

## Seroprevalence of TORCH infections in pregnant women in the city of Likasi, Democratic Republic of Congo

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World Journal of Advanced Research and Reviews, 2025, 28(02), 2192-2200

Publication history: Received 18 October 2025; revised on 24 November 2025; accepted on 26 November 2025

Article DOI: <https://doi.org/10.30574/wjarr.2025.28.2.3958>

### Abstract

**Introduction:** TORCH infections (Toxoplasmosis, Rubella, Cytomegalovirus, Herpes) are diseases transmissible from mother to fetus, representing a global health problem. They can lead to serious complications such as abortions, fetal death *in utero*, or congenital malformations (cerebral and ocular) in the newborn. Cytomegalovirus (CMV) is the most frequent cause of non-genetic congenital developmental disorders.

**Objective:** This study aimed to contribute to the diagnosis and determine the seroprevalence of TORCH infection in pregnant women in Likasi, DRC.

**Methods:** Our study is of a cross-sectional type. This was a study involving a sample of 60 pregnant women in the city of Likasi, recruited during prenatal consultations (PNC). The sample included 20 women in the first trimester, 20 in the second, and 20 in the third trimester, with an age range varying between 19 and 44 years. Blood was collected and the serum was analyzed for TORCH markers (IgG and IgM) using the solid-phase indirect ELISA immunoenzymatic method.

**Results:** The analysis revealed a particularly high seroprevalence for two pathogens:

- Toxoplasmosis (Toxo): 81.66% seropositive women (49 cases).
- Cytomegalovirus infection (CMV): 76.66% seroprevalence (46 cases).

The other TORCH agents showed significant rates: Rubella (43.33%), Herpes type 1 (20%), and Herpes type 2 (23.33%). Furthermore, the seroprevalence of toxoplasmosis and CMV was high in the first, second, and third trimesters of pregnancy.

**Conclusion:** The strong circulation of *Toxoplasma gondii* and CMV in the city of Likasi highlights a significant risk of materno-fetal transmission and congenital complications (malformations, neurodevelopmental disorders). These observations emphasize the necessity of integrating systematic screening for TORCH infections into prenatal consultation (PNC) programs in Likasi, in order to enable early diagnosis and the implementation of adequate management.

**Keywords:** Seroprevalence; TORCH Infections; Pregnant woman; Likasi

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## 1. Introduction

TORCH infections refer to a group of diseases caused by infectious agents (Toxoplasmosis, Rubella, Cytomegalovirus, Herpes) that can be transmitted from mother to fetus during pregnancy, childbirth, or after birth. Toxoplasmosis is a zoonosis caused by a protozoan parasite: *Toxoplasma gondii*, an obligatory intracellular parasite of the reticuloendothelial system. It is a widespread global health problem in both developed and developing countries. (KHALDI, 2019)

It is a parasitic disease most often responsible for an inapparent or benign infection in the immunocompetent host, but it can be serious in the immunocompromised and during pregnancy due to the transplacental passage of the parasite, exposing the fetus to congenital toxoplasmosis.

Congenital CMV infection is the most frequent cause of non-genetic congenital malformations and developmental disorders (mainly deafness and psychomotor delay) and can lead to prematurity or intra-uterine or neonatal death (Hantz et al., 2024). In the case of a maternal primary infection, intra-uterine transmission rates vary according to gestational age, from approximately 5 to 16% (preconceptional or periconceptional) up to 65% (in the third trimester) (Paquet & Yudin, 2018). Reactivations and reinfections can also cause intra-uterine infection in about 1 to 2% of cases, and it is possible that transmission rates in cases of reinfection are underestimated (Huraux et al., 1999).

Rubella is a cosmopolitan infection that occurs endemically interspersed with epidemics with seasonal resurgence (predominant in spring in temperate climates). Currently, due to mass vaccination of children, rubella is evolving with winter-spring epidemic surges, mainly affecting adolescents, young adults, and unvaccinated children, thus posing a risk to pregnant women (Vauloup-Fellous & Portet-Sulla, 2025).

