Journal of Biosciences and Medicines, 2025, 13(4), 347-358
https://www.scirp.org/journal/jbm
ISSN Online: 2327-509X
ISSN Print: 2327-5081

Macrophage Migration Inhibitory Factor, CD163 and Its Association with Placental Malaria

Amal Hussain Mohammed

Department of Medical Laboratories, College of Applied Medical Sciences, Qassim University, Buraydah, Saudi Arabia
Email: Ama.Ali@qu.edu.sa

How to cite this paper: Mohammed, A.H. (2025) Macrophage Migration Inhibitory Factor, CD163 and Its Association with Placental Malaria. *Journal of Biosciences and Medicines*, **13**, 347-358.
https://doi.org/10.4236/jbm.2025.134028

Received: March 10, 2025
Accepted: April 20, 2025
Published: April 23, 2025

Abstract

Background: Malaria has severe effects on pregnancy, and it is a primary cause of maternal and perinatal death in Sudan. This study aimed to investigate the expression of macrophage migration inhibitory factor (MIF) and CD163 in the placentas of malaria-positive and malaria-negative women and explore their potential association with placental malaria pathogenesis. **Methodology:** This case-control study involved 54 malaria-positive and 54 malaria-negative pregnant women at Gadarif Hospital, Sudan. Mothers' body mass index was calculated. Babies were weighed immediately following birth, and maternal hemoglobin concentrations were estimated. Malaria infection was detected using Giemsa-stained blood smears and light microscopy. Maternal, peripheral, and cord blood films were investigated, and immunohistochemical techniques for detecting MIF and CD163 were obtained. **Result:** No significant difference was observed in MIF and CD163 expression between malaria-positive and malaria-negative placentas ($p > 0.05$). Clinical parameters, including age, parity, BMI, maternal hemoglobin levels, and neonatal birth weight, showed no significant variation between the groups. **Conclusion:** The findings suggest that MIF and CD163 expression are not significantly associated with placental malaria in this population. Further research is needed to clarify the immunopathological mechanisms of placental malaria.

Keywords

Placental Malaria, Immune Markers, MIF, CD163, Pregnancy Outcomes, Infection, Malaria, Immune System

1. Introduction

Malaria remains a major public health issue in tropical and subtropical regions

[1]. In Sudan, it has severe implications for pregnant women, acting as a leading cause of maternal and perinatal death [2]. During pregnancy, malaria infection often leads to low birth weight in newborns [3]. One key mechanism involves the sequestration of *Plasmodium*-infected red blood cells in the maternal vascular bed of the placenta [4].

Placental malaria triggers local inflammation. The extent of immune cell infiltration varies among individuals and is inversely related to acquired immunity. Monocytes and macrophages are commonly found in affected placental tissues [5]. A higher number of these cells have been associated with worsened clinical outcomes, including maternal anemia and low birth weight [5] [6]. This likely results from inflammation-induced damage to placental villi, impairing the exchange of nutrients and gases between mother and fetus [7].

One important immune factor in this process is macrophage migration inhibitory factor (MIF), a pro-inflammatory cytokine found at elevated levels in malaria-infected placentas [8]. MIF-related cytokines are also elevated in women infected at higher altitudes. A study conducted at Medani Maternity Hospital in Central Sudan examined MIF's role in malaria severity during pregnancy. However, it found no significant association between MIF levels, birth weight, or maternal hemoglobin [9].

Despite this, other studies suggest that increased activation of monocytes/macrophages—along with their soluble receptors and immune mediators—may drive placental malaria pathology, contributing to anemia and poor fetal outcomes [10] [11]. The presence of *Plasmodium*-infected erythrocytes in the placental intervillous spaces leads to an immune response dominated by monocyte and macrophage infiltration [12]. Elevated MIF levels are often found in the intervillous blood of malaria-infected placentas and are linked with poor outcomes such as stillbirth and low birth weight [13]. Another important marker is CD163, a receptor involved in hemoglobin scavenging. CD163 is expressed on monocytes and macrophages and serves as a marker of anti-inflammatory macrophages. Its soluble form, sCD163, is produced during monocyte/macrophage activation and is elevated in inflammatory conditions. In placental malaria, increased sCD163 levels are inversely associated with maternal hemoglobin, suggesting a link between macrophage activation and malaria-related anemia [14].

