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Toxoplasmosis among Pregnant Women at the Integrated Health Center in Mahajanga (Madagascar): A Preliminary Study

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Abstract - Serological screening for toxoplasmosis in pregnant women is not systematic in Madagascar. However, the urban lifestyle and the often precarious lifestyle of the population are a favorable factor in the onset of the disease. The objectives of this study were to determine the seroprevalence of toxoplasmosis in pregnant women who came for an initial prenatal visit at the Integrated Health Center (IHC) Mahabibo, and to determine the socio-demographic factors that can influence the serological status of these patients in order to propose suggestions for improving the management of toxoplasmosis in pregnant women in Madagascar.

We conducted a prospective, descriptive and analytical study of pregnant women in first prenatal visit at the IHC Mahabibo from May to July 2018. A questionnaire on risk factors and knowledge of the pathology was conducted to participants. They have benefited a blood sample for free serological screening of toxoplasmosis using the Rapid Lab® EIA technique. A second sample was taken after one month for pregnant women who came to 2nd prenatal visit. The biological study was carried out in the laboratory of the PZaGa Mahajanga University Hospital Center.

A total of 96 serologies were performed, including 58 screenings and 38 follow-ups. The seroprevalence of toxoplasmosis was 36.2%. We found 23.7% seroconversion suggestive of recent infection. The seropositivity of toxoplasmosis was 85.7% among pregnant women who consumed raw vegetables and 71.4% among those who ate undercooked meat (p>0,05).

Awareness of the risks of contamination, systematic serological surveillance and hygiene measures should be provided during prenatal visits.

Keywords - EIA, pregnant women, serology, toxoplasmosis.

I. BACKGROUND

Toxoplasmosis is a zooanthroponosis due to *Toxoplasma gondii*. It is a ubiquitous parasitosis affecting less of 10% to 90% of the world's population [1]. The strategy of systematic screening of pregnant women has been successfully developed in some countries to enable early treatment of the mother-fetus couple and care of the child from birth [2]. In Madagascar, there is no systematic

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screening program for toxoplasmosis in pregnant women and access to toxoplasmosis serology is still limited. Peripheral health facilities, which represent a large part of the country's health structures, cannot carry out this analysis.

Severe forms of toxoplasmosis are observed in immunocompromised women and pregnant women with foetopathies fatal to the fetus. The parasite can infect all cell types, especially those with a low immune response (brain, heart, skeletal muscle) where it remains throughout the host's life. It can be transmitted to the fetus by transplacental route [3]. In humans, horizontal contamination is caused either by ingestion of oocysts on soiled food, soiled cooking equipment or drinking water, or by cysts in raw or undercooked meat [4]. Blood transplantation or transfusion from infected donors, accidental dermal-mucosal inoculation is also routes of contamination [5]. Hence the importance of early screening and regular follow-up of pregnant women to detect early seroconversion and thus adapt management according to the risk to the fetus. The objectives of this study were to determine the seroprevalence of toxoplasmosis in pregnant women and the associated risk factors to prevent the occurrence of congenital toxoplasmosis.

II. METHODOLOGY

A prospective, descriptive and analytical cross-sectional study was conducted from May to July 2018. The biological study was carried out in the medical laboratory of the University Hospital Center Professor Zafisaona Gabriel, and the questionnaire sessions and blood samples from pregnant women were carried out in the Mahabibo IHC. This study involved pregnant women who met the following criteria: be in the first prenatal visit and have given their informed consent to the study. These women were screened free of charge during prenatal consultations. The pregnant women included completed a questionnaire and collected 5mL of venous blood from a dry vacutainer tube for toxoplasmosis serology. The samples were then transported to the laboratory. After centrifugation at 2500 rpm for 15 minutes, the serums were aliquoted. Serology was performed with the EIA technique (Rapid Labs Limited, United Kingdom; Lot No. 1712218, expiry date 22/07/2019 for IgG and Lot No. 1804001 with expiry date 15/09/2019 for IgM).

To identify possible seroconversion, a 2nd sample was taken after 4 weeks during the 2nd prenatal visit.

In the questionnaire, the survey covered occupation, history of spontaneous abortion, keeping domestic cat, culinary habits, hygiene habits and knowledge about toxoplasmosis.

Data analysis

Data were entered on Epi info 7.0 software, and analyzed on R software French version 3.5.2 (developed by Center for Disease Control and Prevention, Atlanta, USA).

III. RESULTS

Fifty-eight pregnant women were included with whom 38 have performed a 2nd prenatal visit and received serological follow-up for toxoplasmosis. A total of 96 serologies were performed during the study period.