Herpes simplex virus (HSV) is one of the most widespread viruses globally. It is responsible for acute or recurrent infections in humans. Once in humans, HSV infects the sensory neurons innervating the mucous membranes involved in the acute infection. It then migrates to the regional sensory ganglia and remains latent (Picone, 2017). During reactivations, the virus can lead to the appearance of vesicles and ulcers. There are two species of HSV:

- **HSV type 1 (HSV-1)**, also known as *Herpesvirus labialis*, which commonly infects the oral region.
- **HSV-2** (*Herpesvirus genitalis*), which most often infects the genito-anal region.

Both types can cause infections in sites commonly recognized for the other. Most infected people can excrete the virus periodically even in the absence of clinical manifestation. In its genital form, HSV infection is associated with the acquisition and transmission of HIV. Furthermore, it is responsible for neonatal herpes in the newborn. If untreated, neonatal herpes results in mortality greater than 60%, and survivors retain severe disabilities, such as mental retardation or blindness (Waggoner-Fountain & Grossman, 2004).

The present study is based on the following hypotheses:

- First, congenital CMV infection is the most frequent cause of non-genetic congenital malformations and developmental disorders (mainly deafness and psychomotor delay) and can lead to prematurity or intra-uterine or neonatal death.
- Conversely, toxoplasmic contamination in a pregnant woman generally does not present direct risks for the mothers, and this contamination during pregnancy rests on the risks of causing fetal death or neurological or ophthalmological complications during the first years of the child's life.
- Rubella is a contagious infection caused by a virus. Rubella infection is not serious except in pregnant women, as rubella can have serious consequences for their baby, especially in the first 3 months of pregnancy. This infection is infrequent, thanks to the vaccination which is effective against the virus. In practice, the diagnosis of rubella infection is based on serology.
- Finally, the development of neonatal herpes is heavily dependent on local epidemiology. Populations with high HSV2 seroprevalence have a low prevalence of neonatal Herpes. The major risk of materno-fetal transmission occurs at term, during vaginal delivery. Perinatal infection represents about 85% of all cases of neonatal herpes. The fetus comes into contact with infected vaginal secretions. The entry points are the natural mucous membranes, mainly the eyes, the nasopharynx, but also skin lesions secondary to invasive obstetric maneuvers. Intra-uterine infection represents 5-8% of all cases of neonatal herpes and can result from either the transplacental or ascending route.

This study aims to contribute to the diagnosis of TORCH infection in pregnant women in Likasi. More specifically, it seeks to:

- Search for TORCH antibodies in pregnant women in Likasi.
- Determine the seroprevalence of TORCH infection in pregnant women in Likasi.

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## 2. Materials and Methods

### 2.1. Research Setting

The Dee Service medical laboratory in Lubumbashi was selected as the experimental site for this study. The choice of this location is based on several geographical and environmental factors. Likasi is located in the heart of the Haut Katanga mining region, near the Mitumba and Kundelungu mountains. Situated on the Haut Katanga plateau, Likasi has a mild tropical climate and the average temperature is 20°C. The city is full of hills with an average altitude of 1265 m and isohypses at 1100 and 1400 m (*Guellord et al., 2021*).

### 2.2. Material

For blood collection and laboratory assays, the following materials were used:

- ACCESS2 DE BECKMAN automated analyzer
- EDTA tubes
- Syringes
- Refrigerator
- Centrifuge
- Pipettes of 10-100µl and 100-1000µl
- Tube racks
- Cuvettes
- Tips
- Syringes and needles
- Cotton wool
- Tourniquet
- Test tubes

### 2.3. Study Subjects

Our study is of a cross-sectional type. In the context of this study, our sample size was 60 people. The target population was exclusively pregnant women in the city of Likasi. Among the 60 people, we randomly selected 20 pregnant women in the first trimester, 20 pregnant women in the second trimester, and 20 pregnant women in the third trimester. The age range varied between 19 and 44 years. The results obtained were recorded and statistically analyzed to evaluate the seroprevalence of TORCH infection.

