



# Sero-Epidemiology of *Toxoplasma gondii* Infection in Pregnant Women from Dakar and Thiès Regions in Senegal

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## ABSTRACT

**Background:** Toxoplasmosis in pregnant women can result in many consequences such as abortion, mental retardation, seizures, blindness, and death. Preventive measure against *Toxoplasma gondii* infection during pregnancy is prenatal screening during prenatal consultation.

**Objective:** This study was assessing the seroprevalence of *Toxoplasma gondii* among pregnant women attending to Fann Teaching Hospital in Dakar and Thiès regional hospital in Senegal.

**Methods:** An observational study was conducted at the laboratory of parasitology at Fann Teaching Hospital in Dakar and the laboratory of Thiès regional hospital from August 2015 to May 2016. Pregnant woman attending the Fann Teaching Hospital and Thiès Regional Hospital for antenatal visit provided 10 ml of blood collected into a dry container. *T. gondii* immunoglobulin G (IgG) carriage was assessed using an indirect Enzyme-Linked Immunosorbent Assay (ELISA) method.

**Results:** One hundred thirty-two (132) pregnant women were included in this study (88 in Fann hospital and 44 in Thiès hospital). Overall, 56 samples were positive; *T. gondii* seroprevalence was evaluated at 42.4% (95% CI: 30-55.1). *T. gondii* seroprevalence was higher in 15-20 years group above (50%) compared to other age groups. Seroprevalence was more important in Thiès region (50%) compared to Dakar (28.4%). According to parity, the results of our study showed that the seroprevalence of IgG decreases with parity. The seropositivity rate was higher in the 2<sup>nd</sup> trimester 42.8%. No correlation was found between meat consumption, contact with cat, HIV positive status and other factors with *T. gondii* seroprevalence.

**Conclusion:** The prevalence of *T. gondii* infection in pregnant women is high compared to other studies conducted in Senegal. This study showed that *Toxoplasma gondii* infection is still a public health problem in Senegal. Epidemiological studies on risk factors could improve the management of this infection in order to avoid complication during pregnancy.

**Keywords:** Toxoplasmosis; Seroprevalence; Pregnancy; Senegal

## INTRODUCTION

Toxoplasmosis is a major public health concern worldwide. It's an anthroponosis caused by *Toxoplasma gondii*, an obligate intracellular protozoan that is cosmopolitan and ubiquitous [1,2]. The seroprevalence of Toxoplasmosis in humans varies differently among countries and geographic regions and is influenced

by behavioral factors such as personal hygiene, food handling practices, and culinary preferences within the same country [3,4].

It's estimated that about one-third of the world's population is infected with the parasite. It often remains unrecognized, as most patients do not exhibit symptoms [5].

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During pregnancy, the parasite can be transmitted across the placenta to the fetus resulting in congenital toxoplasmosis which can have grave consequences [6].

Toxoplasmosis in pregnant women is benign or clinically inapparent in more than 80% of cases but potentially serious for the fetus, which is why serology represents the basis for screening, diagnosis, and prevention of toxoplasmosis [1,4,7].

Toxoplasma serology should be repeated on a regular basis if the first test is negative, and seroconversion is considered an active infection. The risk of fetal transmission increases with gestational age, whereas the severity of fetal involvement decreases during pregnancy. Fetal sequelae are more important in the case of early infections [2,8,9].

Indeed, in case of seroconversion during the first trimester, the risk of congenital toxoplasmosis is 4% to 14%, resulting in severe damage. This risk rises to 70%-80% in the third trimester, but usually results in subclinical forms in the newborn [7].

Primary infection with toxoplasmosis in pregnant women can result in abortion and congenital toxoplasmosis.

Manifestations of congenital toxoplasmosis include severe congenital infection, intrauterine fetal death, microcephaly, hydrocephalus, mental retardation, psychomotor retardation, epilepsy, and brain calcifications [3,4,7-10].

In Africa, several studies assessing the seroprevalence of *T. gondii* in pregnant women were conducted. These studies have shown that the prevalence ranges from 18% to 78% among women [1].

In Senegal, the prevalence ranges from 22%-60%. The highest rates have been reported in humid regions where oocysts excreted into the environment are able to sporulate and retain their virulence over a long period of time, in contrast to the dry environment [1].

In Dakar, seroprevalence is 34.5% to 40.2% among women of childbearing age [6,11,12].

