According to B19V IgM Status

| IgM status | n | IL-6 (pg/mL) | TNF-α (pg/mL) | Hemoglobin (g/dL) | Pregnancy loss n (%) |
|---|---|---|---|---|---|
| B19V IgM positive | 14 | 18.59±4.89 | 30.87±5.78 | 11.61±0.98 | 7 (50.0) |
| B19V IgM negative | 76 | 11.14±3.82 | 17.47±6.11 | 11.69±0.76 | 8 (10.5) |

Table 5: Modeled Ultrasonographic Findings and Short-Term Pregnancy Outcomes

| Outcome | Cases n (%) | Controls n (%) | p value |
|---|---|---|---|
| Pregnancy loss | 12 (26.7) | 3 (6.7) | 0.021 |
| Subchorionic hematoma | 12 (26.7) | 2 (4.4) | 0.007 |
| Fetal hydrops | 1 (2.2) | 1 (2.2) | 1.000 |

Table 6: Exploratory Univariable Associations with Threatened-Abortion Case Status

| Candidate factor | OR | 95% CI | p value |
|---|---|---|---|
| B19V IgM positivity | 4.53 | 1.17-17.55 | 0.039 |
| Previous miscarriage | 5.19 | 1.92-14.06 | 0.001 |
| Fever during pregnancy | 3.73 | 1.10-12.64 | 0.051 |
| Known infected contact | 2.00 | 0.77-5.21 | 0.233 |
| IL-6 (per SD) | 14.08 | 4.91-40.44 | <0.001 |
| TNF-α (per SD) | 19.71 | 5.51-70.46 | <0.001 |

Table 3 compares the hematological and inflammatory parameters between the study groups. Women with threatened abortion had significantly lower hemoglobin concentrations and platelet counts than controls ($p = 0.023$ and $p = 0.004$, respectively). Conversely, ESR, IL-6 and TNF-α concentrations were significantly elevated among cases (all $p<0.001$), indicating greater systemic inflammatory activity. No significant difference was observed in the total white blood cell count ($p = 0.174$).

Values are mean ± SD. P values are from Welch independent-samples t tests. ESR, erythrocyte sedimentation rate.

Table 4 and Figure 1 presents the biomarker concentrations and pregnancy outcomes according to B19V IgM status. IgM-positive participants exhibited higher mean IL-6 and TNF-α concentrations than IgM-negative participants. Hemoglobin concentrations were broadly similar between the two serological groups. Pregnancy loss occurred in 50.0% of IgM-positive participants compared with 10.5% of IgM-negative participants. Because inferential *p* values and adjusted estimates are not provided in this table, these findings should be interpreted descriptively.

The ultrasonographic findings and short-term pregnancy outcomes are summarized in Table 5. Pregnancy loss was significantly more frequent among women with threatened abortion than controls (26.7% versus 6.7%, $p = 0.021$). Subchorionic hematoma was also significantly more frequent among cases (26.7% versus 4.4%, $p = 0.007$). Fetal hydrops was uncommon and occurred with equal frequency in the two groups (2.2% each, $p = 1.000$).

Table 6 summarizes the exploratory univariable associations with threatened-abortion status. B19V IgM positivity and previous miscarriage were significantly associated with increased odds of threatened abortion. Increasing IL-6 and TNF-α concentrations demonstrated the strongest associations, with ORs of 14.08 and 19.71 per standard-deviation increase, respectively (both $p<0.001$). Known contact with an infected individual was not significantly associated with case status. Fever during pregnancy demonstrated a borderline association that requires cautious interpretation.

Among the 45 women with threatened abortion, IL-6 showed a moderate positive correlation with TNF-α (Spearman's $\rho = 0.46$, $p = 0.002$) and a weaker positive correlation with ESR ($\rho = 0.38$, $p = 0.010$). TNF-α was also positively correlated with ESR ($\rho = 0.39$, $p = 0.008$). These findings indicate that increasing cytokine concentrations were accompanied by greater systemic inflammatory activity. In contrast, neither IL-6 nor TNF-α was significantly correlated with hemoglobin ($\rho = -0.05$, $p = 0.730$ and $\rho = 0.02$, $p = 0.916$, respectively). ESR demonstrated a weak inverse

correlation with hemoglobin that approached, but did not reach, statistical significance (ρ = −0.28, p = 0.058). Overall, the results suggest coordinated inflammatory activation among threatened-abortion cases, without a clear direct relationship between circulating cytokine concentrations and hemoglobin levels (Figure 2).