### 2.4. Inclusion and Exclusion Criteria

#### 2.4.1. Inclusion criteria were:

- Pregnant women residing in Likasi.
- Pregnant women who gave informed consent to be part of our study.
- Pregnant women not diagnosed positive for the study pathogens.

#### 2.4.2. Exclusion criteria were:

- Pregnant women diagnosed positive for the study pathogens.
- Pregnant women not residing in Likasi.
- Pregnant women who did not give informed consent to be part of our study.

The selection of our study subjects was carried out randomly without distinctions of race, tribe, or social class among all pregnant women who came for prenatal consultations (PNC).

## 2.5. Sample Collection and Processing

Blood collection was performed in the morning between 8:00 a.m. and 10:00 a.m. for the study subjects. The blood was collected in test tubes without anticoagulant, the samples were centrifuged at 2500 revolutions per minute for 10 minutes to obtain the serum, which was collected and kept at 4°C until the analyses, which were performed the same day.

## 2.6. Laboratory Analyses

All TORCH markers were performed using indirect ELISA immunoassays for the quantitative determination of antibodies in human serum or plasma (Maudry *et al.*, 2009).

- **Cytomegalovirus IgG assay:** The principle states that IgG is a test allowing the detection and titration of anti-CMV IgG antibodies in human serum or plasma by an indirect ELISA solid-phase immunoenzymatic method. Inactivated CMV antigen is used to sensitize the microplate. A peroxidase-labeled polyclonal antibody specifically directed against human gamma chains (anti-IgG) is used as the conjugate (Carlier *et al.*, 2010).
- **CMV IgM assay:** The principle states that IgM is a test allowing the qualitative detection of anti-CMV IgM antibodies in human serum or plasma by an immunoenzymatic method with IgM immunocapture on a solid phase. Anti-human  $\mu$  chain antibodies are used to sensitize the microplate. A mixture of CMV antigen and anti-CMV monoclonal antibody labeled with peroxidase is used as the conjugate (Carlier *et al.*, 2010).
- **Toxoplasmosis IgG assay:** The principle states that IgG is a test allowing the detection and titration of anti-*T. gondii* IgG antibodies in human serum or plasma by an indirect ELISA solid-phase immunoenzymatic method. *T. gondii* antigen is used to sensitize the microplate. A peroxidase-labeled monoclonal antibody specifically directed against human gamma chains (anti-IgG) is used as the conjugate (Maudry *et al.*, 2009).
- **Toxoplasmosis IgM assay:** The principle states that IgG is a test allowing the qualitative detection of anti-*T. gondii* IgG antibodies in human serum or plasma by an immunoenzymatic method with IgG immuno-capture on a solid phase. Anti-human  $\mu$  chain antibodies are used to sensitize the microplate. A mixture of *T. gondii* antigen and anti-*T. gondii* monoclonal antibody labeled with peroxidase is used as the conjugate (Maudry *et al.*, 2009).
- **Rubella IgG** is a test for the detection and titration of IgG antibodies against the Rubella virus in human serum or plasma by the indirect ELISA immunoenzymatic method. Rubella antigen is used for microplate coating. A peroxidase-labeled monoclonal antibody specific for human gamma chains (anti-IgG) is used as the conjugate.
- **Rubella IgM** is a qualitative test for the detection of IgM antibodies against the Rubella virus in human serum or plasma by immunoenzymatic assay with IgM capture on a solid phase. Anti-human  $\mu$  chain antibodies are coated on the solid phase (microplate wells). A mixture of Rubella antigen and anti-Rubella monoclonal antibody labeled with peroxidase is used as the conjugate (Grangeot-Keros, 2005).
- **HSV IgM assay:** The test is based on the principle of capturing these immunoglobulins and the subsequent identification of those that are specific by exploiting their ability to bind an antigen conjugated to peroxidase. Capture is achieved using monoclonal antibodies linked to the solid phase (microtiter wells). The antigen is composed of purified and inactivated HSV type 1 and type 2. The conjugate is composed of peroxidase-labeled anti-HSV specific monoclonal antibodies (Bossuyt & Boeynaems, 2001).
- **HSV IgG assay:** The test for IgG assay is based on the ELISA (Enzyme-Linked Immunosorbent Assay) technique. The antigen, composed of purified and inactivated Herpes Virus types 1 and 2, is linked to the solid phase (8-well strips). Specific immunoglobulins are linked to the antigen by incubation with diluted human serum. After washing to remove unreacted proteins, incubation is performed with the conjugate, composed of peroxidase-labeled human IgG monoclonal antibodies. Unbound conjugate is removed and the peroxidase substrate is added. The color that develops is proportional to the concentration of specific antibodies present in the serum sample (Bossuyt & Boeynaems, 2001).