2. Research Gap and Justification

While previous studies have explored the individual roles of MIF and CD163 in placental malaria, their combined impact and potential interaction remain unclear. Understanding how MIF expression relates to CD163-positive macrophages could shed light on the mechanisms driving placental inflammation and adverse pregnancy outcomes. This study aims to investigate MIF and CD163 expression in placental tissues from both malaria-positive and malaria-negative women to clarify their roles in placental malaria pathogenesis.

Key Factors in Malaria Infection

1) Species identification.

2) Five *Plasmodium* species infect humans: *P. falciparum*, *P. vivax*, *P. malariae*, *P. ovale*, and *P. knowlesi*. *P. falciparum* is the most lethal, while *P. vivax* has dormant liver stages [15].

3) Parasitemia levels.

4) Parasitemia—the concentration of parasites in the blood—is a critical indicator of infection severity. It can be measured as a percentage of infected red blood cells or as parasites per microliter [16].

5) Timing of infection.

6) The incubation period depends on the species, generally ranging from 1 to 4 weeks post-infection. *P. falciparum* gametocytes appear 7 - 10 days after symptom onset, while *P. vivax* can produce gametocytes even before symptoms emerge [17].

7) Confounding factors.

8) Environmental and socioeconomic factors—such as bed net use, climate, and access to healthcare—significantly influence malaria transmission and outcomes [18].

3. Materials and Method

3.1. Study Design

A case-control study was conducted at the labor ward of the Gadarif Maternity Hospital in eastern Sudan. This tertiary hospital is for women who receive antenatal care there or are referred from other clinics and hospitals.

3.2. Sampling Method and Selection Criteria

To determine the minimum required sample size, we applied the formula

$$n_0 = Z_2 \cdot p \cdot (1 - p) E_2 n_0 = E_2 Z_2 \cdot p \cdot (1 - p)$$

Z = Z-value (based on the confidence level) = 1.96.

p = Estimated proportion of the population = 0.5.

E = Margin of Error: Approximately 0.1335 or 13.35%.

3.3. Calculation

$$(1.96)^2 \cdot 0.5 \cdot (1 - 0.5) \text{E254} = \text{E}_2\ (1.96)^2 \cdot 0.5 \cdot (1 - 0.5) = 54$$

With these parameters, you would calculate a sample size of approximately 54, in each arm of study (mean case-control), indicating the minimum number of participants needed for effective representation in our study. The cases represent a woman with malaria infection. A consecutive woman who delivered next to the case without malaria infection was taken as a control for each case. Women pregnant (case or controls) with twins and those with hypertension, diabetes mellitus, or antepartum hemorrhage were excluded from the study. After obtaining signed informed consent, women in the case and control groups were enlisted to participate in the study. Body mass index was calculated by measuring maternal weight and height, which was expressed as weight (kg)/height $(m)^2$. Babies were weighed im-

mediately following birth to the nearest 10 g on a Salter scale. Scales were checked for accuracy every week. Maternal hemoglobin concentrations were estimated using a Hemo-Cue hemoglobinometer (HemoCue AB, Angelhom, Sweden).

Malaria infection was detected using Giemsa-stained blood smears and light microscopy. Maternal, peripheral, and cord blood films were prepared, and the resultant slides were stained with 10% Giemsa. If no parasites were detected in 100 oil immersion fields of a thick blood film, blood films were considered negative. Films were counted and double-checked blindly by an expert microscopist.

3.4. Sample Collection, Preservation, and Handling

1) Blood Sample Collection

- Maternal Venous Blood: Approximately 5 mL of venous blood was collected from each participant using sterile syringes and transferred into EDTA tubes to prevent coagulation.
- Peripheral and Cord Blood: Maternal peripheral blood was obtained via fingertip prick. Cord blood was collected immediately after delivery through venipuncture of the umbilical vein using heparinized syringes.
- Placental Blood: Intervillous blood was aspirated from the maternal side of the placenta post-delivery using a sterile syringe.

