

Seroepidemiology of toxoplasmosis in pregnant women attending antenatal clinics at the center for maternal and child health care in Daloa in Ivory Coast.

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ABSTRACT

Background: Serological tests remain the basis of toxoplasmosis diagnosis since direct detection of *Toxoplasma gondii* by microscopy is less sensitive and PCR is inaccessible for routine screening. Unfortunately, serological screening in pregnant women for *T. gondii*-specific antibodies is not mandatory in the assessment of antenatal care in Ivory Coast. Furthermore the latest data published are about a decade.

Aim: To determine the seroprevalence in relation with certain risk factors and discuss epidemiological issue of different patterns seen among pregnant women in Daloa, in Ivory Coast.

Study design and setting: This is a prospective study. Pregnant women attending antenatal care clinics in maternal and child health care center were screened for IgG and IgM antibodies against *T. gondii* from February to August 2014.

Results:

Out of 385 women, 113 (29.35%) were in their first pregnancy and the average age was 26 years with 14 and 47 years of age respectively for the youngest and the oldest mother. A hundred and sixty five (42.85%) of the women had ever been in contact with cats and 305 (79.22%) had ever eaten raw vegetables. 108 (28.05%) out of 385 women were in the third trimester of pregnancy.

226 out of 385 sera (58.70%) had positive IgG and all sera were tested negative for IgM. The remaining sera 159 (41.3%) had no IgG or IgM. The seroprevalence of IgG was correlated to the presence of cats in the household ($P < 0.001$) and age group ($P < 0.05$), whereas no significant association was observed with the consumption of raw vegetables, education and the type of housing. Out of the 159 sera tested negative, 45 (28.3%) were collected in the third trimester of pregnancy.

Conclusion: Our study shows that the prevalence of toxoplasmosis among women of reproductive age, particularly pregnant women is not insignificant in Ivory Coast and deserves more attention from the health officials.

Keywords: Toxoplasmosis, seroprevalence, epidemiology, Daloa, Ivory Coast.

1. INTRODUCTION

Toxoplasmosis is among the global major zoonotic diseases [1, 2]. It is the third leading infectious cause of food-borne and waterborne death after salmonellosis and listeriosis [3] and affect about one third of the humanity [4]. The disease is caused by *Toxoplasma gondii*, an Apicomplexa protozoan parasite [5], with cats as the definitive host, and warm-blooded animal as intermediate hosts [6]. Humans can be infected by contact with infected animals, eating raw food contaminated with oocysts, such as vegetables and fruits but also by water and meat containing encysted bradyzoites [7]. The seroprevalence of toxoplasmosis varies substantially depending on sanitation level of the population, dietary habits, contact with cats, contact with contaminated soil [8, 9], *T. gondii* virulence [10] and varies according to geographic areas [11].

The majority of infections are asymptomatic in immunocompetent individuals, possibly because the organism has co-evolved with its many vertebrate hosts and has developed multiple strategies to persist asymptotically for the lifetime of the host [12]. Its severity lies in cases where the parasite invades immunocompromised subjects [7] and pregnant women. In this latter case, it may cause a serious type of foetopathy hypotonia, microcephaly, chorioretinitis [13]. In women, primary infection with *T. gondii* during the third trimester of pregnancy carries a higher risk of congenital transmission than if it is acquired during the first trimester [14, 15]. Antenatal serological screening of *T. gondii* infection based on IgG and IgM detection is the mainstay in monitoring the risk for congenital toxoplasmosis [16]. The direct demonstration of parasites (microscopy, mouse inoculation, cell culture, PCR) is tricky and inaccessible in the context of routine screening [17, 8].