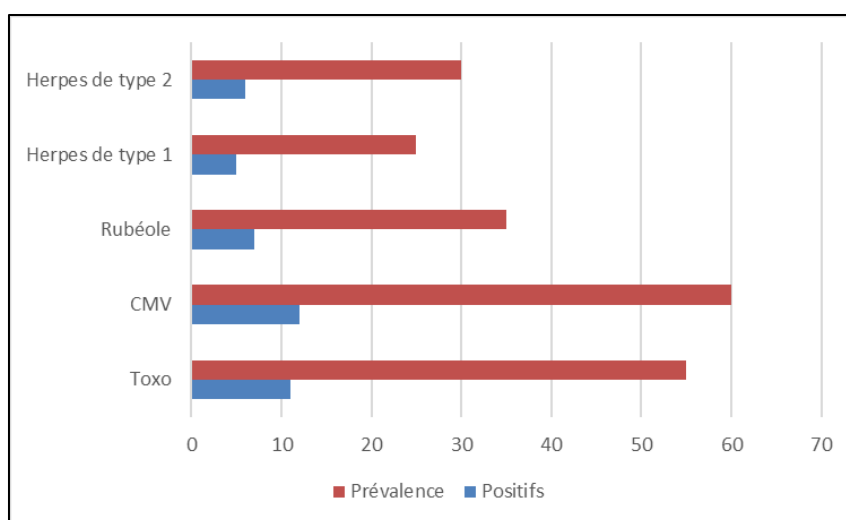
## 2.7. Statistical Analysis

In this study, frequencies and percentages allowed us to calculate the proportion of seropositive pregnant women for each TORCH agent (Toxoplasmosis, Rubella, Cytomegalovirus, Herpes Simplex Virus) and for all agents. The statistical software R STUDIO was used for descriptive statistics in this study. (Bouyer, 2017) (Lesko *et al.*, 2022).

