

Seroprevalence of toxoplasmosis among pregnant women in Mbouda and protestant hospitals of Ngaoundere, Cameroon

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Abstract

Toxoplasmosis is one of the most widespread zoonoses in the world. Although it is usually benign, it seriously affects pregnant women. It is caused by an obligate intracellular protozoan called *Toxoplasma gondii*. We carried out a prospective study involving blood samples from 100 pregnant women aged between 18 and 45 years. These patients presented for a prenatal consultation at the maternity ward and laboratory at the District Hospital of Mbouda and the Protestant Hospital of Ngaoundere, during a period of two months (June 19-August 15, 2022). Our study revealed an overall seroprevalence of 47.8 2 % by the immunochromatography technique. Through this study, it appears that the frequency of women being in their first trimester of pregnancy is 81.81 % and 63.63 %; 88 % and 83.33 % of the toxoplasmosis-positive population have already had a miscarriage at the District Hospital of Mbouda and the Protestant Hospital of Ngaoundere respectively. Concerning the presence of animals, age group and profession of women, many women with domestic animals are the sickest, i.e. 65.51%, women whose age group is between [26-33[and [18 - 25[were the most affected and those working as housekeepers were less affected than those working outside the home. Ultimately, toxoplasmosis causes miscarriages in pregnant women, the consumption of raw vegetables and the presence of a domestic animal (cat) in the home remain the important factors in contamination by the parasite (District Hospital of Mbouda and the Protestant Hospital of Ngaoundere).

Keywords: Toxoplasmosis, Seroprevalence, pregnant women, Associated factors, Immuno chromatography.

1. Introduction

Toxoplasmosis is a parasitic disease caused by the intracellular protozoan *Toxoplasma gondii*. It is a cosmopolitan anthroozoonosis common both to animals and humans [1, 2]. This parasite is transmitted to humans through ingestion of food or water contaminated by oocysts excreted by domestic animals, or by poorly cooked meat containing tissue cysts [3]. Young children can become infected by accidentally ingesting contaminated soil. In pregnant women, primary infection can lead to congenital toxoplasmosis. The placenta represents an intermediate target of infection, from which the fetus is contaminated. The risk of maternal-fetal transmission estimated at 29 % increases with the age of pregnancy (from around 6 % at 13 weeks to around 72 % at 36 weeks) [4]. The risk of transmission is estimated at 15-20 % during the first trimester, compared with 60-90 % during the third trimester [5].

It is a parasitosis most often responsible for an inapparent or benign infection in the immunocompetent subject, but it can be serious in the immunocompromised and during pregnancy through transplacental passage of the parasite, exposing the fetus to congenital toxoplasmosis [6]. Acquired toxoplasmosis is generally asymptomatic, except in immunocompromised patients, whereas congenital toxoplasmosis has serious consequences for the fetus, the newborn and the child if not treated quickly. The manifestations of congenital toxoplasmosis are diverse: spontaneous abortions, fetal death in utero, and congenital malformations in the newborn, the most frequent of which are cerebral (hydrocephalus, microcephalus, cerebral calcifications) [7]. It is a widespread public health problem in both developed and developing countries [8].

According to the WHO, the annual incidence of congenital toxoplasmosis worldwide is estimated at 190.100, with a prevalence of 1.5 per 1.000 live births [9]. Seroprevalence varies considerably from country to country, with the highest figures (>50 %) found in France, Latin America and Sub-Saharan Africa. In the United States, the figure is 15 %, and between 20% and 40% in Canada [10]. In Cameroon, a study by Giroud (2004) and Guemgne Tadjom (2019) found a prevalence of 77 % and 45.5 % respectively. These high prevalences may be due to the fact that this condition is often unrecognized in our regions and its screening is sometimes neglected [11]. Studies carried out in recent years have shown that serological monitoring during pregnancy combined with chemoprophylaxis in the event of proven seroconversion significantly reduce the risk and severity of fetal infection, hence the necessity for systematic prenatal infection screening and seroprevalence monitoring at national level. Thus, the need for early diagnosis for better management and determination of seroprevalence for the development of a good policy by the Ministry of Public Health.