Besides the major African endemic diseases such as malaria, trypanosomiasis and schistosomiasis, toxoplasmosis may seem secondary, even if it affects 14-92.5% of patients across countries [19, 20, 21]. Indeed, if pregnant women in developed countries benefit from a special monitoring where *Toxoplasma gondii* serology is one of the examinations required in early pregnancy [22], this test is not mandatory in the assessment of pregnancy in many African countries [23,24]. The lack of public health schemes to manage the spread of this pathogen places African women populations at risk of ongoing and possibly increasing incidence and prevalence, as well as a corresponding increase in mortality and morbidity due to toxoplasmosis. In Ivory Coast, there is no screening program during antenatal care for pregnant women and the most recent study available in the literature dates back ten years [23] although the incidence on pregnancies and newborns is not insignificant. Therefore this study aims to determine the serological status in a population of pregnant women from the city of Daloa in the Center-West of Ivory Coast, to draw the appropriate epidemiological consequences.

2. MATERIALS AND METHODS

2.1 Study Design

This is a prospective study carried out at the maternal and child health care center in Daloa among patients attending antenatal care who gave their consent, from February to August 2014.

2.2 Study Population and Serological diagnosis

Random sampling was used in selecting pregnant women attending antenatal clinics without notion of prior serological results. They were asked for consent and those who consented were interviewed and sera were collected and stored at -20°C prior to Enzyme Linked Fluorescent Assay (ELFA) test. The lab technician routinely informed a questionnaire including socio-demographic factors such as age, type of housing, presence of cats, consumption of raw fruit and vegetables, trimester of pregnancy and number of pregnancy. The sample size was determined according to the formula $N = \frac{Z^2 \cdot P \cdot (1-P)}{E^2}$ [25].

N=minimum sample size required for the study; Z=the standard normal deviation, usually 1.96 at 95% confidence level; P=prevalence rate 60% prevalence of toxoplasmosis in women [23]. E= precision rate (5%), degree of accuracy. Therefore N=368 parturient women (but 385 were used).

Toxoplasmosis IgG **rate** was conducted by ELFA technique on a controller Vidas^R (BioMérieux^R). Results are expressed in International Units per mL. Serology with an IgG **rate** greater than 8 was considered positive. The Toxo **IgM ISAGA** test (BioMérieux^R) allowed the specific detection of anti-toxoplasma IgM by immunocapture technique.

2.3 Data Analysis

Sociodemographic data related with serology results were registered in an excel database and analyzed by a chi-square statistical test using STATA ($\alpha < 5\%$) [26].

2.4 Ethical Issues

This study was approved by the Research National Ethics Committee **under the N° 0308/MSLS/CNER-P** before implementation. All participants provided written informed consent. The tests were conducted free of charge for women wishing to participate in the study.

3. RESULTS

3.1 Sociodemographic data

In this prospective study, 385 sera were collected from pregnant women attending the maternal and child health care centre. **Out of** the 385 women, 113 (29.35%) were in their first pregnancy and the average age was 26 years with 14 and 47 years of age respectively the youngest and the oldest mother. **165** (42.85%) of the women **had ever** been in contact with cat and 305 (79.22%) had ever eaten crude vegetable. **108** (28.05%) out of 385 women were in the third trimester of pregnancy (Table 1).

3.2 Serological data

Out of the 385 sera tested, 226 (58.70%) had positive IgG and all sera were tested negative for IgM. The remaining sera 159 (41.3%) had no IgG or IgM. The seroprevalence of IgG was correlated to the presence of cats in the household ($P < 0.001$) and age group ($P < 0.05$), whereas no significant association was observed with the consumption of raw vegetables, education and the type of housing. **Out of** the 159 five sera tested negative, 45 (28.3%) were collected in third trimester of pregnancy (Table 1).