These data showed that toxoplasmosis is prevalent in Senegal, but it's considered as a public health priority. The serology is often used for the screening of the disease during antenatal care visit for early detection of toxoplasmosis and to prevent the consequences. In order to improve health care of population and to better guide health policy, it's become relevant to update the epidemiological data concerning the distribution of the diseases.

It was in this context the study was conducted with the objective of assessing the seroprevalence of toxoplasmosis in pregnant women attending to the laboratories of Parasitology the Fann University Hospital in Dakar and the Thiès Regional Hospital (CHR).

## MATERIALS AND METHODS

### Study design and participants

An observational study was conducted at the laboratory of parasitology at Fann Teaching Hospital in Dakar and the laboratory of Thiès regional hospital from August 2015 to May 2016.

Pregnant women attending the laboratory of parasitology at the

Fann Teaching Hospital (located in Dakar, capital city of Senegal) and Thiès regional hospital (located at 70 km from Dakar) for toxoplasmosis serological testing were enrolled in the study if they were up to 18 years. Pregnant women, who were previously screened for toxoplasmosis within the same study period, were excluded. Eligible participants were recruited using a consecutive sampling method. A code was given to each enrolled participants and data on pregnant women socio-demographic characteristics, history of pregnancy, and residency were collected from participant's medical records. Data obtained from participants medical records were assigned on a Case Report Form (CRF).

### Data collection methods

**Samples collection:** For each participant, blood venous sample (10 ml) was collected in a dry sterile tube without anticoagulant. The sample was then labelled and centrifuged at 1500 rpm for 10 min; the separated serum was transferred into Eppendorf tube and stored at -20°C until the day of analysis. Samples storage duration was on average 15 days for each specimen.

**Serological testing:** For the detection of anti *T. gondii* immunoglobulin G (IgG), indirect ELISA method was performed using the Platelia Toxo IgG kit as described elsewhere. Platelia Toxo IgG is a test for the detection and titration of IgG antibodies to *T. gondii* in human. In brief, *T. gondii* antigen is used for coating the microplate titration. A monoclonal antibody labelled with peroxidase which is specific for human gamma chains (anti-IgG) is used as the conjugate. Patient's samples, calibrators and controls were diluted on 1/21 and then distributed in the wells of the microplate and incubated at 37°C for 1 hr. This incubation period was followed by a washing period. In the second step of the essay, the conjugate which is a peroxidase labelled monoclonal antibody specific for human gamma chains was added to the microplate wells and incubated at 37°C for 1 hr. After washing, the presence of immune complexes such as *T. gondii* antigen, IgG antibodies to *T. gondii*, and anti-IgG conjugate was revealed by adding in each well an enzymatic development solution. After incubation at room temperature, the enzymatic reaction was stopped by the addition of 1N sulphuric acid solution. Optical density was obtained using a spectrophotometer (TescanTM) set at 450/620 nm. Optical densities were then converted onto UI/ml using a standard curve calibrated against the WHO international standard TOX-M [12].

### Statistical methods

**Sample size assumptions:** with 1200 pregnant women were sampled, the study was powered at 90% to detect 5% variation in *T. gondii* seroprevalence, assuming a seroprevalence of 35% based on previous studies with alpha at 0.05 (two sided). Data were entered in Excel TM software and analyzed using STATA 14 software.

For binary data, percentage was used to assess the frequency of each outcome with a 95% confidence interval. For continuous data, mean and standard deviation were used to describe normally distributed variables.

Samples were considered as positive if IgG concentration was equal or greater than 9 UI/ml. *T. gondii* seroprevalence was

calculated and expressed as proportion with 95% CI; proportions were compared using Chi square test (univariate analysis). Significance level of the different tests was 0.05, two sided.

## RESULTS

### General characteristics and medical history of pregnant women

A total of 132 pregnant women were enrolled during the study period with the mean age  $\pm$  Standard Deviation (SD) of  $28.3 \pm 5.4$  years. According to age categories, study population was mainly constituted by women aged 20-30 years with 57.6%. Two thirds (66.6%) of the pregnant women came from the parasitology laboratory of the CHNU of Fann.

Regarding the marital status, the study population was mainly represented by married pregnant women (92.4%). The majority (62.8%) of pregnant women had an income generating activity. According to the level of education, most pregnant women were educated (87.1%).

When assessing the risk factors of *T. gondii* infection the results of the study showed that most pregnant women declared consuming red meat (97.7%), having lived with cats in 44.7%. In 93.9%, they declared to respect the hygiene rules (Table 1).