2) Placental Tissue Collection

- A full-thickness placental biopsy (~1 cm^3) was taken from the central region of the placenta, avoiding areas with visible infarcts or calcifications. Each sample was rinsed thoroughly in phosphate-buffered saline (PBS) to eliminate excess blood and tissue debris.

3.5. Sample Preservation and Handling

1) Blood Samples

- Blood smears were prepared immediately after collection and air-dried.
- Thick and thin smears were stained with Giemsa for microscopic detection of *Plasmodium* spp.
- Aliquots of blood were stored at −20˚C for subsequent molecular and biochemical analyses.

2) Placental Tissue

- One section of each tissue sample was fixed in 10% neutral buffered formalin for histopathological and immunohistochemical analysis.
- Additional portions were snap-frozen and stored at −80˚C for future molecular studies.

3.6. Laboratory Processing

1) Microscopy for Malaria Detection

- Giemsa-stained smears were examined under light microscopy at 1000× magnification.
- Parasite density was calculated by counting infected red blood cells per 500

leukocytes.

2) Immunohistochemistry (IHC) for MIF and CD163

- Formalin-fixed tissues were embedded in paraffin, sectioned at 4 μm thickness, and mounted on glass slides.
- Sections underwent deparaffinization, rehydration, and antigen retrieval.
- Tissue sections were then incubated with primary antibodies specific to macrophage migration inhibitory factor (MIF) and CD163 to assess protein expression.

Immediately after delivery of the placenta, samples were sent to the pathology laboratory fixed in 10% neutral buffered formalin. After formalin fixation, samples were cut into 3 - 5 mm slices and processed in a standard manner for the preparation of paraffin wax blocks. Then, sections were treated using standard automated immunohistochemical techniques to detect MIF and CD163. Full automation Immunohistochemistry staining closed system cell Marque antibodies of Ventanas Medical System (Ventanas Medical System, Inc, USA), the procedure started with cut Sections (3 μm) from the paraffin-embedded block of placental tissues was cut by using Rotary microtome and was mounted on poly-L-lysine-coated slides and dried overnight at 58˚C, loaded slides, antibody (Anti-CD163 is mouse monoclonal antibody from supernatant diluted in Tris-buffered saline, PH 7.3 - 7.7, with protein base, and preserved with Sodium azide) and UltraView detection kit dispensers on to Bench Mark instrument, selected CC1 standard pretreatment, started the run, when the staining run was completed, moved slides from device, rinsed well with wash buffer and mounted in DPX. IHC assay and proof slides were coupled with negative and positive controls provided by the manufacturer for each marker, and reactions were observed appropriately. Immunohistochemically stained sections were examined under a light microscope (Olympus CX41, Optical Co., Ltd., Japan). Objective pieces 4× and 10× were used to screen the slide, determine the intensity, and localize the area suitable for counting. Macrophages showing a cytoplasmic brownish color (exhibited by antigen, antibody, and chromogen complex) were considered immune-positive for MIF and CD163. MIF and CD163 immune-positive cells were counted using objective 40× and eyepiece 10×, giving a maximum magnification of 400. Counting was done in the area rich for positive cells and ten consecutive high-power fields (10HPF) using a counter. A single pathologist evaluated each marker's intensity of immunohistochemical staining to reduce intra-observer variability. It was scored based on subjective evaluation of the brownish color as 0 for negative (no color), +1 for a week (light brown color), +2 for moderate (dark brown color), and +3 for strong (very dark brown color). Visualization was achieved using an appropriate secondary antibody and chromogenic substrate. By implementing stringent sample collection and preservation procedures, this study ensured the reliability and reproducibility of malaria detection and immunohistochemical analysis.

3.7. Statistical Analysis

Data was entered into the computer using SPSS for data analysis. T-test and Chi-

square test were used to compare continuous and categorized data between the two groups.

3.8. Ethics Approval

Ethical approval for this study was obtained from the research ethical committee of the Faculty of Medicine, University of Gadarif, Gadarif, Sudan (#08/2021). All participants provided informed consent after receiving information about the study's objectives, methods, and ethics, including confidentiality and the right to withdraw from the study at any time.

4. Results

Fifty-four women were enrolled in each arm of this study. There were no significant differences in the means (SD) in terms of age, parity, BMI, hemoglobin levels, and birth weight, MIF and CD163 expression in the placentas in women with placental malaria and women without placental malaria.