Table I shows the characteristics of our study population with the distribution of the seroprevalence of toxoplasmosis. The age of pregnant women ranged from 15 to 42 years with an average of 25.8 ± 6.9 years and a median at 25 years old. The gestational age varied between 8 SA and 37 SA 4 days with an average of 21.3 ± 6.7 SA. The gestity varied from 1 to 8 with an average of 2.5 ± 1.5. No pregnant woman has known her serological status with regard to toxoplasmosis because none of them has performed toxoplasmosis serology before our study. No notion of fetal malformation or comitativeness in the baby was noted in the gynaecological-obstetrical history of these pregnant women. None of them had a history of blood transfusion. Only one pregnant woman (1.7%) has heard of toxoplasmosis but she has not had enough knowledge about the toxoplasmosis transmission mode and its consequences in the fetus. Her serology was positive for IgG. The majority (98.3%) have never heard of this pathology. No patients had cervical or occipital adenopathies.

At the first serology, 21 pregnant women (36.2%) had only anti-toxoplasmic IgG antibodies (IgG+,IgM-), and 37 (63.8%) were seronegative (IgG-, IgM-) or non-immune. We found 23.7% of cases with a significant increase in optical density of anti-toxoplasmic IgG (above 2 times the confidence interval at 95%, 0,84 after the calculation) among pregnant women who did a serological follow up. No cases of anti-toxoplasmic IgM+, either during screening or serological follow-up, were found.

Table I : Sociodemographic characteristics of pregnant women and seroprevalence of toxoplasmosis

Variable			IgG +	IgG -	*p*
		N=58 (100%)	n=21 (36,2%)	n=37 (63,8%)	
Age (years old)	< 20	14 (24,1)	4 (28,6)	10 (71,4)	*0.89*
	[20-30[	27 (46,6)	10 (37,0)	17 (63,0)	
	≥ 30	17 (29,3)	7 (41,2)	10 (58,8)	
Gestational age (amenorrhea week)	<16	17 (29,3)	7 (41,2)	10 (58,8)	*0,74*
	[16-28[	35 (60,4)	11 (31,4)	24 (68,6)	
	≥28	6 (10,3)	3 (50,0)	3 (50,0)	
Gestity	Primigeste	16 (27,6)	7 (43,8)	9 (56,2)	*0,54*
	Multigeste	42 (72,4)	14 (33,3)	28 (66,7)	
Level of study	Illiterate	7 (12,0)	0 (0,0)	7 (100,0)	*0,21*
	Primary school	6 (10,4)	3 (50,0)	3 (50,0)	
	Secondary school	30 (51,7)	13 (43,3)	17 (56,7)	
	High school	11 (19,0)	4 (36,4)	7 (63,6)	
	University	4 (6,9)	1 (25,0)	3 (75,0)	
Occupation	Farmer	2 (3,4)	0 (0,0)	2 (100,0)	*0,42*
	Housewife	35 (60,4)	12 (34,3)	23 (65,7)	
	Professionnal	17 (29,3)	6 (35,3)	11 (64,7)	
	Employee	4 (6,9)	3 (75,0)	1 (25,0)	

Figure 1 and figure 2 show factors associated with seropositivity of toxoplasmosis in pregnant women. No significant association between toxoplasmosis and the various factors studied was found too (p>0,05).

IV. DISCUSSION

The prevalence of toxoplasmosis among pregnant women in our study, which is 36.2%, was lower than that found among pregnant women in Antananarivo in 1992 (83.5%)[6] and in Mahajanga in 2016 (61%)[7]. This prevalence is also low compared to that of the general population between 1990 and 1995 (52.1%) [8] and a female population in 2000 (41.3%) [9]. The differences in results can be explained by the different recruitment patterns of patients. Compared to other countries, our result is close to some African countries (Benin, Senegal, Burkina Faso) whose prevalence has ranged from 31 to 36%[10-13] and lower compared to the prevalence found in Morocco (50.6% in 2007)[14] in Central Africa in 2012 (50.9%)[15], Ghana (92.5% in 2010)[16] and Gabon (80.8% in 2009)[17]. Lower prevalences have been reported among pregnant women in China (15.2% in 2015) [18], Italy (28.3% in 2010) [19] and Denmark (27.4% in 1993) [20]. The prevalence of toxoplasmosis can vary from one country to another and even sometimes within the same country, especially when moving from an urban to a rural environment. Geo-climatic conditions, recognizing that heat and humidity are factors that favour the conservation of oocysts in the soil and thus contribute to maintaining a high prevalence, but also other risk factors related to lifestyles and diet, including hygiene and the way food is consumed or cooked, have been mentioned to explain these differences [4,21,22]. Some authors reported that contamination was more related to poor hygiene in poor countries, while in rich countries it was more related to eating habits [21,22].

We found 23.7% of cases with a significant increase in optical density of anti-toxoplasmic IgG among pregnant women who did a serological follow up, suspecting seroconversion and therefore recent infection, which implies the presence of a risk of fetal transmission of the parasite.