The aims of this study were to determine the seroprevalence of toxoplasmosis among pregnant women at the District Hospital of Mbouda and the Protestant Hospital of Ngaoundere to describe sociodemographic factors, to assess the level of parasitological knowledge of these pregnant women and to determine risk factors for this pathology.

2. Material and methods

2.1. Duration of study

We conducted a prospective study. This study was carried out over a period of 2 months, from June 19 to August 15, 2022, at the Laboratory of the Protestant Hospital of Ngaoundere and the District Hospital of Mbouda.

2.2. Study population

Our study concerns all pregnant women attending antenatal clinics and showing up at the laboratory at District Hospital of Mbouda and at that of Protestant Hospital of Ngaoundere for antenatal tests.

2.3. Selection criteria

-inclusion criteria : were included in our study : all women attending the district hospital of Mbouda and protestant hospital of Ngaoundere for maternity consultations.

-Non -inclusion criteria : were excluded from our study : all women who came to the maternity ward of the District Hospital of Mbouda and the Protestant Hospital of Ngaoundere for consultation but were not pregnant.

2.4. Sampling technique

For the conduct of our study, we used a random sampling method to include patients in the study as they came to the laboratory.

2.5. Sample size

We were able to achieve a sample size of 100 participants, including 50 at the District Hospital of Mbouda and 50 at the Protestant Hospital of Ngaoundere, corresponding to the number of patients received during the data collection period.

2.6. Method of collection data

Data collection was based on a data collection form and a questionnaire designed to assess socio-demographic parameters (age, sex), knowledge of toxoplasmosis, the dangers it can cause to the foetus, and risk factors. The authorization of the directors of two hospitals and the free and informed consent of all pregnant women who agreed to take part in our study enabled us to take blood samples for laboratory analysis. Blood sampling was performed at the elbow or on the palm of the hand, at the level of a vein. After asepsis of the puncture site using a syringe, the blood sample was introduced into a dry tube.

2.7. Analytical phase

The TOX IgG-IgM test kit device is highly specific to antigen antibody and immuno-chromatographic assay reactions for the qualitative detection of *Toxoplasma gondii*-IgG and IgM in patient serum or plasma. The kit contains recombinant *Toxoplasma gondii* IgM antigen pre-fixed to the membrane in the test region (T1), recombinant *Toxoplasma gondii* IgG antigen pre-fixed to the membrane in the test region (T2), and the corresponding antibodies in the quality control region. The sample (serum/plasma) was deposited in the sample well of the cassette, then mixed with the pre-coated colloidal gold particles in the test.

This mixture migrated chromatographically along the test strip and interacted with immobilized anti-human IgG and IgM. If positive, colloidal gold will bind to anti-toxoplasma gondii antibodies during the chromatography process, then the mixture has been read to the antigen which has bound to the membrane, a red line has appeared in the T test region (T1 and T2 or T1/T2). If negative, there will be no red line in the test region (T1 and T2). The control line indicates the kit's validity [12].

2.8. Post analytical phase

Patient results were recorded in a register and then kept in the secretariat for scanning before being communicated to patients.

2.9. Statistical Analysis

All data collected were entered and curves plotted on Excel 2016. Seroprevalence of toxoplasmosis in pregnant women was defined as the quotient of the number of women with positive serology by the total number of pregnant women tested during that period. The seroprevalence rate was determined as seroprevalence multiplied by 100.

3. RESULTS

3.1. Distribution of population according to their age

The 100 pregnant women aged between 18 and 45 were grouped together to determine the age distribution of the study population, the results of which are shown in Figure 1 (A and B). Figure 1A shows the distribution of subjects according to age. This figure shows that women aged between 26 and 32 are the most represented (50 %), while those aged between 38 and 45 are the least represented (6 %). According to figure 1B, a very large proportion of the women we met were aged [18-25], with a percentage equal to 36 %. This is followed by proportions equal to 32 % for women aged [26-32], while women aged [33-38] represent only 20 % of the study population. The lowest percentage is found among women aged [38-45], with a proportion of 6 %.

