



Seroepidemiology and Associated Risk Factors for *Toxoplasma gondii* Infection among Pregnant Women Attending Antenatal Clinics at the Sino-Gabonese Friendship Hospital in Franceville, South East Gabon

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clinics at the Sino-Gabonese Friendship Hospital in Franceville from 03 January to 28 November 2022 were sorted and recorded for this study. With a mean age of 30 ± 7.38 years, 67 cases were reported to be positive for anti- *Toxoplasma gondii* antibodies (IgG and/or IgM), indicating an overall seroprevalence of 25.77% (67/260; 95% CI:[0.21-0.32]). While IgG antibodies to *Toxoplasma gondii* were detected in 23.85% (62/260) of pregnant women, 1.92%(5/260) was positive for IgM. Being aged between 31 and 40 years (Odds Ratio=4.58; CI95% [2.52;8.32], $p<0.001^*$) and between 41 and 43 years (Odds Ratio=0.24; CI 95%[0.05;1.05], $p=0.040^*$), single (Crude Odds Ratio=6.12. 95% CI [3.13; 11.9], $p<0.001^*$), a primary education level (Crude Odds Ratio=4.57; 95% CI [2.53-8.26], $p<0.001^*$), a shopkeeper (Crude Odds Ratio=2.93; 95% CI [1.51; 5.68], $p=0.000^*$), or a housewife (Crude Odds Ratio=0.24; 95% CI [0.11; 0.52], $p<0.001^*$), living in a rural area (Crude Odds Ratio=3.02; 95%CI [0.54; 5.1], $p=0.001^*$), having between 2 and 3 pregnancies (Crude Odds Ratio=2.62; 95% CI [1.4; 4.63], $p=0.000^*$) and being in the first trimester of a new pregnancy (Crude Odds Ratio=2.55; 95% CI [1.25; 4.83], $p=0.003^*$), were significant predictors of *Toxoplasma gondii* seroprevalence in the pregnant women studied.

Conclusion: 25.77% was the overall seroprevalence rate of *Toxoplasma gondii* infection among pregnant women in the present study. This relatively high rate was significantly associated with a number of socio-demographic characteristics. It would therefore be important to guarantee quality maternal healthcare and health education for pregnant women in both urban and rural areas of Gabon. This would help to reduce the prevalence or prevent *Toxoplasma gondii* infection in the population.

Keywords: Seroepidemiology; Pregnant women; *Toxoplasma gondii*; Franceville; Gabon

Abstract

Background: Generally asymptomatic in pseudo-healthy individuals, *Toxoplasma gondii* infection can lead to serious pathological complications among pregnant women and their fetuses, as well as in immuno compromised patients. This study was conducted to investigate the seroepidemiology and associated risk factors of this infection among pregnant women attending antenatal clinics at the Sino-Gabonese Friendship Hospital in Franceville.

Patients and Methods: This retrospective cross-sectional descriptive study was based on the consultation of randomly selected records and test results for the detection of *Toxoplasma gondii* in pregnant women attending antenatal clinics from 03 January to 28 November 2022 at the Sino-Gabonese Friendship Hospital in Franceville. Socio demographic information and obstetrical data of pregnant women were collected and analysed using R software version 4.2.1, and the results were considered significant for a value of $p \leq 0.05$.

Results: A total of 260 records and test results for the detection of *Toxoplasma gondii* from pregnant women attending antenatal

Introduction

During pregnancy, women are exposed to numerous infections and risks that can have an impact on their health and the outcome of their foetus. This is the case with infection by *Toxoplasma gondii*, known as Toxoplasmosis [1]. It is estimated that a third of the world's population is infected with *Toxoplasma gondii* [2]. Three serious sequelae have major implications for human health: congenital infection, ocular toxoplasmosis and cerebral toxoplasmosis, in HIV-infected and immuno-compromised individuals [3]. Oocysts are the robust environmental stage of the parasite and are excreted only in the faeces of domestic cats and wild felids. *Toxoplasma gondii* oocysts can contaminate soil and water, where they can persist in extreme environmental conditions and can remain infective for one or more years [4]. This disease, which is generally benign (asymptomatic), can cause fetal death in pregnant women or malformations in their newborns [5]. This is why it is necessary to make a serological diagnosis of this infection, during antenatal visits during pregnancy [6]. When toxoplasmosis occurs during pregnancy, its parasite crosses the placenta and infects the foetus [7]. This is known as congenital toxoplasmosis. This is the fatal form of the disease, resulting in abortion, fetal death in utero or serious malformations with damage to the central nervous system. Increasing with age, numerous