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Table 1. Distribution of *Toxoplasma gondii* seroprevalence along with Sociodemographic characteristics

Demographic characteristics	IgG seroprevalence		X ²	p-value
	Number examined	Number of positive (%)		
Age group (Year)				
14-23	158	89(56.32)	7.04	0.02
24-33	190	108(56.84)		
34-43	37	29(78.37)		
Type of housing				
Common home	215	130(60.46)	1.41	0.49
Individual housing	170	96(56.47)		
Education				
Illiterate	176	109(61.93)	2.44	0.48
Primary	115	61(53.04)		
Secondary	76	46(60.52)		
University	18	10(55.56)		
Gravidity				
Primigravid	113	63(55.75)	0.89	0.63
Multigravid	143	88(61.53)		
Grandmultipare	129	75(58.13)		
Cat				
No	220	108(49.09)	19.9	0.0001
Yes	165	118(71.51)		
Consumption of raw vegetable				
No	80	47(58.75)	0.001	0.99
Yes	305	179(58.68)		
Trimester of pregnancy				
First Trimester	136	77(56.61)	0.72	0.69
Second Trimester	141	86(60.99)		
Third trimester	108	63(58.33)		

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118 DISCUSSION

119 This study was conducted, at the center for maternal and child health care, in Daloa city, in
 120 Ivory Coast, among pregnant women without a notion of prior serological results. The aim of

our study was to determine the initial serological status of parturient women and to determine actions to be taken. The overall seroprevalence of toxoplasmosis in our population (58.7%) is statistically consistent with that reported on women of reproductive age in previous study in Abidjan, the economic capital of Ivory Coast [23] and may be stable on a decade from south to west of the country.

When comparing the toxoplasmosis seroprevalence of our study to those found in west-African countries were literature do exist for at least the latest decade (2004-2013), it appears that toxoplasmosis seroprevalence is very higher in Ghana and low in Nigeria, Burkina Faso and Benin (Tableau 2).

Table 2. *Toxoplasma gondii* seroprevalence in West African women from 2004 to 2014

Year	Country	Sample size	Prevalence (%)	References
2004	Ivory coast	1025	60.00	Adou-Bryn et al[23]
2006	Burkina Faso	336	25.30	Simpore et al[27]
2009	Ghana	159	92.50	Ayi et al, [19]
2011	Nigeria	276	32.60	Deji-Agboola et al[28]
2011	Senegal	941	34.50	N'Diaye et al[29]
2012	Burkina Faso	306	31.00	Bamba et al[30]
2013	Nigeria	173	27.75	Alayande et al[31]
2014	Benin	283	30.00	De Paschale et al[32]

This variability between countries of the same region may have climate causes. Indeed, Adoubryn et al[23] had already noted that humid countries have higher seroprevalence than Sahelian ones. In a study conducted in Ghana in 2009, the 159 women aged 15-40 randomly investigated showed an overall seroprevalence of 92.5% [19]. This is the highest prevalence reported in West Africa till now. In Benin, the seroprevalence of 30% observed in 2014 by De Paschale et al [32] is very different from that observed there about two decades before by Rodier et al[33] which was 53.6%. Recent studies conducted in Nigeria by Deji-Agboola et al[28] in 2011 and Alayande et al[31] in 2013 give 32.6% and 27.75% as prevalence respectively that are statistically identical ($P < 0.05$) but very different from what was reported two decades ago by Onadeko et al, [34], which was 75.4%. This difference could be due to variations in geographic patterns within the same country. Indeed, Nigeria is a vast country with a large area covering wet and dry regions as well. The same observation is made in Senegal, where a recent study showed a prevalence of 34.5% [29] whereas 10 years before, the prevalence was around 60% [35]. Regarding Burkina Faso, the prevalence is stable according to Simporé et al [27] in 2006 and Bamba et al[30] who reported the respective prevalence of 25.3% and 31%, confirming that the lowest prevalence of toxoplasmosis are mostly recorded in dry regions [23,36].

In this study, it should be noted that none of the sera tested had positive IgM. This pattern is slightly different from the one previously published by Adoubryn et al[23] where 0.4% of IgG were associated with the presence of IgM in the southern Ivory Coast. The presence of IgG and absence of IgM (IgG^+ and IgM^-) in our study population for a first toxoplasma serology without any prior information in pregnant women, advocates a former probable immunity. In this context, 58.7% of women are not likely to transmit the infection to their children without any immunosuppression. Then, they would not require additional supervision during

pregnancy if a toxoplasmosis monitoring program was introduced [37]. However, in such case, we recommended a second serological test three weeks after the first serodiagnosis, to monitor changes in IgG level. Thus, a stable **titer** would definitely conclude to past infection while an increase in antibody **rate** after these three weeks should lead physicians to a more precise dating of the infection with the help of additional serological techniques to assess the risk to the fetus.