**Table 1:** General characteristic of pregnant women (N=132).

Variable	Frequency (n)	Percentage (%)	95% CI
<b>Age group</b>			
<20 ans	6	4.5	1.9-9.6
[20-30[	76	57.6	48.7-66.1
[30-40[	45	34.1	26.1-42.84
$\geq 40$ ans	5	3.8	1.2-8.6
<b>Origin</b>			
Fann teaching hospital	88	66.7	57.9-74.6
Regional hospital Thiès	44	33.3	25.37-42.06]
<b>Marital status</b>			
Married	122	92.5	86.5 -96.3
Unmarried	10	7.5	3.6-13.9
<b>Occupation</b>			
With occupation	83	62.9	54.1-71.1
Housewife	49	37.1	27.5-49.1
<b>Education level</b>			
Educated	115	87.2	80.2-92.3
Illiterate	17	12.8	7.5-20.6
<b>Meat consumption</b>			
Yes	129	97.7	93.5-99.5
No	3	2.3	0.4-6.6
<b>Living with cat</b>			
Yes	59	44.7	36.1-53.6
No	73	55.3	43.3-69.5
<b>Regular hand washing before meal</b>			
Yes	124	93.9	88.4-97.4
No	8	6.1	2.6-11.9

According to parity, the study population was mainly composed of primiparous women (45.45%), pauciparous women (39.39%) and multiparous women (14.39%). Ten (10) pregnant women had a history of blood transfusion (7.6%). Four (04) pregnant women declared that they were infected with HIV (3.03%) (Table 2).

**Table 2:** Medical history of pregnant women (N=132).

Parameters	Frequency (n)	Percentage (%)	95% CI
<b>Parity</b>			
Primipares (G1)	60	45.5	36.7-54.4
Paucipares (G2-G3)	52	39.4	31-48.3
Multipares ( $\geq$ G4)	20	15.1	9.3-23.4
<b>Pregnancy trimester (months)</b>			
1 <sup>st</sup> Trimester (0-3)	77	58.4	49.4-66.8
2 <sup>nd</sup> Trimester (4-6)	42	31.8	23.9-40.5
3 <sup>rd</sup> Trimester (7-9)	13	9.8	5.4-16.3
<b>Blood transfusion</b>			
Yes	10	7.6	3.7-13.5
No	122	92.4	76.7-99.9
<b>HIV status</b>			
Positive	4	3.1	0.8-7.6
Negative	128	96.9	89.9-99.9

### Seroprevalence of toxoplasmosis in pregnant women

The overall seroprevalence of toxoplasmosis in our series was 42.42%, representing 56 pregnant women with anti-toxoplasma antibodies out of 132. Among the 56 pregnant women with anti-toxoplasma antibodies, 47 were carriers of anti-*Toxoplasma gondii* IgG (seroprevalence of anti-toxoplasma IgG of 35.61%) and 9 were carriers of detectable anti-*Toxoplasma gondii* IgM (sero-conversion rate of 6.82%) (Table 3).

**Table 3:** Seroprevalence of IgG and IgM anti-*Toxoplasma gondii* in pregnant women.

<b>IgG</b>			
	Frequency (n)	Percentage (%)	95% CI
Positive	47	35.6	27.5-44.4
Negative	85	64.4	55.6-72.5
Total	132	100	
<b>IgM</b>			
	Frequency (n)	Percentage (%)	95% CI
Positive	9	6.8	3.2-12.5
Negative	123	93.2	87.5-96.8
Total	132	100	

### Variations of IgG seroprevalence according to the general characteristics of pregnant women

According to age categories, IgG seroprevalence decreased with age, ranging from 50% in the youngest age category to 20% in the oldest women. However, the difference was not significant ( $p=0.78$ ). Regarding the recruitment site, IgG seroprevalence was higher in Thiès (50%) compared to Dakar where seroprevalence was 28.41% with a statistically significant difference ( $p=0.015$ ). Married pregnant women had a low IgG seropositivity rate

(33.6%) compared to unmarried women (60%) ( $p=0.09$ ). By occupation, seroprevalence was higher among women with no income (38.8%) compared to those with income, whose seroprevalence was 33.7% ( $p=0.55$ ). According to education level, educated pregnant women had a higher IgG seropositivity rate (37.4%) than uneducated women (23.5%) ( $p=0.26$ ). The highest IgG seroprevalence was found in women who reported not eating red meat (66.7%) compared to those who did (34.8%) ( $p=0.25$ ). In women who lived with cats, IgG seropositivity was higher (42.4%) compared to those who did not live with cats (30.2%). IgG seroprevalence was higher in women who did not follow hand hygiene (50%) compared to 34.7% in those who followed hygiene measures with no statistically significant difference ( $p=0.38$ ) (Table 4).