epidemiological studies have indicated that the prevalence rate of *Toxoplasma gondii* infection in pregnant women varies considerably between countries and regions depending on dietary habits, health standards and socio-economic level [8]. Unlike European countries, where the prevalence of *Toxoplasma gondii* infection in pregnant women varies from 9% to 67%, low prevalences of this infection have been found in Asian countries. This is the case in Korea (0.8%) and Vietnam (11.2%) [9-11]. In the United States of America, the annual burden of *Toxoplasma gondii* infection has been estimated at 10,964 Quality-Adjusted Life Years (QALYs), and is the third most important foodborne pathogen after *Salmonella enterica* and *Campylobacter* species in terms of costs of illness, at \$2.9 billion [12]. In the African region, the overall combined seroprevalence of *Toxoplasma gondii* infection in pregnant women was 51.01% (95% CI; 37.66, 64.34) while the incidence rate of congenital toxoplasmosis is around 1.5 cases per 1000 live births [13,14]. In Gabon, despite the existence of a number of studies on *Toxoplasma gondii* infection, such as that carried out in Koula-Moutou and the surrounding departments, in which a seroprevalence of 80.41% was reported among pregnant women in this region [15]. The diagnosis of this disease is not always systematic in pregnant women. In view of the frighteningly high rates recorded in this part of the vulnerable population, the main objective of this study was to assess the sero-epidemiology and associated factors of *Toxoplasma gondii* infection among pregnant women attending antenatal clinics at the Sino-Gabonese Friendship Hospital in Franceville, in South East Gabon.

Patients and Methodology

Description of the host organization

This study was conducted at the medical analysis laboratory of the Sino-Gabonese Friendship Hospital in Franceville. Located in the 2nd district of the said town, this hospital has adequate technical facilities for routine hospital examinations, and welcomes people from all walks of life.

Type, period and study population

This retrospective, cross-sectional study was conducted in the medical analysis laboratory of the Sino-Gabonese Friendship Hospital in Franceville. It was based on a random selection of records and test results for the detection of IgM or IgG antibodies against the toxoplasmosis virus in pregnant women from 03 January to November 2022, entered into a database in the hospital's health information management system. The aim of the study was therefore to determine the prevalence and associated risk factors of *Toxoplasma gondii* infection recorded during the study period.

Inclusion and exclusion criteria

Only results obtained from qualitative screening for anti-*Toxoplasma gondii* antibodies using the DIALAB IgG-IgM test Enzyme Linked Immunosorbent Assay (ELISA), available on the market, carried out on blood serum/plasma taken from pregnant women attending antenatal clinics from 03 January to November 2022, at the Sino-Gabonese Friendship Hospital in Franceville, were taken into account in the present study. For this test, the correlation between serological status and LDBio ICT (Immunochromatographic Test) is excellent: sensitivity Se=100%, specificity Sp=97.7%. Incomplete records and unexploitable or doubtful results were excluded from the study.

Sampling method

Purposive sampling was used to target and focus only on the results of analyses of serum/blood plasma samples taken from pregnant women attending antenatal clinics from 03 January to November 2022 at the medical analysis laboratory of the Sino-Gabonese Friendship Hospital in Franceville during the study period. To ensure that the study was representative, the sample size depended on the number of cases recorded in the laboratory database.

Procedure for obtaining data

The data used for the study were the results of analyses of serum/blood plasma samples taken from pregnant women attending antenatal clinics from 03 January to November 2022 and obtained at the laboratory of the Sino-Gabonese Friendship Hospital in Franceville, selected at random. Access to these data was facilitated by the collaboration between the management of this hospital and the Masuku University of Science and Technology. The extracted data were made available to us in digital form. All the results obtained from the qualitative screening for anti-*Toxoplasma gondii* antibodies were extracted and used for the present study.