In case of known immunosuppression, the presence of IgG and absence of IgM even if it evokes an old probable infection, may require serological surveillance and must be interpreted in **the** light of the clinical and the degree of immunosuppression. Only the subsequent analysis of two sera at intervals of about three weeks in the same laboratory by the same technique allows a definitive conclusion [19]. **Forty-one point three percent** of the women in this study do not present toxoplasmosis-specific IgG or IgM. This demonstrates a lack of toxoplasma immunity with a risk of gestational seroconversion [37,38]. These women must be monitored regularly to detect any seroconversion (appearance of IgM and IgG). In case of confirmed seroconversion, the fetus is at risk of congenital infection, risks that varies depending on the gestational age at the time of maternal contamination. Parturient affected should get into a rigorous monitoring program (ultrasound and/or parasitological follow-up). It is noteworthy that in cases of antenatal seroconversion with evolution of serum signature and occurrence of clinical signs such as lymphadenopathy, fetal transmission may be probable [37,38]. In this study, the proportion of parturient that is still at risk of seroconversion during late pregnancy because not immunized (41.3%) is substantial. In order to reduce the risk of transmission of congenital toxoplasmosis in 159 non-immune women of our study, two preventive attitudes may be recommended: Firstly, regular control of serology if possible monthly until delivery and even nearest postpartum and secondly, application of hygienic dietary recommendations.

Cats were found as a risk factor for toxoplasmosis infection as already demonstrated in a study conducted in greater Accra region of Ghana [19]. This could be explained by direct contact with cat or by ingesting oocysts released by cats straying. The correlation found between age and seroprevalence just shows that the probability of being infected is increasing with the experienced time. No significant differences were found between factors such as type of housing, education, consumption of raw vegetables and trimester of pregnancy contrary to El-Ghany et al studies in Sharkia Province in Egypt [39] who found a positive correlation between the occurrence of toxoplasmosis and factors such as consumption of raw vegetables and contact with cats. In our study the lack of correlation between the consumption of raw vegetables and the occurrence of toxoplasmosis could be explained by improved washing of the vegetables. Regarding the type of housing, mutual housing as a common yard with multiple tenants generally leads to poor hygiene and subsequent health concerns. However, no significant correlation was found in our study between the type of housing and the occurrence of toxoplasmosis contrary to Mohammad et al[40], who found that housing conditions may influence the occurrence of toxoplasmosis.

4. CONCLUSION

The high prevalence rate observed in this study shows that toxoplasmosis is endemic in Daloa area. We recommend the establishment of serological surveillance program as soon as possible during pregnancies with counseling on the risk factors associated with toxoplasmosis combined with clear information on lifestyle, dietary rules and hygiene.

COMPETING INTEREST

The authors declare that they have no competing interests.

AUTHORS' CONTRIBUTIONS

This work was carried in collaboration among all authors.

M. Koffi designed the study and wrote the first draft of the manuscript which was approved by all authors.

I. Konaté and D.P. Sokouri wrote the protocol and managed the literature search.

J. C. Bosson and T. Konan participated in the collection of specimens and data.

B. Ahouty performed the statistical analysis.

All authors read and approved the final version of this manuscript.

ACKNOWLEDGEMENTS

We thank the Ivorian Ministry of Scientific Research, which Assigns research premium for lecturers and researchers. Our sincere thanks go to the team of laboratory technicians at maternal and child health care centre in Daloa and all women who agreed to participate in the study.

CONSENT AND ETHICAL APPROVAL

All authors hereby declared that this research was approved by national ethical committee of Ivory Coast and that an informed consent was obtained from the parturient.

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