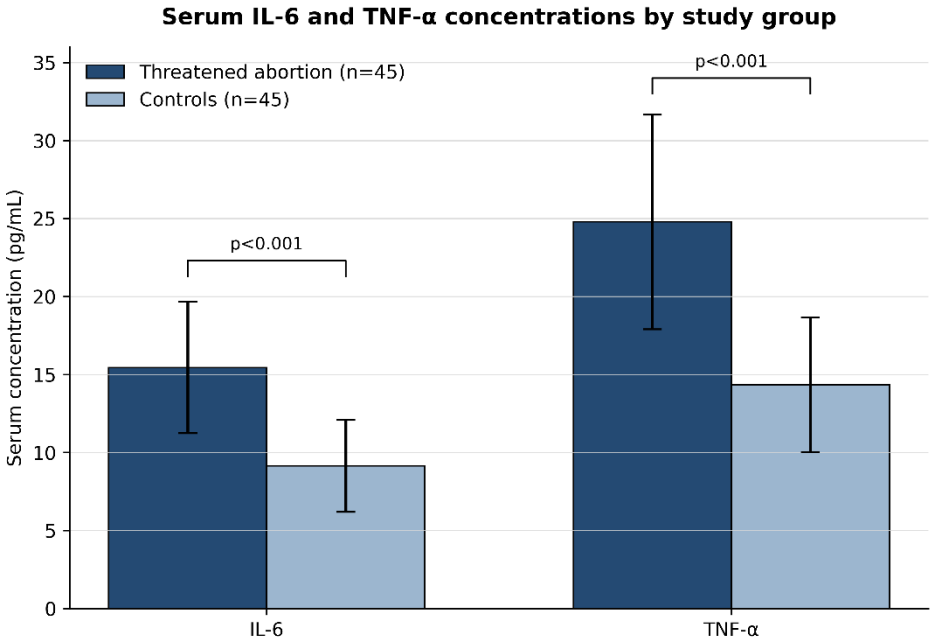


Figure 1: Serum IL-6 and TNF-α Concentrations among Women with Threatened Abortion and Healthy Pregnant Controls