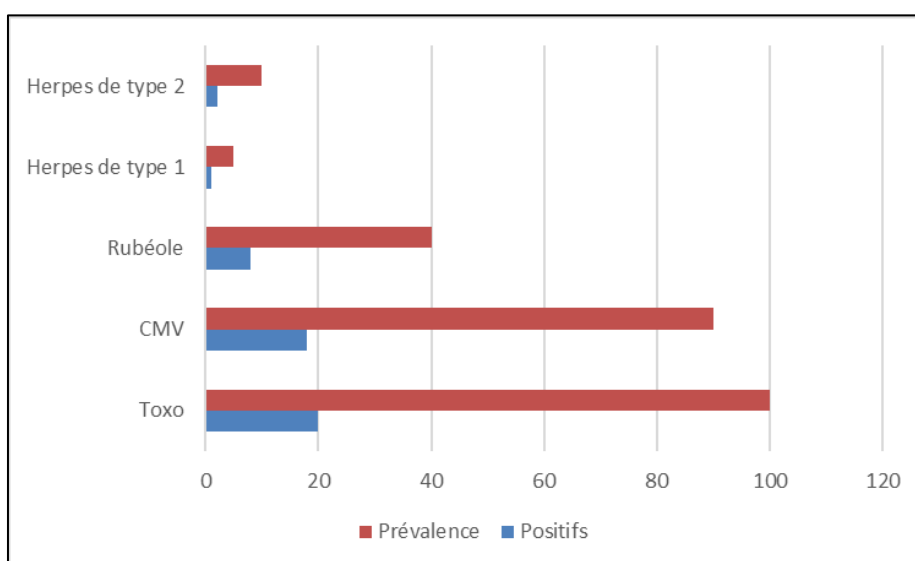
### 3. Results

**Table 1** Seroprevalence of TORCH Infections in Pregnant Women in Likasi

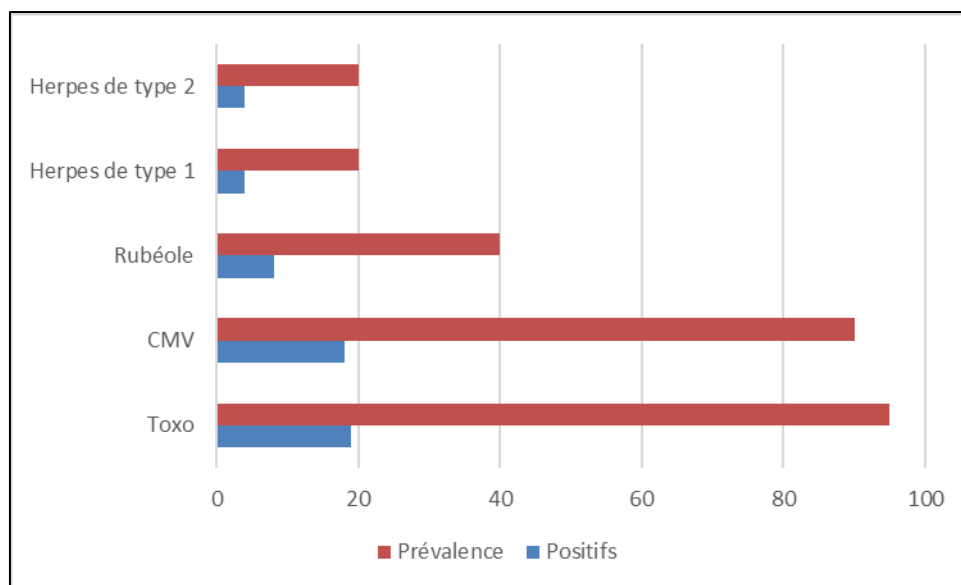
Marker	Positive Cases	Prevalence (%)
Toxo	49	81.66
CMV	46	76.66
Rubella	26	43.33
Herpes type 1	12	20
Herpes type 2	14	23.33



**Figure 1** Seroprevalence of TORCH infections in pregnant women in the First Trimester in Likasi.



**Figure 2** Seroprevalence of TORCH infections in pregnant women in the Second Trimester in Likasi.



**Figure 3** Seroprevalence of TORCH infections in pregnant women in the Third Trimester in Likasi.

Analysis of Table 1 (above) as well as Figures 1, 2, and 3 allowed for the following observations:

- In the pregnant women population of Likasi, Toxoplasmosis and CMV infection were identified more frequently than the other TORCH infections, with respective proportions of 81.66% and 76.66% versus Rubella and Human Simplex Virus type 1 and 2 infections (Table 1).
- In addition, seroprevalence in the first, second, and third trimesters showed high rates concerning Toxoplasmosis and CMV infection versus Rubella and HSV-1 and 2 infections (Figures 1, 2, and 3).