MIF was expressed in 44.5% and 50.0%, of the placentas of women with malaria positive compared with placentas of women with malaria negative, respectively (p = 0.563).

The intensity of MIF expression was mild (16.7% and 14.8%, p = 0.791), moderate (20.4%, and (24.1%, p = 0.643), and intense (7.4% and 11.1%, p = 0.506) in placentas of women with malaria positive compared with placentas of women with malaria negative, respectively (Table 1, Figure 1).

CD163 was expressed in 37.0% and 55.7% of the placentas of women with malaria positive compared with the placentas of women with malaria negative, respectively (p = 0.081). The intensity of CD163 expression was mild (9.3% and 20.4%, p = 0.104), moderate (22.2%, and 27.8%, p = 0.505), and intense in 5.6 % (in each) in placentas of women with malaria positive compared with placentas of women with malaria negative, respectively (Table 2, Figure 2).

Based on Table 1 and Figure 1, none of the clinical and obstetric characteristics show a statistically significant difference between women with and without placental malaria.

Table 1. Mean (±SD) of the clinical and obstetric characteristics of women with placental malaria and women without placental malaria.

Variable	Malaria Positive (n = 54)	Malaria Negative (n = 54)	p-value
Age (years)	27.7 ± 5.4	28.0 ± 4.7	0.315
Parity	2.5 ± 1.8	2.8 ± 1.7	0.678
Gestational Age (weeks)	39.4 ± 1.3	38.9 ± 1.2	0.562
Body Mass Index (kg/m^2)	24.9 ± 2.4	24.7 ± 2.1	0.334
Hemoglobin (g/dL)	10.5 ± 1.2	10.7 ± 1.4	0.265
Birth Weight (g)	3136.9 ± 443.0	3137.8 ± 420.0	0.699

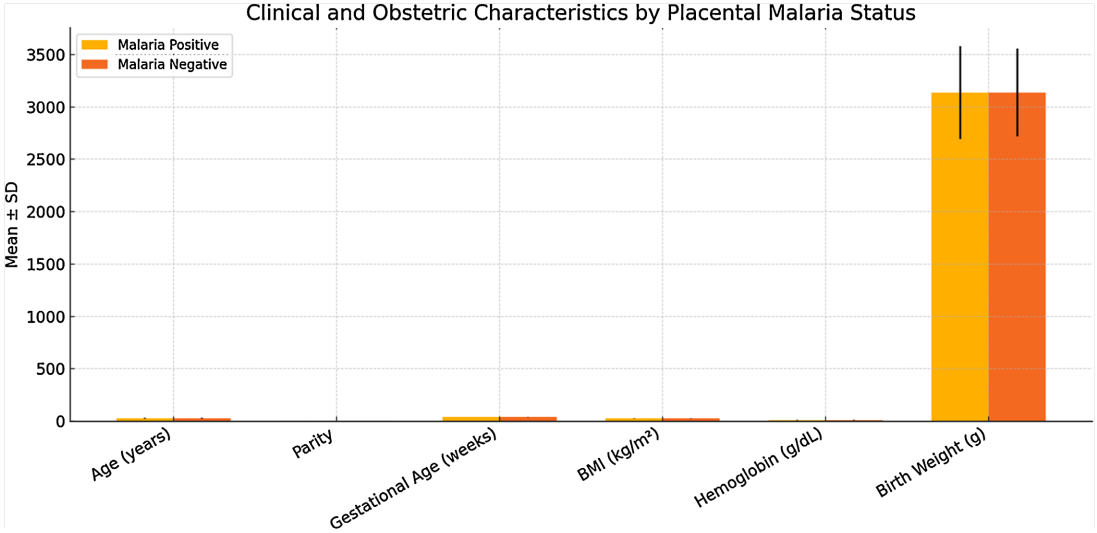

Figure 1. Bar graph displaying the mean ± SD for each clinical and obstetric variable, comparing women with and without placental malaria.

Table 2. Scoring as *n* and (%) of the intensity of MIF and HIF-1a immunostaining in placental malaria positive and placental malaria negative.