Data collection

During this study, variables such as socio-demographic characteristics (age, place of residence (urban and semi-urban areas), marital status, professional status, number of months pregnant, number of pregnancies) were collected. The results of the Toxoplasmosis diagnostic test and the titre of the reaction were evaluated

Ethical considerations

The data received did not include patient identity or personal information

Statistical analysis of the data

Entered into a Microsoft Excel 2016 format, the data collected from the laboratory registers were then analysed using R software version 4.2.1. Measures of central tendency and dispersion were determined for age with a 95% confidence interval. The χ^2 test was used to compare proportions. The search for an association between the study variables and the prevalence of toxoplasmosis was carried out by univariate analysis. Odds Ratios (OR) was calculated along with their confidence intervals (95% CI). The results were considered statistically significant for a $p \leq 0.05$ value.

Results

Seroprevalence of *Toxoplasma gondii* infection in pregnant women in the study (N=260)

A total of 260 records and test results for the detection of *Toxoplasma gondii* from pregnant women attending antenatal clinics at the Sino-Gabonese Friendship Hospital in Franceville from 03 January to 28 November 2022 were randomly collected for this study. With a mean age of 30 ± 7.38 years, 67 cases tested positive for anti-*Toxoplasma gondii* antibodies (IgG and/or IgM), indicating an overall seroprevalence of 25.77% (67/260; 95% CI; [0.21-0.32]), including 23.85% (62 cases) for IgG and 1.92% (5 cases) for IgM. The study

found that 74.23% (193) of pregnant women were seronegative and likely to be infected during pregnancy.

Seroprevalence of *Toxoplasma gondii* infection according to sociodemographic and obstetric characteristics of pregnant women in the study (N=260)

In a univariate analysis of the prevalence of *Toxoplasma gondii* infection according to the sociodemographic and obstetric characteristics of the pregnant women in the study, it was observed that those aged between 31 and 40 (Odds Ratio=4.58; IC95% [2.52; 8.32], p<0.001*) and between 41 and 43 (Odds Ratio=0.24; CI 95%

[0.05;1.05], p=0.040*), single (Crude Odds Ratio=6.12; CI 95% [3.13; 11.9], p<0.001*), whose level of education or instruction was primary (Crude Odds Ratio=4.57. CI 95% [2.53-8.26], p<0.001*), a shopkeeper (Crude Odds Ratio=2.93; CI 95% [1.51; 5.68], p=0.000*), or a housewife (Crude Odds Ratio=0.24; CI 95% [0.11; 0.52], p<0.001*), living in a rural area (Crude Odds Ratio=3.02; CI 95% [0.54; 5.1], p=0.001*), had between 2 and 3 pregnancies (Crude Odds Ratio=2.62; 95% CI [1.4; 4.63], p=0.000*) and were in the first quarter of a new pregnancy (Crude Odds Ratio=2.55; 95% CI [1.25; 4.83], p=0.003*), were significantly associated with *Toxoplasma gondii* infection (Table 1).

Variables	Number of pregnant women in the study N (%)	Prevalence of <i>Toxoplasma gondii</i> infection		Univariate analysis Crude OR 95% CI	p-value
		Positive	Negative		
		N (%)	N (%)		
Age groups (years)					
≤20	26 (10)	1(3.85)	25(96.15)	Ref	-
21-25	45 (17.31)	8(17.78)	37(82.22)	0.57 [0.25; 1.3]	0.18
26-30	58 (22.31)	14 -24.14	44 -75.86	0.89 [0.45; 1.8]	0.75
31-40	107 (41.15)	42(39.25)	55(60.75)	4.58 [2.52; 8.32]	<0.001*
41-43	24(9.23)	2(8.33)	22(91.67)	0.24 [0.05; 1.05]	0.040*
Marital status					
Married	83(31.92)	6(7.23)	77(92.77)	Ref	-
Single	132(50.78)	54(40.90)	78(59.1)	6,12 [3.13; 11.9]	<0.001*
Cohabiting	45(17.30)	7(15.56)	38(84.44)	0.48 [0.2; 1.13]	0.084
Level of education or training					
Secondary	156 (60)	21(13.46)	135(86.54)	Ref	-
Primary	92 (35.38)	41(45.57)	51(97.75)	4.57 [2.53-8.26]	<0.001*
Superior	12(4.62)	5(4.67)	7(95.33)	2.14 [0.66-7]	0.2
Professional status					
Pupil/Student	64(24.62)	19(29.69)	45(70.31)	1,3 [0.69; 2.43]	0.41
Tradeswoman	47(17.08)	21(44.68)	26(55.32)	2.93 [1.51; 5.68]	0.000*
Housewife	85(32.7)	9(10.59)	76(89.41)	0.24 [0.11; 0.52]	<0.001*
Civil servant	18(6.92)	7(38.89)	11(61.11)	Ref	-
Daily	46 (18.68)	11(23.91)	35(76.09)	0.89 [0.42;1.87]	0.78