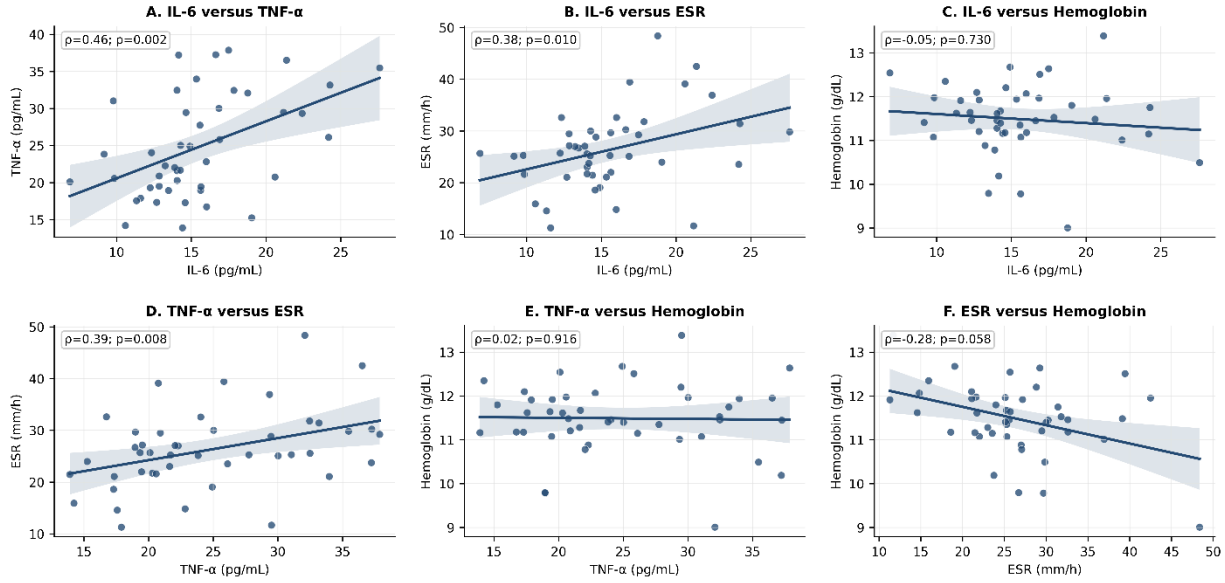


Figure 2: Correlations among Inflammatory and Hematological Parameters in Women with Threatened Abortion

## DISCUSSION

The present case-control study demonstrated three principal findings. First, B19V IgM positivity was significantly more frequent among women with threatened abortion than among healthy pregnant controls. Second, women with threatened abortion exhibited a pronounced inflammatory profile characterized by higher serum IL-6, TNF-α and ESR concentrations, accompanied by modest reductions in hemoglobin and platelet counts. Third, pregnancy loss and subchorionic hematoma were more frequent among cases, while B19V IgM-positive participants exhibited particularly high inflammatory-marker concentrations and a greater frequency of pregnancy loss. These findings indicate a possible relationship between recent B19V infection, systemic inflammatory activation and adverse early-pregnancy outcomes. B19V IgM positivity was detected in 24.4% of cases compared with 6.7% of controls, corresponding to

approximately 4.5-fold higher odds of threatened abortion. This finding is consistent with a systematic review and meta-analysis showing that maternal B19V infection increased the risks of fetal loss, spontaneous abortion and stillbirth [1]. A population-based Danish study similarly found that first-trimester B19V IgM positivity was associated with a 71% increase in fetal-loss risk [2]. Large prospective studies have demonstrated that fetal complications occur predominantly when maternal infection develops during the first 20 weeks of gestation [3,4]. Nevertheless, the relatively wide confidence interval observed in the present study indicates limited statistical precision resulting from the small number of IgM-positive participants. The association between B19V infection and threatened abortion is biologically plausible. B19V demonstrates a marked tropism for erythroid progenitor cells through its interaction with globoside, also known as the P antigen [5]. Viral replication within erythroid precursors causes cell-cycle arrest and apoptosis, leading to a temporary interruption of erythropoiesis [6]. Although this transient suppression is usually tolerated by immunocompetent adults, the fetus is particularly vulnerable because of rapid erythroid expansion, a short erythrocyte lifespan and increasing oxygen requirements. Infection may therefore produce profound fetal anemia, tissue hypoxia, high-output cardiac failure, placental edema, nonimmune hydrops and intrauterine death [7-9]. Hemoglobin concentrations were significantly lower among cases than controls. This observation may be clinically relevant because B19V directly affects erythroid-cell production. However, maternal hemoglobin should not be considered a direct surrogate for fetal anemia. Maternal bleeding, iron deficiency, nutritional status, gestational hemodilution and pre-existing anemia may also influence hemoglobin concentrations. The absence of ferritin, transferrin saturation, reticulocyte count, vitamin B12 and folate measurements limits the interpretation of this result. Similarly, the lower platelet count among cases may reflect inflammatory activation, increased consumption, hemodilution, or unrelated biological variation rather than a specific effect of B19V infection. Pregnancy loss occurred in 26.7% of cases compared with 6.7% of controls. Moreover, pregnancy loss was recorded in 50.0% of B19V IgM-positive women compared with 10.5% of IgM-negative women. These results are directionally consistent with evidence that adverse outcomes are more frequent when maternal infection occurs during early or mid-pregnancy [10]. Previous investigations have shown that gestational B19V infection may cause both hydropic and non-hydropic fetal death [11]. Viral DNA has also been detected in cases of intrauterine fetal death, supporting the potential involvement of B19V even when hydrops is absent [12]. However, the pregnancy-loss rate in the present study was higher than that reported in many population-based investigations, probably because the cases were symptomatic women who had already presented with threatened abortion rather than unselected pregnant women with B19V infection.