**Table 4:** Seroprevalence of IgG antibodies according to general characteristic of pregnant women (N=47).

IgG Seroprevalence				
Variables	Frequency	Percentage (%)	95% CI	P-value
<b>Age group</b>				
[15-20[	3	50	10.3-99.9	
[20-30[	27	35.5	23.4-51.7	
[30-40[	16	35.5	20.3-57.7	
[40-45[	1	20	0.5-99.9	0.78
<b>Origin</b>				
Fann teaching hospital	25	28.4	18.4-41.9	
Regional hospital Thiès	22	50	31.3-75.5	0.015
<b>Marital status</b>				
Married	41	33.6	24.1-45.6	
Unmarried	6	60	22-99.9	0.09
<b>Occupation</b>				
With occupation	28	33.7	22.4-48.7	
Housewife	19	38.8	23.3-60.5	0.55
<b>Education level</b>				
Educated	4	23.5	6.4-60	
Illiterate	43	37.4	27-50.4	0.26
<b>Meat consumption</b>				
Yes	2	66.7	Aug-24	0.25
No	45	34.8	25.4-46.7	
<b>Living with cat</b>				
Yes	22	30.14	18.8-45.6	0.14
No	25	42.37	27.4-62.5	
<b>Regular hand washing before meal</b>				
Yes	4	50	13.6-12.8	0.38
No	43	34.7	25-46.7	

### Variation of seroprevalence according to medical history of pregnant women

According to the parity, the results of our study showed that IgG seroprevalence decreased with parity, from 43.3% in primiparous women to 21.1% in multiparous women. The difference was significant ( $p=0.25$ ).

Depending on the trimester of pregnancy, the overall IgG

seropositivity rate was 35.6%. This rate was higher in the 2<sup>nd</sup> trimester 42.8%. On the other hand, in the 1<sup>st</sup> and 3<sup>rd</sup> trimester, the seroprevalence was 32.5% and 30.7% respectively ( $p=0.49$ ).

The rate of seropositivity was higher among women with no history of blood transfusion (36.1%) compared to those with a history of blood transfusion (30%) ( $p=0.7$ ). Among the four HIV positive pregnant women, three had positive toxoplasma serology (75% seroprevalence) (Table 5).

**Table 5:** Seroprevalence of IgG antibodies according to medical history of pregnant women (N=47).

IgG seroprevalence				
Variables	Frequency	Percentage (%)	95% CI	P-value
<b>Parity</b>				
Primipares (G1)	26	43.33	28.3-63.49	0.25
Paucipares (G2-G3)	17	32.69	19-52.34	
Multipares ( $\geq$ G4)	4	21.05	5.73-53.9	
<b>Pregnancy trimester (months)</b>				
1 <sup>st</sup> Trimester (0-3)	25	32.47	21-47.92	0.49
2 <sup>nd</sup> Trimester (4-6)	18	42.86	25.39-67.73	
3 <sup>rd</sup> Trimester (7-9)	4	30.77	8.3-78.78	
<b>Blood transfusion</b>				
Yes	3	30	6.18-87.67	
No	44	36.07	26.2-48.41	0.7
<b>HIV status</b>				
Positive	3	75	15.46-219	
Negative	29	37.66	25.22-54.08	0.15

## DISCUSSION

The overall seroprevalence of toxoplasmosis in our study was 42.42% (56/132) with anti-toxoplasma antibodies. Among the 56 pregnant women with anti-toxoplasma antibodies, 47 were carriers of anti-*Toxoplasma gondii* IgG (seroprevalence of 35.6%). Similar results were found by other authors. In Senegal, Tine et al. have showed a seroprevalence of 35.4% in pregnant attending to Fann hospital [12]. These results are in line with what found by Ndiaye et al. in 2011 (34.5%) [11]. 40.2% seroprevalence was found by Faye et al. Dakar in 353 women under reproductive age [13]. Bamba et al. in Burkina Faso had found a seroprevalence of 31% (95/306) in pregnant women, defined by the presence of *T. gondii* specific IgG [8]. When assessing the seroprevalence and risk factors of *Toxoplasma gondii* infection in pregnant women, Bamba et al. have found 31.1% seroprevalence [14].