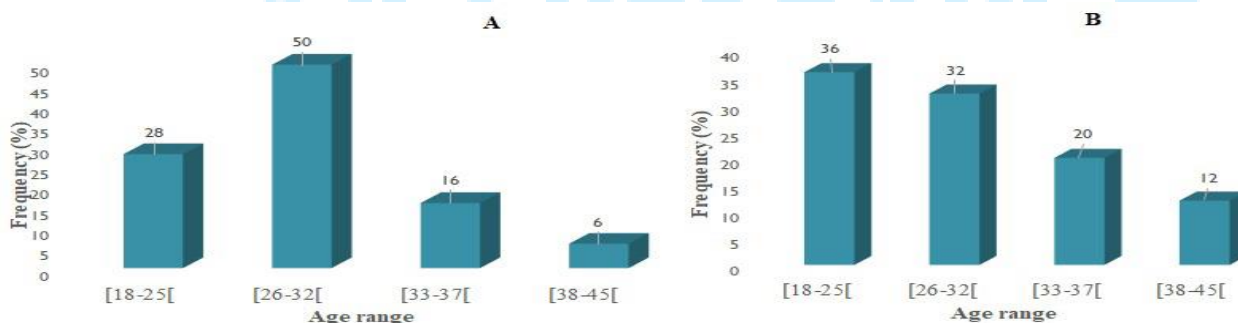


Figure 1: Distribution of population according to their age (A = District hospital of Mbouda et B = Protestant hospital of Ngaoundere)

3.2. Distribution of the study population according to gestational age

Figure 2A shows the distribution of the study population according to consultation. It shows that women who would have come for CPN1 are more represented (88 %), whereas there are 6 % of women in CNP2 and CPN3 at District hospital of Mbouda. The results in figure 2B show that the percentage of CPN2 is 16.20 % and 39 % for CPN3, while the majority of patients are from CPN1 (44.80 %).

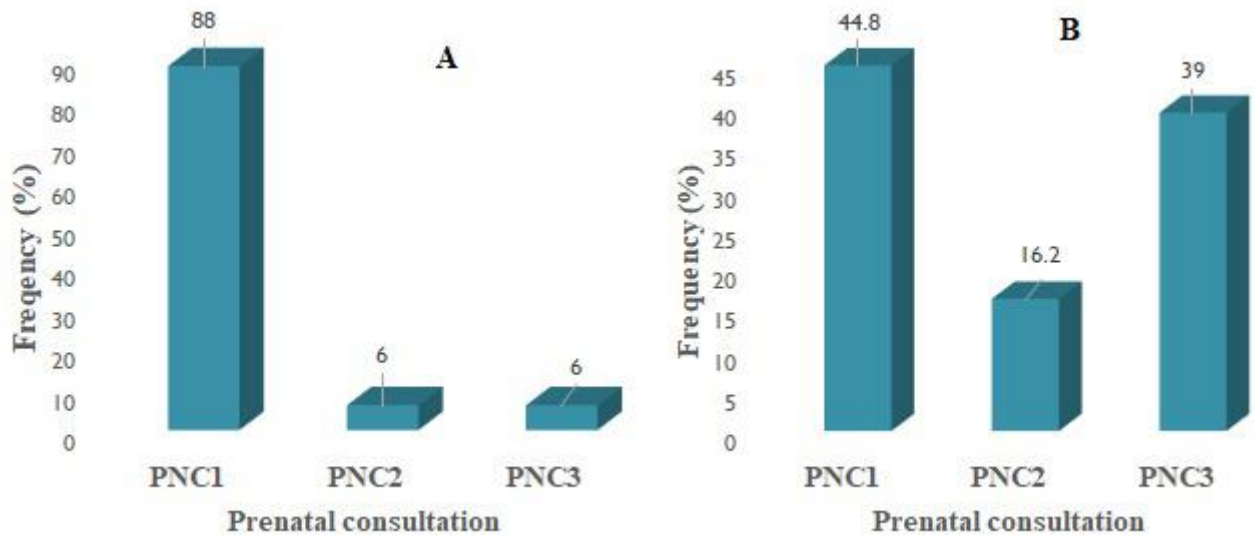


Figure 2: Distribution of the study population according to gestational age (A = District hospital of Mbouda et B = Protestant hospital of Ngaoundere)