Marker	Expression Level	Malaria Positive (%)	Malaria Negative (%)	*p*-value
MIF	Any Expression	44.5	50.0	0.563
	Mild	16.7	14.8	0.791
	Moderate	20.4	24.1	0.643
	Intense	7.4	11.1	0.506
CD163	Any Expression	37.0	55.7	0.081
	Mild	9.3	20.4	0.104
	Moderate	22.2	27.8	0.505
	Intense	5.6	5.6	-

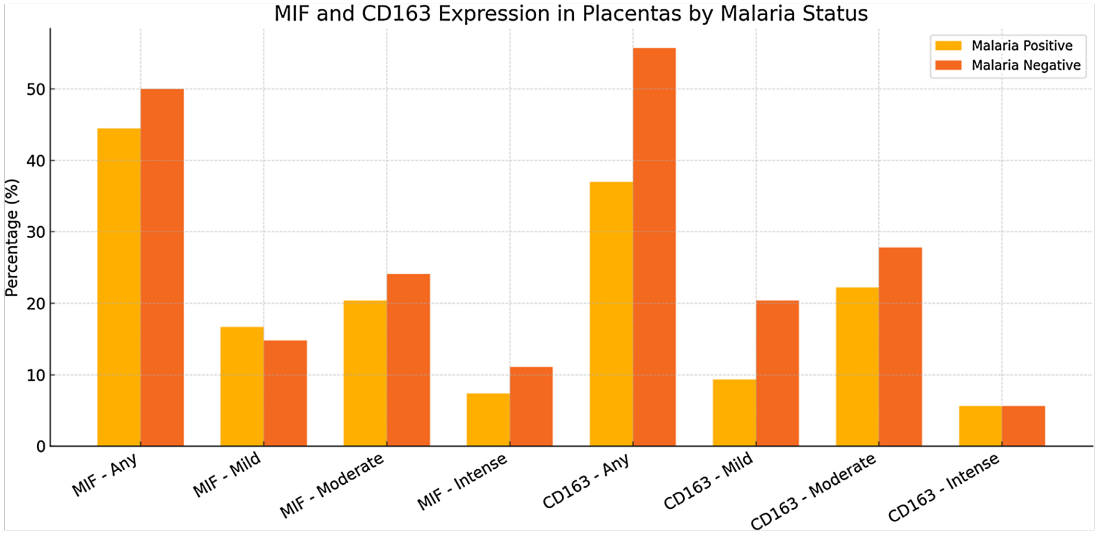

Figure 2. Bar graph comparing MIF and CD163 expression levels in placentas of women with and without malaria. Each expression level is shown by percentage.

The conventional threshold for statistical significance is $p < 0.05$.

All reported p-values are greater than 0.05, indicating no significant differences between the two groups for: Age, parity, gestational age, BMI, hemoglobin levels, birth weight.

There are no statistically significant differences observed in the clinical and obstetric characteristics between women with placental malaria and those without.

Based on the data in **Table 2**, which is reflected in **Figure 2**, there are no statistically significant differences between the malaria-positive and malaria-negative groups for any of the MIF or CD163 expression levels.

p-value < 0.05 is typically considered statistically significant. All p-values in the table are greater than 0.05, meaning none of the comparisons reached statistical significance.

Even the closest value, CD163 "Any Expression" ($p = 0.081$), is not statistically significant, though it suggests a possible trend toward higher CD163 expression in malaria-negative placentas.

No statistically significant differences were found in MIF or CD163 expression between placentas of women with and without malaria in this study.

5. Discussion

This study found no significant difference in the expression of macrophage migration inhibitory factor (MIF) and CD163 in placental tissues between malaria-positive and malaria-negative women. Similarly, there were no significant differences in age, parity, BMI, hemoglobin levels, or birth weight between the two groups. These results contrast with several earlier studies suggesting an immunological response in the placenta due to *Plasmodium falciparum* infection.

For instance, this aligns with Salih *et al.* [5], who proposed that inflammatory infiltrates in the placenta vary among women and are inversely related to acquired immunity, suggesting MIF levels may decline in the later stages of pregnancy as adaptive immunity modulates the response.

However, our findings are consistent with Eltayeb *et al.* [9], who observed no association between MIF levels in maternal or cord blood and placental malaria, maternal anemia, or low birth weight.