#### 4. Discussion

Our study focused on a sample of 60 pregnant women in the city of Likasi, equally distributed across the three trimesters of pregnancy. After analysis, our results reveal a particularly high prevalence for two major pathogens in this population: Toxoplasmosis with 81.66% and Cytomegalovirus (CMV) infection with 76.66% of seropositive women. The other TORCH agents, namely Rubella, Herpes type 1, and Herpes type 2, showed lower but still significant proportions. Furthermore, the seroprevalence for Toxoplasmosis and CMV was high in the first, second, and third trimesters.

The seroprevalence rate of 81.66% for Toxoplasmosis in Likasi is significantly higher than that reported in other regions. For example, a study by L. Messerer (2014) revealed a seroprevalence of 47.8% in pregnant women. This high rate highlights a wide circulation of the parasite *Toxoplasma gondii* in our study area. Toxoplasmosis is a parasitic disease responsible for an infection that, although often inapparent in the immunocompetent, is serious during pregnancy due to transplacental passage, exposing the fetus to congenital toxoplasmosis. The severity of congenital clinical signs (cerebral and ocular malformations, chorioretinitis progressing to blindness, fetal death *in utero*) is linked to infection occurring during the first trimester. However, the observation that only a minority of women in the study population present for prenatal consultations (PNC) in the first trimester of pregnancy poses a public health problem for the diagnosis and early management of recent infections (Yolande *et al.*, 2018).

CMV infection is the leading cause of neurodevelopmental pathology and can lead to severe consequences (deafness, psychomotor delay, polymicrogyria). The seroprevalence rate of 76.66% in Likasi is comparable to other African studies, notably that of Y. Sissinto *et al.* (2018), and indicates that the virus is widespread in the area (Hantz *et al.*, 2024). This high prevalence reinforces the need for systematic screening for Cytomegalovirus for all women in PNC to limit transmission and congenital risks (St-Georges, 2025).

Rubella is generally benign, but it is a cause of spontaneous abortion, fetal death, or congenital malformations if the primary infection occurs in the first trimester of pregnancy. Seroprevalence surveys are of great interest in measuring the prevalence of rubella infection in pregnant women. The risk of fetal infection is maximal before 11 weeks of

amenorrhea (WA) (90%), then decreases, and then increases again at the end of pregnancy. The clinical evolution (embryopathy or fetopathy) depends on the gestational age at which contamination occurs (*Dontigny et al., 2008*).

HSV (types 1 and 2) is one of the most widespread viruses in the world, capable of acute or recurrent infections and latency in the sensory ganglia. The development of neonatal herpes (NHSV) is highly dependent on local epidemiology. A large proportion of HSV-2 seropositive individuals were unaware they were infected (75% to 90%), which makes all newborns potentially vulnerable. The major risk of materno-fetal transmission, representing about 85% of all cases of neonatal herpes, occurs during vaginal delivery (perinatal infection) (*Picone, 2017*). The fetus comes into contact with infected vaginal secretions, with the entry points being the natural mucous membranes or skin lesions secondary to invasive obstetric maneuvers (*Waggoner-Fountain & Grossman, 2004*).

The strong circulation of *Toxoplasma gondii* and CMV observed in this study confirms a significant risk of materno-fetal transmission and congenital complications (malformations, neurodevelopmental disorders) for the fetus and newborn in Likasi. It is therefore imperative to integrate systematic screening for TORCH infections into PNC programs in Likasi, in order to allow for early diagnosis of primary infections and the implementation of adequate management (*Huraux et al., 1999*).