In our study, the seropositivity of toxoplasmosis was higher in women without a history of spontaneous abortion

(Figure 1), but the difference was not significant (p>0,05). Toxoplasmic seroconversion occurring during the periconceptional period could be responsible for 41.6 to 66% of miscarriages [23,24]. But not all miscarriages are due to toxoplasmosis, especially when they are recurrent, other etiologies such as genetic causes, uterine anomalies, thrombophilia, autoimmune diseases, endocrine diseases, psychological factors should be investigated [25].

We found that there was 42,9% of the seropositive pregnant women who used to use soap for hand washing (Figure 2). Therefore, simple hand washing with soap could not be sufficient to prevent toxoplasm infestation, which implies the presence of another source of contamination for these populations. Consumption of rawness was noted in 85.7% of pregnant women seropositive of toxoplasmosis (Figure 1), and all these pregnant women have not used to wash fruits and raw vegetables with soap (Figure 2). Indeed, rawness can be a major source of toxoplasm contamination [4], because fruits and raw vegetables can be contaminated by T. gondii oocysts in different places and in different ways, either by exposure to dust, or via dirty hands. A simple washing of food with water is not enough to get rid of these oocysts. Some authors have reported that the consumption of raw vegetables was significantly related to the seroprevalence of toxoplasmosis [10]. This hypothesis was not confirmed in our study.

Our results show that consumption of undercooked meat was also found in 71.4% of pregnant women seropositive in toxoplasmosis (Figure 1). Several epidemiological studies have agreed on the existence of a risk of toxoplasmosis associated with the consumption of undercooked meat [21]. Meat and poultry from animals raised outdoors may contain *T. gondii* cysts. These cysts are quite resistant, only a freezing at -18°C or proper cooking throughout the thickness of the meat at 72°C have allowed the cysts to be destroyed [21,26]. Some subjects are used to eating malted meat (skewers) accompanied by uncovered raw vegetables at the seaside, which leads to a high risk of contamination by both cysts and oocysts of *T. gondii* [27].

In our case, the seropositivity of toxoplasmosis was too higher among pregnant women who had direct contact with soil at through the household (66,7%) (Figure 1), but not statistically significant (p>0.05). The soil is directly involved in the transmission of toxoplasmosis. The habit of felines to bury their faeces allows contamination of the first ten centimetres of the surface of the soil and prevents desiccation of oocysts. Oocysts remain infectious for 30 to 410 days depending on climatic conditions [28,29]. This makes contact with the soil a risk factor in the acquisition of toxoplasmosis if hand hygiene is insufficient. Contact with soil was identified as the cause of 17% of recent seroconversions or infections in a multi-center study in Europe in 2000 [30]. Other studies have revealed the causal link between contact with soil and toxoplasmic infection (Iran [31] and Egypt [32]). Hence the importance of hygiene in the prevention of toxoplasmosis.

We found that toxoplasmosis was higher in pregnant women who did not have direct contact with domestic cat (61.9%) compared to those who have keeped cat at home (38.1%), but the difference was not significant (p=0.29) (Figure 1). While contact with cats was a risk factor for toxoplasmosis contraction by some authors [4,33] this was not the case in our study. Women were as infected whether or not they were in direct contact with cats, which would have meant the presence of other sources of contamination, such as eating habits and poor hygiene conditions for those who did not have contact with cat. For those who have been in direct contact with the final host, they may also not be infected if they have complied with strict hygiene rules, especially regarding the handling of their litter [34]. Daily change and disinfection of the litter are essential before the oocysts, which would eventually be rejected, are sporulated. Oocysts only become infestant after 2 to 5 days [26]. In addition, toxoplasm contamination of a domestic cat could be prevented if its diet is well controlled from birth. It should not be fed raw meat or raw milk but only with dry or cooked commercial food (croquettes, cans) [35,36]. However, we could not prevent them from hunting rats or mice that are everywhere. Hence, the best prevention would be the strict adherence to hygiene rules.

The results of the survey revealed that toxoplasmosis is very little known by the study population, 98,3% of subjects have no knowledge of the toxoplasmosis's modes of transmission. Our data were comparable to that found in a rural town in Benin where 99,5% of women have no knowledge about this subject [10]. Indeed, knowledge of toxoplasmosis, as well as the sources of contamination, is becoming an imperative for the general public. These results are related to a lack of communication around the pathology because of its asymptomatic nature. In the future, a study on cat parasitemia would be beneficial to assess the risk of exposure of households.