Notes: PNC1 = Prenatal Consultation first trimester of pregnancy, PNC2 = Prenatal Consultation second trimester of pregnancy, PNC3 = Prenatal Consultation third trimester of pregnancy

3.3. Global prevalence of toxoplasmosis according to immunochromatography technique

Figure 3A shows that (46 %) of women have a long-standing toxoplasma infection and (54 %) have no serological evidence in favor of toxoplasma (District Hospital of Mbouda). Figure 3B shows that (41.66 %) of women had a long-standing toxoplasma infection and (58.34 %) had no serological evidence in favor of toxoplasmosis.

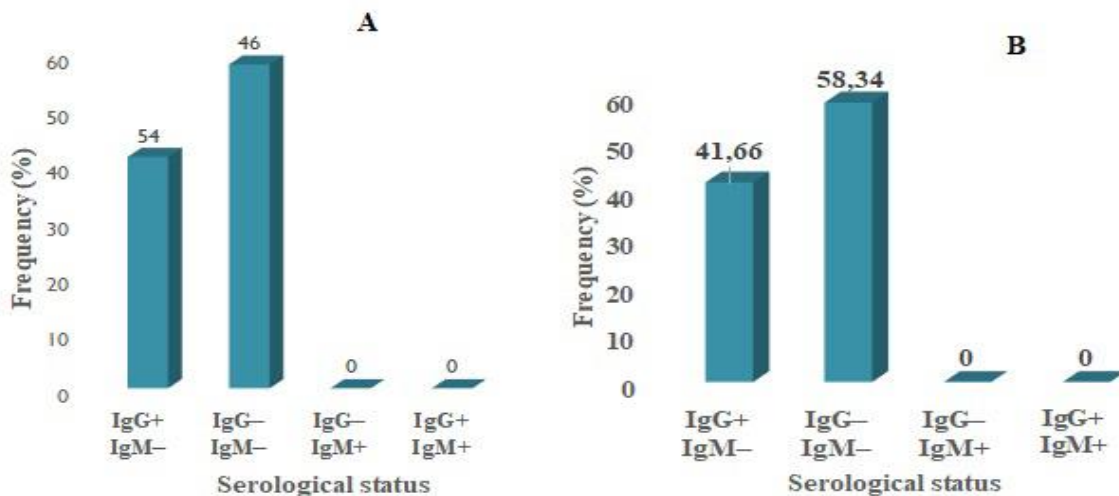


Figure 3 : Global prevalence of toxoplasmosis according to immunochromatography technique (A = District hospital of Mbouda et B = Protestant hospital of Ngaoundere)

3.4. Distribution of the study population according to serological status and age

The table 1 presents the seroprevalence of toxoplasma IgM and IgG in pregnant women who participated in this study according to serological status and age respectively at the District hospital of Mbouda and protestant hospital of Ngaoundere. Analysis of Table 1 shows that most women with positive serology (IGM-, IGG+) are aged between 38 and 45. Table 1 shows that more serology-positive women are aged [38-45], with 66.6 % at District Hospital of Mbouda and 10 % at Protestant Hospital of Ngaoundere.

Table 1: Distribution of the study population according to serological status and age

Age	Seropositif		Seronegatif	
	IgG	IgM	IgG	IgM
District Hospital of Mbouda				
[18-25[47.3 %	0 %	56.6 %	100 %
[26-32[44.4 %	0 %	55.5 %	100 %
[33-37[37.5 %	0 %	62.5 %	100 %
[38-45[66.6 %	0 %	33.3 %	100 %
Protetant Hospital of Ngaoundere				
[18-25[12 %	0 %	88 %	100 %
[26-32[44 %	0 %	56 %	100 %
[33-37[26 %	0 %	74 %	100 %
[38-45[10 %	0 %	90 %	100 %

3.5. Distribution of population according to gestational age

Analysis of Table 2 shows that seropositive subjects at Protestant Hospital of Ngaoundere are found in the first (69.22 %), second (17.32 %) and third trimester (100 %) respectively. Analysis of data from the district hospital of Mbouda shows that seropositive subjects are found in the first (38.10 %), second and third trimesters of pregnancy (100 %) respectively.