Area of residence					
Urban	205 (78.9)	46(22.44)	159(77.56)	Ref	-
Rural	55(21.1)	21(38.18)	24(61.82)	3.02 [0.54; 5.1]	0.001*
Number of pregnancies					
0-1	67(25.77)	12(17.91)	55(82.09)	Ref	-
2-3	106(40.77)	39(36.79)	67(63.21)	2.62 [1.4; 4.63]	0.000*
≤ 4	87(33.46)	16(18.39)	71(81.61)	0.54 [0.29; 1.02]	0.054
Age at pregnancy (Quarter)					
1st Quarter	53(23.18)	22(1.8)	31(98.2)	2.55 [1.25; 4.83]	0.003*
2nd Quarter	95 (31.13)	18(14.89)	77(85.11)	0.55 [0.0; 1.01]	0.06
3rd Quarter	112(45.69)	27(5.80)	85(94.20)	Ref	-
Note: Crude OR=Crude Odds Ratio; CI=Confidence Interval; *=Significant test, Ref=Reference					

Table 1: Univariate analysis of the prevalence of *Toxoplasma gondii* infection according to the socio demographic and obstetric characteristics of the pregnant women in the study (n=260).

Discussion

Because of the risk of infection of the foetus or newborn and of reactivation of the infection in immunocompromised individuals, the estimation of *Toxoplasma gondii* seropositivity in pregnant women is important and necessary. The present study revealed an overall seropositivity of 25.77% for *Toxoplasma gondii* infection in pregnant women attending antenatal clinics at the Sino-Gabonese Friendship Hospital in Franceville, from 03 January to 28 November 2022. Although this result is lower than those found, in northwest Ethiopia (68.4%), Cameroon (70%), Kinshasa, Democratic Republic of Congo (80.3%) and finally, that of a previous study in Gabon (80.41%) [16]. It is practically similar to that found in Saudi Arabia (24.1%) [17]. However, the result of the present study is higher than those of studies conducted in Ethiopia (18.5%), Mozambique (18.7%), Sudan (20.2%) and Burkina Faso (20.3%) [18-21]. The variability and discrepancy observed in *Toxoplasma gondii* infection rates from one study to another may be due to differences in the sensitivity of the diagnostic methods used, climatic conditions, and cultural differences in the hygiene, dietary and even literacy habits of the participants in the different studies. Variations in the degree of human contact, management and interaction with definitive hosts and reservoirs of *Toxoplasma gondii*, and lack of awareness of the disease and its transmission may also explain the difference between studies within the same or different populations. Similar to a previous study an association between age and seroprevalence was observed in the present study [22]. Indeed it was observed that pregnant women aged between 31 and 40 years and between 41-43 years were significantly associated with *Toxoplasma gondii* infection. Contrary to a study conducted in Morocco, which indicated that the prevalence of Toxoplasmosis does not increase linearly with age, this result is in line with the observation made in Germany revealing that the prevalence

rates of *Toxoplasma gondii* infection increased linearly with age [23,24]. This may also be justified by the greater temporal risk of exposure with age. Regarding the marital status of the pregnant women in the study, a statistically significant association was found between *Toxoplasma gondii* infection and celibacy (28.13% IgG and 3.13% IgM). This result is similar to that found in a study conducted in Bamenda, Cameroon [25]. However, this result is contrary to that obtained by Ayeah et al. who found no significant association between marital status and seroprevalence of toxoplasmosis in pregnant women [26]. This could be explained by the fact that the celibacy, which is associated with independence and freedom, may mean that this category of women prefers to go out at night, and is therefore exposed to contaminated food (undercooked meat, fruit and salad) and contaminated water served in "street restaurants" [27]. As the level of knowledge and awareness of toxoplasmosis are considered to be very important factors in preventing the risk of toxoplasmosis, the present study observed a statistically significant association between *Toxoplasma gondii* infection and the level of education or primary schooling among the pregnant women in the study. This result is consistent with that found in a study elsewhere [28]. There it was reported that, women who had no knowledge of toxoplasmosis were at higher risk of this infection. This clear observation demonstrates a lack of information characterised by a lack of awareness on the part of the country's health authorities. A significant interaction was also observed between *Toxoplasma gondii* seropositivity and the occupational status of the study participants. Contrary to a study conducted elsewhere, which indicated that female employees/businesswomen were more likely to be infected with *Toxoplasma gondii* than female farmers (40.0% versus 25.9%) [OR=1.9, 95% CI: 1.2-3.0, p=0.006] [29]. Pregnant housewives and shopkeepers in the present study were respectively 0.24 (p<0.001) and 2.93 (p=0.000) times more likely to be *Toxoplasma gondii* seropositive, compared