Fetal hydrops was uncommon and occurred with equal frequency in cases and controls. This finding should be interpreted in light of the small sample and the rarity of hydrops. The absence of a group difference does not exclude B19V as a potential cause because hydrops may develop several weeks after maternal infection and may be missed when follow-up is short. Contemporary reviews emphasize that maternal infection may progress to fetal anemia before ultrasonographic features of hydrops become evident [13]. When severe fetal anemia is detected, intrauterine transfusion can improve survival, particularly when undertaken in specialized fetal-medicine centers [14]. Middle cerebral artery peak systolic velocity Doppler assessment provides a noninvasive method for identifying moderate-to-severe fetal anemia and is central to the surveillance of pregnancies at risk [15]. Another important finding was the substantial elevation of IL-6 and TNF-α among women with threatened abortion. Pregnancy requires a carefully regulated immune environment that permits antimicrobial defense while maintaining maternal tolerance toward fetal and placental tissues. IL-6 and TNF-α participate in implantation, trophoblast differentiation, vascular adaptation, inflammatory signaling and host defense [16,17]. However, excessive cytokine production may disrupt this balance, promote endothelial activation and coagulation, increase oxidative stress and impair trophoblast survival. Dysregulated maternal and fetal inflammatory responses have been implicated in miscarriage, placental dysfunction, fetal injury and preterm birth [18]. Recent evidence has also associated increased circulating pro-inflammatory cytokines, particularly TNF-α, with unfavorable outcomes following threatened abortion [19]. The correlation findings support the biological coherence of the inflammatory profile. Among cases, IL-6 showed a moderate positive correlation with TNF-α and a weaker positive correlation with ESR. TNF-α was also positively correlated with ESR. These relationships suggest coordinated activation of different components of systemic inflammation. In contrast, neither IL-6 nor TNF-α was significantly correlated with hemoglobin, indicating that cytokine elevation and reduced hemoglobin may represent partially independent processes. Previous studies of threatened miscarriage have reported alterations in TNF-α, IL-6, IL-10, interferon-γ and their relative ratios [20]. Nevertheless, results have not been completely consistent across investigations. Cytokine concentrations are affected by gestational age, maternal BMI, smoking, concurrent infection, medication use, specimen-processing time, storage conditions and analytical platform. In addition, systemic cytokine concentrations may not accurately reflect local immune activity at the decidual and placental interface. Reviews of recurrent pregnancy loss have emphasized that successful pregnancy depends on the balance between inflammatory and regulatory immune pathways rather than isolated elevation or suppression of one cytokine [21,22]. Therefore, IL-6 and TNF-α should presently be regarded as research biomarkers rather than independent diagnostic tests.

Interpretation of B19V serology also requires caution. Maternal IgM positivity generally indicates recent infection, but IgM may remain detectable for several months. Conversely, delayed specimen collection may produce an IgM-negative result after antibody concentrations have declined. Isolated IgG positivity usually indicates previous exposure and probable immunity, whereas concurrent IgM and IgG positivity may reflect recent infection or early convalescence. Equivocal or discordant results should be investigated by repeat serology, demonstration of IgG seroconversion, IgG-avidity testing, or quantitative B19V DNA PCR [23]. However, low-level DNAemia may persist beyond the acute phase; therefore, PCR results must be interpreted together with clinical findings, serological patterns and gestational timing [24].

The absence of a significant difference in IgG positivity suggests that cumulative previous B19V exposure was broadly similar between cases and controls. The clinically important distinction was recent infection, reflected by IgM positivity, rather than past exposure alone. Current professional recommendations do not support universal antenatal screening for B19V. Targeted testing is recommended for pregnant women with compatible symptoms, confirmed exposure, unexplained fetal anemia, nonimmune hydrops, or other suggestive ultrasonographic abnormalities. Women with confirmed acute infection should receive appropriate counseling and serial fetal surveillance [25].