Table 2: Distribution of population according to gestational age

Reason for consultation	Seropositif		Seronegatif	
	IgG	IgM	IgG	IgM
District Hospital of Mbouda				
PNC1	38.10 %	0 %	61.9 %	100 %
PNC2	100 %	0 %	0 %	100 %
PNC3	100 %	0 %	0 %	100 %
Protestant Hospital of Ngaoundere				
PNC1	69.22 %	0 %	30.78 %	100 %
PNC2	17.32 %	0 %	82.68 %	100 %
PNC3	100 %	0 %	0 %	100 %

Notes: PNC1 = Prenatal Consultation first trimester of pregnancy, PNC2 = Prenatal Consultation second trimester of pregnancy, PNC3 = Prenatal Consultation third trimester of pregnancy

3.6. Distribution of miscarriages according to serological status

Figure 4 A shows that (81 %) of the toxoplasmosis-seropositive population had already suffered a miscarriage and (19 %) of the toxoplasmosis-seropositive population had never suffered a miscarriage at District Hospital of Mbouda. On the other hand, at the Ngaoundere Protestant Hospital, figure 4 B shows that (52.1 %) of the toxoplasmosis-seropositive population had already suffered a miscarriage and (47.9 %) of the toxoplasmosis-seropositive population had never suffered a miscarriage. Figures 4A and 4B show that *Toxoplasma gondii* causes miscarriage in pregnant women.

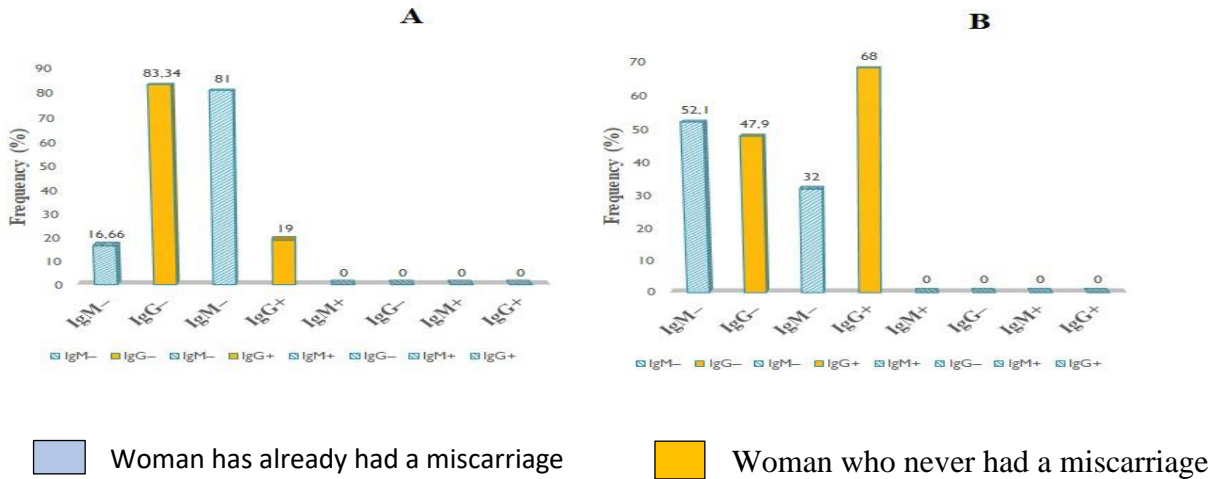


Figure 4: Distribution of miscarriages according to serological status

3.7. Factors favoring the occurrence of toxoplasmosis

The patients' socio-dietary data are presented in Table 3. These data take into account the presence of a pet at home, the type of animal, the consumption of raw vegetables and the consumption of rare meat. For the presence of pets at home, we note a high frequency (58 % and 34.48 %) of the population with one or two pets. People with one or two pets are more affected than those without, with respective seropositivity rates of 65.51 % and 34.48 %, and 41.91 %, 19.78 % and 80.22 % respectively at the district hospital of Mbouda and protestant hospital of Ngaoundere.