A study from Thailand [8] found significantly higher MIF levels in infected placental plasma compared to peripheral plasma ($p < 0.05$), indicating a localized immune response. Our results, however, did not show such differences, reinforcing the idea that placental immune responses are heterogeneous and influenced by multiple factors, including host immunity, timing of infection, and study methodology.

The literature distinguishes between M1 (inflammatory) and M2 (regulatory) macrophages, with CD163 typically associated with the M2 phenotype. The soluble form, sCD163, is released during monocyte/macrophage activation and is a known marker in inflammatory conditions, including pregnancy complications such as preeclampsia [11] [12]. Chua *et al.* reflect this by finding elevated levels of

soluble CD163 (sCD163) in women with placental malaria, particularly in early pregnancy [19]. Elevated sCD163 levels are considered indicators of immune activation rather than protection. MIF, especially in early pregnancy, may serve as a pro-inflammatory mediator during immune responses to malaria infection [20]-[24]. Møller *et al.* [25] reported elevated MIF levels in first-trimester placentas from *P. falciparum*-infected women using immunocytochemical methods. The discrepancy with our findings could be explained by differences in immune status and gestational stage.

Overall, while prior studies suggest that MIF and CD163 are implicated in placental immune responses, our findings did not detect significant differences in their expression, highlighting possible variation in immune profiles, timing of infection, or methodological differences.

Implications

- Reevaluation of Inflammatory Markers: The lack of significant differences in MIF and CD163 expression challenges assumptions about the uniformity of placental inflammatory responses to malaria. This may suggest a more nuanced or context-dependent immune mechanism.
- Clinical Utility: These findings suggest that routine monitoring of MIF and CD163 may have limited diagnostic value in assessing placental health in malaria-exposed pregnancies.
- Public Health Perspective: With no notable differences in key clinical variables (e.g. hemoglobin levels, birth weight), other environmental or health-related factors may play a more significant role in outcomes and warrant further attention.

Future Research Directions

- Alternative Biomarkers: Investigate other immune or inflammatory markers that may better reflect placental responses to malaria.
- Mechanistic Studies: Explore the molecular pathways through which malaria affects placental function, including placental barrier integrity, angiogenesis, and cytokine signaling.
- Longitudinal Research: Study long-term maternal and neonatal outcomes to assess delayed effects of placental malaria.
- Multifactorial Models: Consider co-infections (e.g. HIV, helminths) and other pregnancy-related conditions to understand compound effects.
- Environmental & Socioeconomic Factors: Examine how factors such as nutrition, healthcare access, and malaria prevention strategies influence outcomes.

Study Limitations

This study has several limitations. First, the sample size may not adequately represent the wider population, potentially limiting generalizability. Second, selection and reporting bias could influence the accuracy of the results. Third, as an observational study, causality between macrophage activity and clinical outcomes cannot be inferred. Additionally, not all environmental or comorbid factors were controlled, which may have confounded the findings. Finally, variable follow-up du-

rations could introduce inconsistencies in outcome measurement.

Addressing these limitations in future studies would improve the robustness and applicability of findings.

6. Conclusion

This study found no significant differences in MIF or CD163 expression between malaria-infected and uninfected placentas in pregnant women from eastern Sudan. These findings highlight the complexity of the immune response in placental malaria and suggest a need for further research to identify reliable biomarkers for diagnosis and prognosis.

Acknowledgements

The researcher gratefully acknowledges the laboratory technicians for their precise and diligent work in sample processing and data collection, as well as the statisticians for their support in data analysis. Their contributions were vital to the integrity of this study. Sincere thanks are also extended to all the women who participated and made this research possible.

Funding

This paper received no external funding.

Availability of Data and Material

A data sheet is available.

Authors' Contributions

Amal Hussain Mohammed is responsible for all works of the paper.

Consent to Participate

Written consent was taken from all participants.

Conflicts of Interest

The author declares no conflicts of interest regarding the publication of this paper.