### Strengths and limitations

The strengths of this study include its case-control design, contemporaneous control group, standardized clinical definitions and simultaneous assessment of serological, inflammatory, hematological and ultrasonographic parameters. Reporting odds ratios with confidence intervals also enabled evaluation of both the magnitude and precision of the associations.

Several limitations should be acknowledged. The relatively small, single-city sample limited statistical power and multivariable adjustment. Hospital-based recruitment may have introduced selection bias, while histories of symptoms and exposure were susceptible to recall bias. A single serological measurement could not precisely determine infection timing. The absence of paired serology, IgG-avidity testing, quantitative PCR, placental histopathology and fetal testing may have resulted in infection misclassification. Cytokine concentrations are nonspecific and may have been affected by unmeasured infections, BMI, medications, specimen storage and assay variability. Finally, limited follow-up may have missed later fetal anemia, hydrops, preterm birth and neonatal complications.

## CONCLUSION

B19V IgM positivity was significantly associated with threatened abortion and was accompanied by elevated IL-6, TNF-α, ESR and pregnancy-loss frequency. These findings support a potential relationship between recent B19V infection, maternal inflammatory activation and adverse early-pregnancy outcomes. Larger prospective studies incorporating paired serology, quantitative PCR, IgG-avidity testing, standardized cytokine measurements, comprehensive confounder adjustment and follow-up through delivery are required.

## REFERENCES

[1] Xiong, Y.Q. *et al.* "The risk of maternal parvovirus B19 infection during pregnancy on fetal loss and fetal hydrops: A systematic review and meta-analysis." *Journal of Clinical Virology*, vol. 114, 2019, pp. 12-20. doi:10.1016/j.jcv.2019.03.004.

[2] Lassen, J. *et al.* "Parvovirus B19 infection in the first trimester of pregnancy and risk of fetal loss: A population-based case-control study." *American Journal of Epidemiology*, vol. 176, no. 9, 2012, pp. 803-807. doi:10.1093/aje/kws177.

[3] Enders, M. *et al.* "Fetal morbidity and mortality after acute human parvovirus B19 infection in pregnancy: Prospective evaluation of 1018 cases." *Prenatal Diagnosis*, vol. 24, no. 7, 2004, pp. 513-518. doi:10.1002/pd.940.

[4] Miller, E. *et al.* "Immediate and long-term outcome of human parvovirus B19 infection in pregnancy." *British Journal of Obstetrics and Gynaecology*, vol. 105, no. 2, 1998, pp. 174-178. doi:10.1111/j.1471-0528.1998.tb10048.x.

[5] Brown, K.E. *et al.* "Erythrocyte P antigen: Cellular receptor for B19 parvovirus." *Science*, vol. 262, no. 5130, 1993, pp. 114-117. doi:10.1126/science.8211117.

[6] Young, N.S. and K.E. Brown. "Parvovirus B19." *New England Journal of Medicine*, vol. 350, no. 6, 2004, pp. 586-597. doi:10.1056/NEJMra030840.

[7] Dittmer, F.P. *et al.* "Parvovirus B19 infection and pregnancy: Review of the current knowledge." *Journal of Personalized Medicine*, vol. 14, no. 2, 2024, article 139. doi:10.3390/jpm14020139.

[8] Gigi, C.E. and D.O.C. Anumba. "Parvovirus B19 infection in pregnancy: A review." *European Journal of Obstetrics and Gynecology and Reproductive Biology*, vol. 264, 2021, pp. 358-362. doi:10.1016/j.ejogrb.2021.07.046.

[9] Lamont, R.F. *et al.* "Parvovirus B19 infection in human pregnancy." *BJOG*, vol. 118, no. 2, 2011, pp. 175-186. doi:10.1111/j.1471-0528.2010.02749.x.