In our study, according to age categories, IgG seroprevalence decreased with age, ranging from 50% in the youngest age category to 20% in the oldest women. In fact, the seropositivity rate obtained as early as 15 years of age proves that most of the

contamination takes place during childhood.

According to the level of education, educated pregnant women had a higher IgG seropositivity rate (37.39%) than uneducated women (23.53%) ( $p=0.26$ ). This result is similar to that of Mwambel et al. in Tanzania who found a seroprevalence of 66.67% among educated women. Pangui et al. Dakar found a seroprevalence that varied between 22%-60% [1,10].

Depending on the trimester of pregnancy, the overall IgG seroprevalence rate is 35.61%. This rate was higher in the 2<sup>nd</sup> trimester at 42.8%. On the other hand, in the 1<sup>st</sup> and 3<sup>rd</sup> trimester, the seroprevalence was 32.47% and 30.7% respectively ( $p=0.49$ ). This result is consistent with those of Zemene et al. in 2012 in Ethiopia who found a high seroprevalence of 82.7% [3]. Mwambel et al. in Tanzania had found a seroprevalence of 33.3% among pregnant women in the 2<sup>nd</sup> trimester. This is consistent with the distribution of the infection [9].

Indeed, the risk of fetal transmission increases with gestational age while the severity of fetal involvement decreases during pregnancy. Fetal sequelae are more important in the case of early infections [2,8,9]. Indeed, in case of seroconversion during the first trimester, the risk of congenital toxoplasmosis is 4% to 14%, resulting in severe damage. This risk rises to 70%-80% in the third trimester, but usually results in subclinical forms in the newborn [7].

Primary infection with toxoplasmosis in pregnant women can result in abortion and congenital toxoplasmosis. Manifestations of congenital toxoplasmosis include severe congenital infection, intrauterine fetal death, microcephaly, hydrocephalus, mental retardation, psychomotor retardation, epilepsy, and brain calcifications [3,4,7-10].

The highest IgG seroprevalence was found in women who did not consume red meat (66.67%) compared to those who consumed red meat (34.8%). However, the difference was not significant ( $p=0.25$ ). Indeed, meats are consumed well cooked in Senegal [6]. HIV status seems to be explained by other factors.

Among women who live with cats, IgG seropositivity was higher (42.4%) compared to those who do not live with cats (30.1%). Our results are similar to those of Gebremedhin et al. in 2013 in Ethiopia who found a strong association between seroprevalence of toxoplasmosis in pregnant women cohabiting with cats (84.9%,  $p<0.05$ ) [15]. The association between the notion of contact with the cat, dirty hands and seroprevalence of toxoplasmosis in pregnant women has been established [16]. In our study, the seroprevalence of IgG was higher in women who did not follow hand hygiene (50%) compared to 34.7% in those who did follow hygiene measures without any statistically significant difference ( $p=0.38$ ). This shows the role of cats in environmental contamination.

Concerning the *T. gondii* and HIV co-infection, out of the four persons with a positive HIV serology, 3 had a positive toxoplasma serology (seroprevalence of 75%). Our result is similar to that of Gebremedhin et al. in Ethiopia who found a high seroprevalence of *T. gondii* in pregnant women infected with HIV at 88.4% [14,15].

## CONCLUSION

The prevalence of *T. gondii* infection in pregnant women is high compared to other studies conducted in Senegal. This study showed that *Toxoplasma gondii* infection is still a public health problem in Senegal. Risk factors were not clearly identified in this study. Epidemiological studies on risk factors could improve the management of this infection in order to avoid complication during pregnancy.

## DECLARATIONS

### Ethical considerations

Inform consent was required prior the participation to the study. Pregnant women who didn't accept to sign inform consent form were not included in the study. This study was conducted according to the Declaration of Helsinki and existing national legal and regulatory requirements. To respect the confidentiality, an identification code was given to each participant. The study was approved by the Ethic Committee of University Cheikh Anta Diop of Dakar (408/2019/CER/UCAD).

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## CONFLICT OF INTEREST

The authors declare that they have no competing interest

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## AUTHOR CONTRIBUTIONS

KS conceived and designed the study, KS and HAS supervised the data collection, HAS realized serological assessment, KS analyzed the data and wrote the first draft of the manuscript. All authors read and approved the final manuscript.

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