References

[1] Tangpukdee, N., Duangdee, C., Wilairatana, P. and Krudsood, S. (2009) Malaria Diagnosis: A Brief Review. *The Korean Journal of Parasitology*, **47**, 93-102. https://doi.org/10.3347/kjp.2009.47.2.93

[2] Adam, I., Adam, G.K., Mohmmed, A.A., Salih, M.M., Ibrahuim, S.A. and Ryan, C.A. (2009) Placental Malaria and Lack of Prenatal Care in an Area of Unstable Malaria Transmission in Eastern Sudan. *Journal of Parasitology*, **95**, 751-752. https://doi.org/10.1645/ge-1912.1

[3] Adam, I., Babiker, S., Mohmmed, A.A., Salih, M.M., Prins, M.H. and Zaki, Z.M. (2007) Low Body Mass Index, Anaemia and Poor Perinatal Outcome in a Rural Hospital in

Eastern Sudan. *Journal of Tropical Pediatrics*, **54**, 202-204. https://doi.org/10.1093/tropej/fmm110

[4] Reeder, J.C., Hodder, A.N., Beeson, J.G. and Brown, G.V. (2000) Identification of Glycosaminoglycan Binding Domains in *Plasmodium falciparum* Erythrocyte Membrane Protein 1 of a Chondroitin Sulfate A-Adherent Parasite. *Infection and Immunity*, **68**, 3923-3926. https://doi.org/10.1128/iai.68.7.3923-3926.2000

[5] Salih, M.M., Mohammed, A.H., Mohmmed, A.A., Adam, G.K., Elbashir, M.I. and Adam, I. (2011) Monocytes and Macrophages and Placental Malaria Infections in an Area of Unstable Malaria Transmission in Eastern Sudan. *Diagnostic Pathology*, **6**, Article No. 83. https://doi.org/10.1186/1746-1596-6-83

[6] Batran, S.E., Salih, M.M., Elhassan, E.M., Mohmmed, A.A. and Adam, I. (2013) CD20, CD3, Placental Malaria Infections and Low Birth Weight in an Area of Unstable Malaria Transmission in Central Sudan. *Diagnostic Pathology*, **8**, Article No. 189. https://doi.org/10.1186/1746-1596-8-189

[7] Menendez, C., Ordi, J., Ismail, M.R., Ventura, P.J., Aponte, J.J., Kahigwa, E., *et al.* (2000) The Impact of Placental Malaria on Gestational Age and Birth Weight. *The Journal of Infectious Diseases*, **181**, 1740-1745. https://doi.org/10.1086/315449

[8] Chaiyaroj, S.C., Rutta, A.S.M., Muenthaisong, K., Watkins, P., Na Ubol, M. and Looareesuwan, S. (2004) Reduced Levels of Transforming Growth Factor-β1, Interleukin-12 and Increased Migration Inhibitory Factor Are Associated with Severe Malaria. *Acta Tropica*, **89**, 319-327. https://doi.org/10.1016/j.actatropica.2003.10.010

[9] Eltayeb, R., Bilal, N., Abass, A., Elhassan, E.M., Mohammed, A. and Adam, I. (2015) Macrophage Migration Inhibitory Factor and Placental Malaria Infection in an Area Characterized by Unstable Malaria Transmission in Central Sudan. *F*1000*Research*, **4**, Article 824. https://doi.org/10.12688/f1000research.7061.1

[10] Moormann, A.M., Sullivan, A.D., Rochford, R.A., Chensue, S.W., Bock, P.J., Nyirenda, T., *et al.* (1999) Malaria and Pregnancy: Placental Cytokine Expression and Its Relationship to Intrauterine Growth Retardation. *The Journal of Infectious Diseases*, **180**, 1987-1993. https://doi.org/10.1086/315135

[11] Awandare, G.A., Goka, B., Boeuf, P., Tetteh, J.K.A., Kurtzhals, J.A.L., Behr, C., *et al.* (2006) Increased Levels of Inflammatory Mediators in Children with Severe *Plasmodium falciparum* Malaria with Respiratory Distress. *The Journal of Infectious Diseases*, **194**, 1438-1446. https://doi.org/10.1086/508547

[12] Mantovani, A., Sica, A., Sozzani, S., Allavena, P., Vecchi, A. and Locati, M. (2004) The Chemokine System in Diverse Forms of Macrophage Activation and Polarization. *Trends in Immunology*, **25**, 677-686. https://doi.org/10.1016/j.it.2004.09.015