Raw vegetables were consumed by 98 % and 100 % of the subjects, unlike rare meat, which was consumed by 3 %. We also note that 47.82 % and 44.89 % of raw vegetable consumers are toxoplasmosis positive (District hospital of Mbouda and protestant hospital of Ngaoundere).

Tableau 3: Factors favoring the occurrence of toxoplasmosis

Features	Modality	Study population		Toxoplasmosis			
				IgG+	IgG-	IgM+	IgM-
		N	%	%	%	%	%
District Hospital of Mbouda							
Domestic animal	Yes	29	58	65.51	34.48	0	100
	No	21	42	19.04	80.95	0	100
Type of animals	Cat	28	97	64.28	35.71	0	100
	Dog	1	3	100	0	0	100
Consumption of raw vegetables	Yes	49	98	44.89	55.50	0	100
	No	1	2	100	0	0	100
Consumption of rare meat	Yes	1	2	100	0	0	100
	No	49	98	44.89	59.10	0	100
Protestant Hospital of Ngaoundere							
Domestic animal	No	27	41.91	19.78	80.22	0	100
	Yes	1	66.56	90,01	9.99	0	100
Types of animals	Cat	30	50.30	54.93	45.07	0	100
	Dog	11	2	70.21	29.79	0	100
Consumption of raw vegetables	No	3	17	100	0	0	100
	Yes	20	100	41	59	0	100
Consumption of rare meat	No	21	77.10	100	0	0	100
	Yes	25	22.90	35	65	0	100

N: Effective

3.8. Level of knowledge of the study population about toxoplasmosis

Table 4 shows the level of knowledge of the study population about toxoplasmosis in association with the prevalence of toxoplasmosis. These risk factors are knowledge of toxoplasmosis, danger to the fetus, and whether the examination was performed beforehand. This table shows that, out of 100 women studied at the District Hospital of Mbouda, 57.6 % and 34.72 % had undergone the examination beforehand, but only 42.5 % and 50 % had ever heard of toxoplasmosis. In the present study,

57.5 % and 50 % of the latter were seropositive for toxoplasmosis at Mbouda District Hospital and Ngaoundere Protestant Hospital respectively. Also, 20 % of the study population were unaware of the dangers to which fetuses are exposed in the event of seropositivity.

Tableau 4: Level of knowledge of the study population about toxoplasmosis

Features	Modality	Study population		Toxoplasmosis			
				IgG+	IgG-	IgM+	IgM-
		N	%	%	%	%	%
District Hospital of Mbouda							
Knowledge about toxoplasmosis	Yes	38	58	57.5	42.5	0	100
	No	21	42	13.5	18/42	0	100
Danger to the fetus	Cat	28	97	45	55	0	100
	Dog	1	3	50	50	0	100
Completed the exam before	Yes	49	98	42.5	57.5	0	100
	No	1	2	53.34	46.66	0	100
Protestant Hospital of Ngaoundere							
Knowledge about toxoplasmosis	Yes	14	60.56	65.28	34.72	0	100
	No	9	39.13	22.23	77.77	0	100
Danger to the fetus	Cat	8	57.14	62.5	37.50	0	100
	Dog	6	42.85	66.66	33.33	0	100
Completed the exam before	Yes	23	100	50	50	0	100
	No	0	0	0	0	0	100

N: Effective

3.9. Distribution of serological status according to occupation

Table 5 shows that housewives are less affected than all other women: 40.91 % and 44 % of positive cases for housewives; 50 % and 47.4 % of positive cases for other women at District Hospital of Mbouda and Protestant Hospital of Ngaoundere respectively.