with other occupations. This result is in agreement with the study conducted in Nigeria [30]. In contrast, a study conducted in Tanzania reported no significant association between occupational status and *Toxoplasma gondii* seropositivity [31]. On the other hand, pregnant housewives and shopkeepers are at greater risk of *Toxoplasma gondii* seropositivity, probably due to greater exposure to contaminated meat, vegetables and fruit, salads, in markets (whether at home, in the cafeteria or restaurant). Contrary to a study which indicated that the prevalence of *Toxoplasma gondii* infection in rural areas (63.1%) was lower than in urban areas (81.3%). The 2nd arrondissement of the city of Franceville, which geographically hosts the study hospital, is surrounded by villages and most women from these rural areas attend antenatal clinics at this hospital [32]. A statistically significant association was also observed between *Toxoplasma gondii* infection and rural area in the present study. This result is in line with that obtained in other countries [33]. This could be explained by the fact that, generally speaking, traditional upbringing in Africa assigns all household tasks to women, during which the risks of parasitic contamination could be higher (handling contaminated water, food of dubious quality, vegetables and fruit, contaminated salads, etc.) [34]. Furthermore, in rural areas, there is close proximity between the population and the livestock in the households. This state of affairs is responsible for the contamination of soils in this area by *Toxoplasma gondii* oocysts, which can survive there for years. Examination of the obstetric results in the present study indicated that pregnant women with between 2 and 3 pregnancies were almost 3 times more likely to be infected with *Toxoplasma gondii* [35]. This result is consistent with a study conducted in France, which established the relationship between infection with this parasite and multigravid women [36]. Furthermore, studies conducted in Saudi Arabia have shown that the seroprevalence of IgG against *Toxoplasma gondii* was significantly correlated with an increase in parity. This could be explained by an increase in women's exposure to the disease [37]. Since infection in the first quarter most often leads to miscarriage, the present study observed a significant association between women who were in the first trimester of their new pregnancy and *Toxoplasma gondii* infection. This result is consistent with the fact that risk increases with gestational age [38]. On the other hand, previous studies have shown that *Toxoplasma gondii* infections acquired by the mother during the first quarter of gestation have a 10%-15% risk of congenital transmission to the fetus with serious consequences, whereas mothers acquiring the infection during the second or third quarter has an increasing risk (up to 68%) of infecting her unborn baby, with less serious consequences [39]. This indicates that the category of HIV-positive pregnant women identified in this study presented a risk of transmitting the infection to their fetus.

Strengths and limitations of the study

- This is the first study of infection in pregnant women at the Sino-Gabonese Friendship Hospital in Franceville.
- It provides information on the immunological status against of pregnant women in this referral hospital, which has been unexplored until now.
- An average seroprevalence of infection was found among the pregnant women studied.
- Ongoing work is showing that risk factors for infection found in pregnant women can help plan measures against toxoplasmosis and its sequelae.

- The low rate of seropositivity to has not made it possible to find other associations between the characteristics of pregnant women and infection.
- The use of the rapid diagnostic immunological test for the simultaneous detection and differentiation of anti-*Toxoplasma gondii* (T. *gondii*) IgG and IgM, would have been appropriate.

Conclusion

The results obtained in this study revealed a high seropositivity to *Toxoplasma gondii* among pregnant women in the study area. Among the predictors evaluated in this study, having an average monthly income, professional status, the habit of washing hands after handling raw meat and sources of water for drinking proved to be significant predictors that increase the risk of *Toxoplasma gondii* seropositivity in pregnant women. Consequently, further studies on bioassays, isolation and genotyping of the pathogen are crucial. This also requires measures to prevent and control *Toxoplasma gondii* infection in pregnant women, which poses a potential threat to the foetus.

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