[13] Owino, F., Kijogi, C., Anzala, O., Walong, E., Jael, O., Nyanjom, S.G., *et al.* (2025) Placental Malaria Infection Is Associated with Downregulation of STAT-6 and ANG-1 in Decidual Macrophages. *Frontiers in Immunology*, **16**, Article 1497936. https://doi.org/10.3389/fimmu.2025.1497936

[14] Singh, P.P., Lucchi, N.W., Blackstock, A., Udhayakumar, V. and Singh, N. (2012) Intervillous Macrophage Migration Inhibitory Factor Is Associated with Adverse Birth Outcomes in a Study Population in Central India. *PLOS ONE*, **7**, e51678. https://doi.org/10.1371/journal.pone.0051678

[15] World Health Organization (2024) Malaria. https://www.who.int/news-room/fact-sheets/detail/malaria

[16] Centers for Disease Control and Prevention (2024) Malaria. https://www.cdc.gov/dpdx/malaria/index.html

[17] Acherar, A., Tannier, X., Tantaoui, I., Brossas, J., Thellier, M. and Piarroux, R. (2024)

Evaluating *Plasmodium falciparum* Automatic Detection and Parasitemia Estimation: A Comparative Study on Thin Blood Smear Images. *PLOS ONE*, **19**, e0304789. https://doi.org/10.1371/journal.pone.0304789

[18] Medicines for Malaria Venture: Lifecycle of the Malaria Parasite. https://www.mmv.org/malaria/about-malaria/lifecycle-malaria-parasite

[19] Chua, C.L.L., Brown, G.V., Hamilton, J.A., Molyneux, M.E., Rogerson, S.J. and Boeuf, P. (2013) Soluble CD163, a Product of Monocyte/Macrophage Activation, Is Inversely Associated with Haemoglobin Levels in Placental Malaria. *PLOS ONE*, **8**, e64127. https://doi.org/10.1371/journal.pone.0064127

[20] Rouamba, T., Nakanabo-Diallo, S., Derra, K., Rouamba, E., Kazienga, A., Inoue, Y., *et al.* (2019) Socioeconomic and Environmental Factors Associated with Malaria Hotspots in the Nanoro Demographic Surveillance Area, Burkina Faso. *BMC Public Health*, **19**, Article No. 249.

[21] Alvarado-Vazquez, P.A., Bernal, L., Paige, C.A., Grosick, R.L., Moracho Vilrriales, C., Ferreira, D.W., *et al.* (2017) Macrophage-Specific Nanotechnology-Driven CD163 Overexpression in Human Macrophages Results in an M2 Phenotype under Inflammatory Conditions. *Immunobiology*, **222**, 900-912. https://doi.org/10.1016/j.imbio.2017.05.011

[22] Nunes, P.R., Romão-Veiga, M., Peraçoli, J.C., Araujo Costa, R.A., de Oliveira, L.G., Borges, V.T.M., *et al.* (2019) Downregulation of CD163 in Monocytes and Its Soluble Form in the Plasma Is Associated with a Pro-Inflammatory Profile in Pregnant Women with Preeclampsia. *Immunologic Research*, **67**, 194-201. https://doi.org/10.1007/s12026-019-09078-8

[23] Gordon, S. (2003) Alternative Activation of Macrophages. *Nature Reviews Immunology*, **3**, 23-35. https://doi.org/10.1038/nri978

[24] Knudsen, T.B., Gustafson, P., Kronborg, G., Kristiansen, T.B., Moestrup, S.K., Nielsen, J.O., *et al.* (2005) Predictive Value of Soluble Haemoglobin Scavenger Receptor CD163 Serum Levels for Survival in Verified Tuberculosis Patients. *Clinical Microbiology and Infection*, **11**, 730-735. https://doi.org/10.1111/j.1469-0691.2005.01229.x

[25] Møller, H.J., Grønbæk, H., Schiødt, F.V., Holland-Fischer, P., Schilsky, M., Munoz, S., *et al.* (2007) Soluble CD163 from Activated Macrophages Predicts Mortality in Acute Liver Failure. *Journal of Hepatology*, **47**, 671-676. https://doi.org/10.1016/j.jhep.2007.05.014