[10] Ornoy, A. and Z. Ergaz, "Parvovirus B19 infection during pregnancy and risks to the fetus." *Birth Defects Research*, vol. 109, no. 5, 2017, pp. 311-323. doi:10.1002/bdra.23588.

[11] Enders, M. *et al.* "Risk of fetal hydrops and non-hydropic late intrauterine fetal death after gestational parvovirus B19 infection." *Journal of Clinical Virology*, vol. 49, no. 3, 2010, pp. 163-168. doi:10.1016/j.jcv.2010.07.014.

[12] Tolfvenstam, T. *et al.* "Frequency of human parvovirus B19 infection in intrauterine fetal death." *The Lancet*, vol. 357, no. 9267, 2001, pp. 1494-1497. doi:10.1016/S0140-6736(00)04647-X.
[13] Kagan, K.O. *et al.* "Prenatal parvovirus B19 infection." *Archives of Gynecology and Obstetrics*, vol. 310, no. 5, 2024, pp. 2363-2371. doi:10.1007/s00404-024-07644-6.
[14] Kosian, P. *et al.* "Intrauterine transfusion in 103 fetuses with severe anemia caused by parvovirus infection: A multicenter retrospective study." *Archives of Gynecology and Obstetrics*, vol. 308, no. 1, 2023, pp. 117-125. doi:10.1007/s00404-022-06712-z.
[15] Mari, G. *et al.* "Noninvasive diagnosis by Doppler ultrasonography of fetal anemia due to maternal red-cell alloimmunization." *New England Journal of Medicine*, vol. 342, no. 1, 2000, pp. 9-14. doi:10.1056/NEJM200001063420102.
[16] Yockey, L.J. and A. Iwasaki. "Interferons and proinflammatory cytokines in pregnancy and fetal development." *Immunity*, vol. 49, no. 3, 2018, pp. 397-412. doi:10.1016/j.immuni.2018.07.017.
[17] Mor, G. *et al.* "The unique immunological and microbial aspects of pregnancy." *Nature Reviews Immunology*, vol. 17, no. 8, 2017, pp. 469-482. doi:10.1038/nri.2017.64.
[18] Green, E.S. and P.C. Arck. "Pathogenesis of preterm birth: Bidirectional inflammation in mother and fetus." *Seminars in Immunopathology*, vol. 42, no. 4, 2020, pp. 413-429. doi:10.1007/s00281-020-00807-y.
[19] Papadatou, V. *et al.* "Viral-induced inflammation can lead to adverse pregnancy outcomes." *Folia Medica*, vol. 65, no. 5, 2023, pp. 744-752. doi:10.3897/folmed.65.e90054.
[20] Calleja-Agius, J. *et al.* "Obstetric outcome and cytokine levels in threatened miscarriage." *Gynecological Endocrinology*, vol. 27, no. 2, 2011, pp. 121-127. doi:10.3109/09513590.2010.487614.
[21] Saini, V. *et al.* "Cytokines in recurrent pregnancy loss." *Clinica Chimica Acta*, vol. 412, no. 9-10, 2011, pp. 702-708. doi:10.1016/j.cca.2011.01.002.
[22] Christiansen, O.B. *et al.* "Inflammation and miscarriage." *Seminars in Fetal and Neonatal Medicine*, vol. 11, no. 5, 2006, pp. 302-308. doi:10.1016/j.siny.2006.03.001.
[23] Gallinella, G. "The clinical use of parvovirus B19 assays: Recent advances." *Expert Review of Molecular Diagnostics*, vol. 18, no. 9, 2018, pp. 821-832. doi:10.1080/14737159.2018.1503537.
[24] Bonvicini, F. *et al.* "Parvovirus B19 infection in pregnancy: Awareness and opportunities." *Current Opinion in Virology*, vol. 27, 2017, pp. 8-14. doi:10.1016/j.coviro.2017.10.003.
[25] Society for Maternal-Fetal Medicine. "Human parvovirus B19 in pregnancy: Clinical guidance." 27 Aug. 2024. *Society for Maternal-Fetal Medicine*, www.smfm.org/parvovirus-b19. Accessed 5 Sept. 2026.