However, our study presents certain limitations:

- The study was conducted on a total sample of only 60 pregnant women, divided into 20 subjects per trimester of pregnancy. Such a limited sample size detracts from the representativeness of the total population of pregnant women in Likasi and limits the generalization of seroprevalence results to the entire city.
- The study is cross-sectional in nature (each woman was tested only once at a stage of her pregnancy). There is no follow-up of positive (or negative) cases of the same woman from the first to the third trimester.
- The study used the indirect ELISA method to detect IgG and IgM antibodies. Although the detection of IgM suggests a recent infection, these antibodies can persist for several months. The article does not mention the use of IgG avidity tests, which is an essential tool for dating the infection (differentiating a very recent infection from one from the previous year). Without avidity data, it is difficult to determine with certainty which are the cases of acute primary infection (infection that occurred during pregnancy) that pose the highest risk of fetal transmission.
- Conversely, the fact that most women begin prenatal consultations (PNC) in the second trimester or later poses a public health problem and a limitation for the study, as it leads to a selection bias and prevents the early diagnosis and management of infections that may have occurred *before* the first contact with the health system.

## 5. Conclusion

Our study aimed to determine the seroprevalence of TORCH infections (Toxoplasmosis, Rubella, Cytomegalovirus, Herpes) in pregnant women in the city of Likasi. The size of our sample was 60 people. The target population was exclusively pregnant women in the city of Likasi. Among the 60 people, we selected 20 pregnant women in the first trimester, 20 pregnant women in the second trimester, and 20 pregnant women in the third trimester. The age range varied between 19 and 44 years.

The results revealed a particularly high prevalence for two major pathogens within the studied population:

- **Toxoplasmosis (Toxo)** with 81.66% of seropositive pregnant women.
- **Cytomegalovirus infection (CMV)** with 76.66% seroprevalence.

The other TORCH agents also showed significant rates, notably Rubella at 43.33%, Herpes type 1 at 20%, and Herpes type 2 at 23.33%. The analysis of seroprevalence according to the trimesters of pregnancy also showed high rates for Toxoplasmosis and CMV from the first to the third trimester. This strong circulation of TORCH infectious agents, particularly *Toxoplasma gondii* and CMV, in the city of Likasi, highlights a significant risk of materno-fetal transmission and congenital complications (malformations, neurodevelopmental disorders, intrauterine death) for the fetus and newborn.

These observations emphasize the necessity of integrating systematic screening for TORCH infections into prenatal consultation (PNC) programs in Likasi in order to enable:

- The early diagnosis of primary infections.
- The implementation of adequate management aimed at reducing the risks of fetal contamination.

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## Compliance with ethical standards

### *Acknowledgments*

We extend our thanks to our wives and children, to the pregnant women of the city of Likasi, to the members of the journal's editorial board, as well as to the Dee Service Laboratory in Lubumbashi.

### *Disclosure of conflict of interest*

This work was carried out with impartiality and complete independence of mind. No conflict of interest was reported.

### *Statement of ethical approval*

The free and informed consent of each pregnant woman was obtained. Data confidentiality was guaranteed, with the collected data being anonymized as soon as possible to protect the participants' identity, and the selection of participants was fair, avoiding the exploitation of vulnerable groups.

### *Authors' Contribution*

Arold Fazili designed and supervised the study, wrote the main manuscript and validated the final version, validated the data, contributed to the discussion, and gave final approval of the version to be submitted.

- Cynthia BUTEKA designed and supervised the study, wrote the main manuscript and validated the final version.
- Prisca NDAY designed and supervised the study, wrote the main manuscript and validated the final version.
- Dan KASANGA designed and supervised the study, wrote the main manuscript and validated the final version.
- Armand ABASI participated in data collection, statistical analysis, and contributed to the interpretation of results and critical review of the manuscript.
- Gradine Muinga ensured the bibliographic review and participated in the formatting of the document.

All authors contributed to the interpretation of the results and the critical review of the manuscript. All authors read and approved the final version of the manuscript.

### *Funding*

This article was entirely funded by the authors themselves.

### *Statement of informed consent*

Informed consent was obtained from all individual participants included in the study.

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