Tableau 5: Distribution of serological status according to occupation

Occupation	Effective	Frequency	Serological status			
			IgG+	IgG-	IgM+	IgM-
District Hospital of Mbouda						
Housewife	22	%	40.91 %	59.09 %	0 %	100 %
Non-housewife	28	%	50 %	50 %	0 %	100 %
Protestant Hospital of Ngaoundere						
Housewife	17	%	44 %	56 %	0 %	100 %
Non-housewife	33	%	47.4 %	52.6 %	0 %	100 %

4. Discussion

Toxoplasmosis is a ubiquitous parasitosis, the most serious form of which is congenital toxoplasmosis acquired during pregnancy. Prevention of this form relies on serological screening, antenatal diagnosis and management of fetal infection. According to the literature, measures of *Toxoplasma gondii* seroprevalence differ from one study to another. The aims of this study were to determine the seroprevalence of toxoplasmosis in pregnant women attending antenatal clinics at the District Hospital of Mbouda and the Protestant Hospital of Ngaoundere, and to attempt to establish the factors favouring the occurrence of this parasitosis.

In a total of 100 pregnant women surveyed for toxoplasma serology, 22 were toxoplasma antibody carriers, i.e. a prevalence of 54 %, and 17, i.e. a prevalence of 41.66 %, respectively at District Hospital of Mbouda and Protestant Hospital of Ngaoundere. Recent studies confirm this trend. In northern Tanzania, they account for 56.7 % of the 67 women surveyed [13] and in Dakar by Faye et al. [14], 40.2 %. This value is roughly equal to that obtained by Guemgne in 2019 [15] in Bandjoun in the same region (45.5 %) and lower than the national prevalence in Cameroon (77 %) 17 years ago [16]. This drop in seroprevalence is thought to be due to the efforts made by public health authorities to eradicate this parasite. Seropositivity increases with age, but not with maternity.

In our study, the seronegativity rate was 46 % and 58.34 % respectively at District Hospital of Mbouda and Protestant Hospital of Ngaoundere. This shows that a non-negligible proportion of pregnant women are at high risk of contracting the disease during pregnancy, and consequently of passing the parasite to the fetus. The fact that the test is only prescribed in the first trimester of pregnancy means that we are unable to assess cases of seroconversion in pregnant women. However, contamination can occur throughout pregnancy, with the consequent possibility of infection of the fetus after the first trimester

[11]. In our study, we noted that seroprevalence increased with patient age: 47.3 % for the [18-25[age group and 66.6 % for the [38-45[age group. Several studies show that toxoplasmosis seroprevalence increases with age. In France, studies have shown an increase in toxoplasmosis seroprevalence in pregnant women of French nationality with age [17]. A study carried out in Brazil in 2017 also showed that seroprevalence increases linearly with age [18]. Similar observations have been made in Nigeria [19] and Morocco [20]. These findings corroborate those of Singh in 2004, for which prevalence was 38.5 % for the [20-25[age group and 77.8 % for the [35-39[age group. This increase in seroprevalence with the age of the patient is thought to be due to the increased duration of exposure to the disease's risk factors. This highlights the importance of educating young women of childbearing age about the risk factors for toxoplasma infection. After analyzing toxoplasma seroprevalence in pregnant women, we directed our attention to assessing the link between toxoplasmosis and miscarriage. Spontaneous miscarriage, late fetal death or congenital anomalies may occur. Some known cases of abortion are thought to be due to toxoplasmosis. The results showed that 81 % and 32 % of women who had already miscarried were seropositive for toxoplasmosis at District Hospital of Mbouda and Protestant Hospital of Ngaoundere respectively. Adjé's study in Senegal showed that 22.6 % of women who had abortions had anti-toxoplasma antibodies. The frequency of spontaneous abortions reported during our study was higher than that reported in Nigeria (22.8 %) [21]. This difference is not related to methodology, as Hany et al [22] also used ELISA as their test method. This observed seropositivity could be one of the causes of unexplained abortions in the female population surveyed. Indeed, the presence of IgG is indicative of chronic toxoplasmosis infestation, which is one of the main causes of miscarriage [23]. Some cases of abortion in pregnant women may be due to neosporosis.