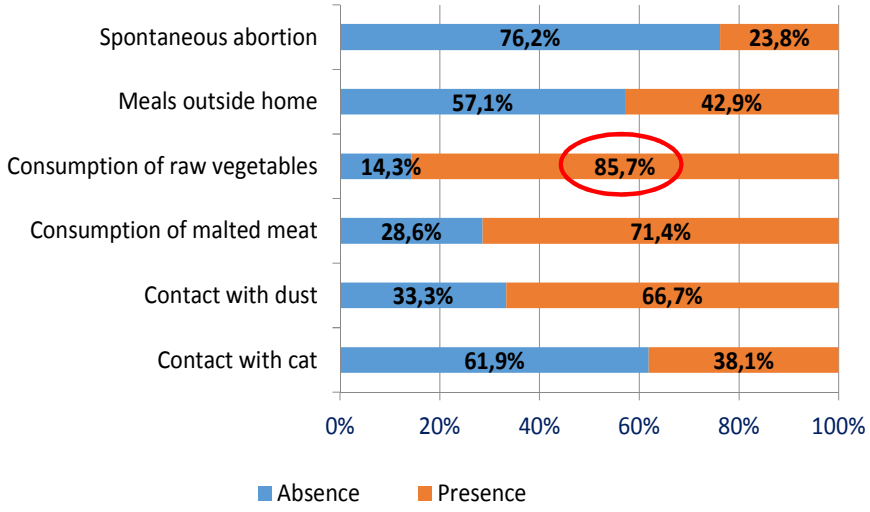

Figure 1 : Distribution of pregnant women seropositive with toxoplasmosis according to the exposure to risk factors and history of spontaneous abortion (n=21)

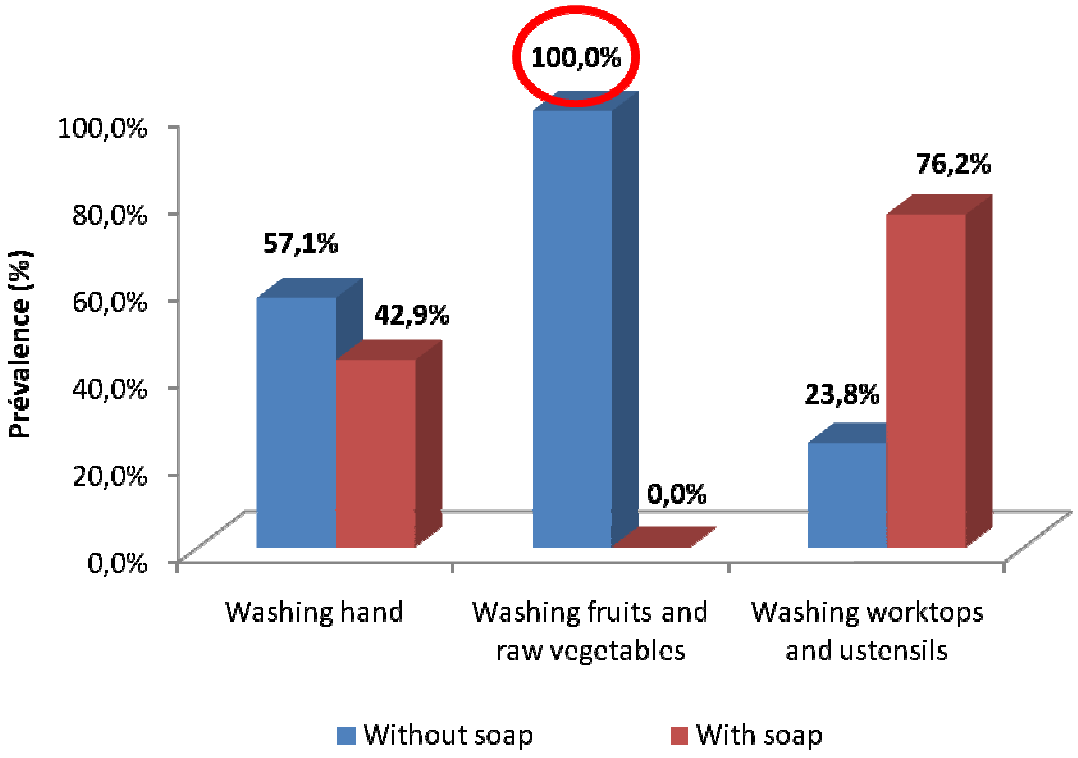

Figure 2: Distribution of pregnant women seropositive with toxoplasmosis according to their hygiene habits (n=21)

V. CONCLUSION

At the end of our study, we could say that populations must therefore be better informed about the disease (the parasite cycle and modes of transmission) in order to understand and more easily adopt hygiene-dietary recommendations that reduce exposure to the parasite. The results of this work confirmed that infestation by parasitoses is real for a young and very active population. Awareness of the risks of contamination, systematic serological surveillance and hygiene measures should be proposed during prenatal consultations at both the peripheral health centres and the University Hospital, in order to prevent toxoplasmic seroconversions in pregnant women and to protect the child. Our work was only a preliminary study that could serve as the basis for a larger study with a more representative sample number of Mahajanga population in perspective.

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