Although developing the prevalence of toxoplasmosis is a major concern, determining risk factors remains an important element in identifying this parasitosis. Several authors have demonstrated the difference in seropositivity between stray and domestic cats. Indeed, stray cats have a higher seropositivity than domestic cats. Milan et al [24] assert that the main source of environmental contamination could be stray cats. Studies carried out on stray cats in Bangkok [25] and Tehran [26] found prevalences of 75 % and 90 % respectively. These trends are not consistent with the data obtained. We found that 65.51 % of women with anti-toxoplasma antibodies have pets (cats), whereas 19.04% of these women have no pets.

Multivariate analysis enabled us to identify raw vegetable consumption as a factor associated with toxoplasmosis. Consumption of raw vegetables is not outdone: 44.89 % of women who consume raw vegetables have a positive toxoplasmosis serological status, which remains an important factor. Consumption of rare meat, on the other hand, results in 100 % toxoplasmosis-positive women at the district hospital of Mbouda and the Protestant hospital of Ngaoundere respectively. However, recent studies in Benin have shown that pig and goat meat intended for human consumption was parasitized by *T. gondii* in 25 % and 53 % of cases respectively [27, 28]. Several studies have shown that the presence of cats in the household and the consumption of poorly cooked meat are potential risks for the acquisition of anti-toxoplasmic antibodies [29, 30]. This may be explained by the fact that the cat completes the parasite's cycle, releasing oocysts into the environment [31]. Consequently, the consumption of raw vegetables and the presence of cats in the home are risk factors for toxoplasma infection.

As for the level of knowledge about toxoplasmosis in the District Hospital of Mbouda, 57.89 % are affected; this could be due to the fact that women have acquired this knowledge because they had already come into contact with the parasite.

Women unaware of the danger of this parasite for their foetus are the most affected. Housewives were less affected, with prevalences of 40.91 % and 44 %, compared with 50 % and 47.4 % for non-housewives at District Hospital of Mbouda and Protestant Hospital of Ngaoundere respectively. However, Sissinto and Tové [32] found a significant association between occupation as a businesswoman or office worker and toxoplasmosis ($p = 0.006$), showing the impact of economic level on dietary habits [33]. This may be explained by the fact that women who have to leave the house to work often eat out, sometimes in unhygienic conditions. Consumption of undercooked meat and vegetables, and lack of knowledge about toxoplasmosis, its modes of transmission and means of prevention, are the main risk factors for transmission of the disease.

Conclusion

Toxoplasma gondii is one of the most widespread parasites not only geographically, but also zoologically. Congenital toxoplasmosis occurs in pregnant women infected with *T. gondii* for the first time during pregnancy, when the parasite crosses the placenta and infects the fetus. The data obtained have enabled us to gain a better understanding of toxoplasmosis in the Adamawa and West regions in terms of seroprevalence among pregnant women, and the consumption of raw vegetables, rare meat and the presence of cats in the home have been significantly identified as risk factors linked to contamination. Seroprevalence of toxoplasmosis among pregnant women attending antenatal clinics was 47.82 % and 46 % at District Hospital of Mbouda and Protestant Hospital of Ngaoundere respectively. The present study shows that 83.33 % and 63.63 % of the toxoplasmosis-seropositive population had already suffered a miscarriage, and 65.51 % and 60.86 % of women with pets were seropositive at Mbouda District Hospital and Ngaoundere Protestant Hospital respectively.

The results of our survey have enabled us to gain a better understanding of toxoplasmosis in the Adamawa and West regions, in terms of seroprevalence among pregnant women, and to identify the main risk factors associated with contamination. Socio-demographic factors, notably the consumption of raw vegetables and rare meat, and the presence of cats in the home, all contribute to an increase in the prevalence of toxoplasmosis. We recommend the implementation of a national strategy focused on serodiagnosis and education of seronegative pregnant women in order to reduce the incidence and prevalence of toxoplasmosis and its serious consequences for the fetus.

Conflicts of Interest

The authors declare no conflicts of interest

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